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| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/36631597  Title: The cytoplasmic localization of ADNP through 14-3-3 promotes sex-dependent neuronal morphogenesis, cortical connectivity, and calcium signaling.  Defective neuritogenesis is a contributing pathogenic mechanism underlying a variety of neurodevelopmental disorders. Single gene mutations in activity-dependent neuroprotective protein (ADNP) are the most frequent among autism spectrum disorders (ASDs) leading to the ADNP syndrome. Previous studies showed that during neuritogenesis, Adnp localizes to the cytoplasm/neurites, and Adnp knockdown inhibits neuritogenesis in culture. Here, we hypothesized that Adnp is localized in the cytoplasm during neurite formation and that this process is mediated by 14-3-3. Indeed, applying the 14-3-3 inhibitor, difopein, blocked Adnp cytoplasmic localization. Furthermore, co-immunoprecipitations showed that Adnp bound 14-3-3 proteins and proteomic analysis identified several potential phosphorylation-dependent Adnp/14-3-3 binding sites. We further discovered that knockdown of Adnp using in utero electroporation of mouse layer 2/3 pyramidal neurons in the somatosensory cortex led to previously unreported changes in neurite formation beginning at P0. Defects were sustained throughout development, the most notable included increased basal dendrite number and axon length. Paralleling the observed morphological aberrations, ex vivo calcium imaging revealed that Adnp deficient neurons had greater and more frequent spontaneous calcium influx in female mice. GRAPHIC, a novel synaptic tracing technology substantiated this finding, revealing increased interhemispheric connectivity between female Adnp deficient layer 2/3 pyramidal neurons. We conclude that Adnp is localized to the cytoplasm by 14-3-3 proteins, where it regulates neurite formation, maturation, and functional cortical connectivity significantly building on our current understanding of Adnp function and the etiology of ADNP syndrome. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/35775424  Title: A convergent mechanism of high risk factors ADNP and POGZ in neurodevelopmental disorders.  ADNP and POGZ are two top-ranking risk factors for autism spectrum disorder and intellectual disability, but how they are linked to these neurodevelopmental disorders is largely unknown. Both ADNP and POGZ are chromatin regulators, which could profoundly affect gene transcription and cellular function in the brain. Using post-mortem tissue from patients with autism spectrum disorder, we found diminished expression of ADNP and POGZ in the prefrontal cortex, a region highly implicated in neurodevelopmental disorders. To understand the functional role of these neurodevelopmental disorder risk factors, we used viral-based gene transfer to investigate how Adnp or Pogz deficiency in mouse prefrontal cortex affects behavioural, transcriptomic and synaptic function. Mice with prefrontal cortex deficiency of Adnp or Pogz exhibited specific impairment of cognitive task performance. RNA-sequencing revealed that Adnp or Pogz deficiency induced prominent upregulation of overlapping genes enriched in neuroinflammation, similar to the elevation of pro-inflammatory genes in humans with neurodevelopmental disorders. Concomitantly, Adnp or Pogz deficiency led to the significant increase of pro-phagocytic microglial activation in prefrontal cortex, as well as the significant decrease of glutamatergic transmission and postsynaptic protein expression. These findings have uncovered the convergent functions of two top risk factors for autism spectrum disorder and intellectual disability in prefrontal cortex, providing a mechanism linking chromatin, transcriptional and synaptic dysregulation to cognitive deficits associated with neurodevelopmental disorders. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/35538192  Title: SH3- and actin-binding domains connect ADNP and SHANK3, revealing a fundamental shared mechanism underlying autism.  De novo heterozygous mutations in activity-dependent neuroprotective protein (ADNP) cause autistic ADNP syndrome. ADNP mutations impair microtubule (MT) function, essential for synaptic activity. The ADNP MT-associating fragment NAPVSIPQ (called NAP) contains an MT end-binding protein interacting domain, SxIP (mimicking the active-peptide, SKIP). We hypothesized that not all ADNP mutations are similarly deleterious and that the NAPV portion of NAPVSIPQ is biologically active. Using the eukaryotic linear motif (ELM) resource, we identified a Src homology 3 (SH3) domain-ligand association site in NAP responsible for controlling signaling pathways regulating the cytoskeleton, namely NAPVSIP. Altogether, we mapped multiple SH3-binding sites in ADNP. Comparisons of the effects of ADNP mutations p.Glu830synfs\*83, p.Lys408Valfs\*31, p.Ser404\* on MT dynamics and Tau interactions (live-cell fluorescence-microscopy) suggested spared toxic function in p.Lys408Valfs\*31, with a regained SH3-binding motif due to the frameshift insertion. Site-directed-mutagenesis, abolishing the p.Lys408Valfs\*31 SH3-binding motif, produced MT toxicity. NAP normalized MT activities in the face of all ADNP mutations, although, SKIP, missing the SH3-binding motif, showed reduced efficacy in terms of MT-Tau interactions, as compared with NAP. Lastly, SH3 and multiple ankyrin repeat domains protein 3 (SHANK3), a major autism gene product, interact with the cytoskeleton through an actin-binding motif to modify behavior. Similarly, ELM analysis identified an actin-binding site on ADNP, suggesting direct SH3 and indirect SHANK3/ADNP associations. Actin co-immunoprecipitations from mouse brain extracts showed NAP-mediated normalization of Shank3-Adnp-actin interactions. Furthermore, NAP treatment ameliorated aberrant behavior in mice homozygous for the Shank3 ASD-linked InsG3680 mutation, revealing a fundamental shared mechanism between ADNP and SHANK3. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/34865853  Title: Novel ADNP Syndrome Mice Reveal Dramatic Sex-Specific Peripheral Gene Expression With Brain Synaptic and Tau Pathologies.  ADNP is essential for embryonic development. As such, de novo ADNP mutations lead to an intractable autism/intellectual disability syndrome requiring investigation. Mimicking humans, CRISPR (clustered regularly interspaced short palindromic repeats)-Cas9 editing produced mice carrying heterozygous Adnp p.Tyr718∗ (Tyr), a paralog of the most common ADNP syndrome mutation. Phenotypic rescue was validated by treatment with the microtubule/autophagy-protective ADNP fragment NAPVSIPQ (NAP). RNA sequencing of spleens, representing a peripheral biomarker source, revealed Tyr-specific sex differences (e.g., cell cycle), accentuated in females (with significant effects on antigen processing and cellular senescence) and corrected by NAP. Differentially expressed, NAP-correctable transcripts, including the autophagy and microbiome resilience-linked FOXO3, were also deregulated in human patient-derived ADNP-mutated lymphoblastoid cells. There were also Tyr sex-specific microbiota signatures. Phenotypically, Tyr mice, similar to patients with ADNP syndrome, exhibited delayed development coupled with sex-dependent gait defects. Speech acquisition delays paralleled sex-specific mouse syntax abnormalities. Anatomically, dendritic spine densities/morphologies were decreased with NAP amelioration. These findings were replicated in the Adnp+/- mouse, including Foxo3 deregulation, required for dendritic spine formation. Grooming duration and nociception threshold (autistic traits) were significantly affected only in males. Early-onset tauopathy was accentuated in males (hippocampus and visual cortex), mimicking humans, and was paralleled by impaired visual evoked potentials and correction by acute NAP treatment. Tyr mice model ADNP syndrome pathology. The newly discovered ADNP/NAP target FOXO3 controls the autophagy initiator LC3 (microtubule-associated protein 1 light chain 3), with known ADNP binding to LC3 augmented by NAP, protecting against tauopathy. NAP amelioration attests to specificity, with potential for drug development targeting accessible biomarkers. |
| ADNP, ARID1B, TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34858139  Title: Wnt/β-Catenin-Dependent Transcription in Autism Spectrum Disorders.  Autism spectrum disorders (ASD) is a heterogeneous group of neurodevelopmental disorders characterized by synaptic dysfunction and defects in dendritic spine morphology. In the past decade, an extensive list of genes associated with ASD has been identified by genome-wide sequencing initiatives. Several of these genes functionally converge in the regulation of the Wnt/β-catenin signaling pathway, a conserved cascade essential for stem cell pluripotency and cell fate decisions during development. Here, we review current information regarding the transcriptional program of Wnt/β-catenin signaling in ASD. First, we discuss that Wnt/β-catenin gain and loss of function studies recapitulate brain developmental abnormalities associated with ASD. Second, transcriptomic approaches using patient-derived induced pluripotent stem cells (iPSC) cells, featuring mutations in high confidence ASD genes, reveal a significant dysregulation in the expression of Wnt signaling components. Finally, we focus on the activity of chromatin-remodeling proteins and transcription factors considered high confidence ASD genes, including CHD8, ARID1B, ADNP, and TBR1, that regulate Wnt/β-catenin-dependent transcriptional activity in multiple cell types, including pyramidal neurons, interneurons and oligodendrocytes, cells which are becoming increasingly relevant in the study of ASD. We conclude that the level of Wnt/β-catenin signaling activation could explain the high phenotypical heterogeneity of ASD and be instrumental in the development of new diagnostics tools and therapies. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/33453943  Title: Activity-dependent neuroprotective protein (ADNP)-end-binding protein (EB) interactions regulate microtubule dynamics toward protection against tauopathy.  The 1102-amino-acid activity-dependent neuroprotective protein (ADNP) was originally discovered by expression cloning through the immunological identification of its 8-amino-acid sequence NAPVSIPQ (NAP), constituting the smallest active neuroprotective fragment of the protein. ADNP expression is essential for brain formation and cognitive function and is dysregulated in a variety of neurodegenerative diseases (Alzheimer's disease, Parkinson's disease, and schizophrenia). ADNP has been found to be mutated in autism, with an estimated prevalence of 0.17% (together, these autism cases now constitute ADNP syndrome cases) and our recent results showed somatic mutations in ADNP in Alzheimer's disease brains correlating with tauopathy. Furthermore, Adnp haploinsufficiency in mice causes an age-dependent reduction in cognitive functions coupled with tauopathy-like features such as an increased formation of tangle-like structures, defective axonal transport, and Tau hyperphosphorylation. ADNP and its derived peptides, NAP and SKIP, directly interact with end-binding proteins (EBs), which decorate plus-tips of the growing axonal cytoskeleton-microtubules (MTs). Functionally, NAP and SKIP are neuroprotective and stimulate axonal transport. Clinical trials have suggested the potential efficacy of NAP (davunetide, CP201) for improving cognitive performance/functional activities of daily living in amnestic mild cognitive impairment (aMCI) and schizophrenia patients, respectively. However, NAP was not found to be an effective treatment (though well-tolerated) for progressive supranuclear palsy (PSP) patients. Here we review the molecular mechanism of NAP activity on MTs and how NAP modulates the MT-Tau-EBs crosstalk. We offer a molecular explanation for the different protective potency of NAP in selected tauopathies (aMCI vs. PSP) expressing different ratios/pathologies of the alternatively spliced Tau mRNA and its resulting protein (aMCI expressing similar quantities of the dynamic Tau 3-MT binding isoform (Tau3R) and the Tau 4-MT binding isoform (Tau4R) and PSP enriched in Tau4R pathology). We reveal the direct effect of truncated ADNPs (resulting from de novo autism and newly discovered Alzheimer's disease-related somatic mutations) on MT dynamics. We show that the peptide SKIP affects MT dynamics and MT-Tau association. Since MT impairment is linked with neurodegenerative and neurodevelopmental conditions, the current study implicates a paucity/dysregulation of MT-interacting endogenous proteins, like ADNP, as a contributing mechanism and provides hope for NAP and SKIP as MT-modulating drug candidates. |
| ADNP, FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32926339  Title: Sex-and Region-Dependent Expression of the Autism-Linked ADNP Correlates with Social- and Speech-Related Genes in the Canary Brain.  The activity-dependent neuroprotective protein (ADNP) syndrome is an autistic-like disorder, instigated by mutations in ADNP. This syndrome is characterized by developmental delays, impairments in speech, motor function, abnormal hearing, and intellectual disabilities. In the Adnp-haploinsufficient mouse model, many of these impediments are evident, appearing in a sex-dependent manner. In zebra finch songbird (ZF; Taeniopygia guttata), an animal model used for song/language studies, ADNP mRNA most robust expression is observed in the cerebrum of young males, potentially corroborating with male ZF exclusive singing behavior and developed cerebral song system. Herein, we report a similar sex-dependent ADNP expression profile, with the highest expression in the cerebrum (qRT-PCR) in the brain of another songbird, the domesticated canary (Serinus canaria domestica). Additional analyses for the mRNA transcripts of the ADNP regulator, vasoactive intestinal peptide (VIP), sister gene ADNP2, and speech-related Forkhead box protein P2 (FoxP2) revealed multiple sex and brain region-dependent positive correlations between the genes (including ADNP). Parallel transcript expression patterns for FoxP2 and VIP were observed alongside specific FoxP2 increase in males compared with females as well as VIP/ADNP2 correlations. In spatial view, a sexually independent extensive form of expression was found for ADNP in the canary cerebrum (RNA in situ hybridization). The songbird cerebral mesopallium area stood out as a potentially high-expressing ADNP tissue, further strengthening the association of ADNP with sense integration and auditory memory formation, previously implicated in mouse and human. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/32918531  Title: Developmental Predictors of Cognitive and Adaptive Outcomes in Genetic Subtypes of Autism Spectrum Disorder.  Approximately one-fourth of autism spectrum disorder (ASD) cases are associated with a disruptive genetic variant. Many of these ASD genotypes have been described previously, and are characterized by unique constellations of medical, psychiatric, developmental, and behavioral features. Development of precision medicine care for affected individuals has been challenging due to the phenotypic heterogeneity that exists even within each genetic subtype. In the present study, we identify developmental milestones that predict cognitive and adaptive outcomes for five of the most common ASD genotypes. Sixty-five youth with a known pathogenic variant involving ADNP, CHD8, DYRK1A, GRIN2B, or SCN2A genes participated in cognitive and adaptive testing. Exploratory linear regressions were used to identify developmental milestones that predicted cognitive and adaptive outcomes within each gene group. We hypothesized that the earliest and most predictive milestones would vary across gene groups, but would be consistent across outcomes within each genetic subtype. Within the ADNP group, age of walking predicted cognitive outcomes, while age of first words predicted adaptive behaviors. Age of phrases predicted adaptive functioning in the CHD8 group, but cognitive outcomes were not clearly associated with early developmental milestones. Verbal milestones were the strongest predictors of cognitive and adaptive outcomes for individuals with mutations to DYRK1A, GRIN2B, or SCN2A. These trends inform decisions about treatment planning and long-term expectations for affected individuals, and they add to the growing body of research linking molecular genetic function to brain development and phenotypic outcomes. LAY SUMMARY: Researchers have found many genetic causes of autism including mutations to ADNP, CHD8, DYRK1A, GRIN2B, and SCN2A genes. We found that each genetic cause had different early developmental milestones that explained the overall functioning of the children when they were older. Depending on the genetic cause, the age that a child first starts walking and/or talking may help to better understand and support a child's development who has a mutation to one of the above genes. Autism Res 2020, 13: 1659-1669. © 2020 International Society for Autism Research and Wiley Periodicals LLC. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/32661233  Title: Tauopathy in the young autistic brain: novel biomarker and therapeutic target.  Given our recent discovery of somatic mutations in autism spectrum disorder (ASD)/intellectual disability (ID) genes in postmortem aged Alzheimer's disease brains correlating with increasing tauopathy, it is important to decipher if tauopathy is underlying brain imaging results of atrophy in ASD/ID children. We concentrated on activity-dependent neuroprotective protein (ADNP), a prevalent autism gene. The unique availability of multiple postmortem brain sections of a 7-year-old male, heterozygous for ADNP de novo mutation c.2244Adup/p.His559Glnfs\*3 allowed exploration of tauopathy, reflecting on a general unexplored mechanism. The tested subject exhibited autism, fine motor delays, severe intellectual disability and seizures. The patient died after multiple organ failure following liver transplantation. To compare to other ADNP syndrome mutations, immortalized lymphoblastoid cell lines from three different patients (including ADNP p.Arg216\*, p.Lys408Valfs\*31, and p.Tyr719\* heterozygous dominant mutations) and a control were subjected to RNA-seq. Immunohistochemistry, high-throughput gene expression profiles in numerous postmortem tissues followed. Comparisons to a control brain and to extensive datasets were used. Live cell imaging investigated Tau-microtubule interaction, protecting against tauopathy. Extensive child brain tauopathy paralleled by multiple gene expression changes was discovered. Tauopathy was explained by direct mutation effects on Tau-microtubule interaction and correction by the ADNP active snippet NAP. Significant pathway changes (empirical P value 50% of the tested genes), including NLGN1, NLGN2, PAX6, SMARCA4, and SNAP25, converging on nervous system development and tauopathy. NAP provided protection against mutated ADNP disrupted Tau-microtubule association. In conclusion, tauopathy may explain brain-imaging findings in ADNP syndrome children and may provide a new direction for the development of tauopathy protecting drug candidates like NAP in ASD/ID. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/32072336  Title: Microbiota changes associated with ADNP deficiencies: rapid indicators for NAP (CP201) treatment of the ADNP syndrome and beyond.  Activity-dependent neuroprotective protein (ADNP) and its protein snippet NAP (drug candidate CP201) regulate synapse formation and cognitive as well as behavioral functions, in part, through microtubule interaction. Given potential interactions between the microbiome and brain function, we now investigated the potential effects of the ADNP-deficient genotype, mimicking the ADNP syndrome on microbiota composition in the Adnp+/- mouse model. We have discovered a surprising robust sexually dichotomized Adnp genotype effect and correction by NAP (CP201) as follows. Most of the commensal bacterial microbiota tested were affected by the Adnp genotype and corrected by NAP treatment in a male sex-dependent manner. The following list includes all the bacterial groups tested-labeled in bold are male Adnp-genotype increased and corrected (decreased) by NAP. (1) Eubacteriaceae (EubV3), (2) Enterobacteriaceae (Entero), (3) Enterococcus genus (gEncocc), (4) Lactobacillus group (Lacto), (5) Bifidobacterium genus (BIF), (6) Bacteroides/Prevotella species (Bac), (7) Clostridium coccoides group (Coer), (8) Clostridium leptum group (Cluster IV, sgClep), and (9) Mouse intestinal Bacteroides (MIB). No similarities were found between males and females regarding sex- and genotype-dependent microbiota distributions. Furthermore, a female Adnp+/- genotype associated decrease (contrasting male increase) was observed in the Lactobacillus group (Lacto). Significant correlations were discovered between specific bacterial group loads and open-field behavior as well as social recognition behaviors. In summary, we discovered ADNP deficiency associated changes in commensal gut microbiota compositions, a sex-dependent biomarker for the ADNP syndrome and beyond. Strikingly, we discovered rapidly detected NAP (CP201) treatment-dependent biomarkers within the gut microbiota. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/31992971  Title: VIP/PACAP-Based Drug Development: The ADNP/NAP-Derived Mirror Peptides SKIP and D-SKIP Exhibit Distinctive in vivo and in silico Effects.  Activity-dependent neuroprotective protein (ADNP) was discovered and first characterized in the laboratory of Prof. Illana Gozes to be regulated by vasoactive intestinal peptide (VIP), and pituitary adenylate cyclase-activating peptide (PACAP) toward neuroprotection. Importantly, ADNP is a master regulator of >400 genes, essential for brain formation, while its haploinsufficiency causes cognitive impairments. Recently, de novo mutations in ADNP were identified as leading to the autism-like ADNP syndrome, mimicked by the Adnp-deficient mouse model. Furthermore, novel peptide derivatives of the neuroprotective ADNP-snippet NAP (NAPVSIPQ), developed in our laboratory, include SKIP and the mirroring all D-amino acid SKIP (D-SKIP). We now extended previous evidence suggesting potential antagonistic features for D-SKIP, compared with the neuroprotective peptide SKIP, as was observed by NMR analysis and social/olfactory functional testing. Here, an impact of the Adnp genotype was observed in the Morris Water Maze (MWM) test measuring cognition, coupled with improvement by SKIP, opposing the inert/exacerbating effect of D-SKIP. In the elevated plus-maze and open field tests measuring anxiety-related behaviors, contrasting effects of SKIP and D-SKIP were found, with SKIP improving/preserving the normal phenotype of the mouse, and D-SKIP causing alterations. Lastly, an in silico analysis suggested that SKIP and D-SKIP bind the microtubule end binding (EB) proteins EB1 and EB3 in different conformations, thereby indicating distinctive natures for the two peptides, potentially mediating differential in vivo effects. Altogether, our findings corroborate the notion of D-SKIP acting as an antagonist, thus distinguishing it from the neuroprotective SKIP. |
| ADNP, CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/31872500  Title: Molecular causes of sex-specific deficits in rodent models of neurodevelopmental disorders.  Neurodevelopmental disorders (NDDs) such as intellectual disability and autism spectrum disorder consistently show a male bias in prevalence, but it remains unclear why males and females are affected with different frequency. While many behavioral studies of transgenic NDD models have focused only on males, the requirement by the National Institutes of Health to consider sex as a biological variable has promoted the comparison of male and female performance in wild-type and mutant animals. Here, we review examples of rodent models of NDDs in which sex-specific deficits were identified in molecular, physiological, and/or behavioral responses, showing sex differences in susceptibility to disruption of genes mutated in NDDs. Haploinsufficiency in genes involved in mechanisms such as synaptic function (GABRB3 and NRXN1), chromatin remodeling (CHD8, EMHT1, and ADNP), and intracellular signaling (CC2D1A and ERK1) lead to more severe behavioral outcomes in males. However, in the absence of behavioral deficits, females can still present with cellular and electrophysiological changes that could be due to compensatory mechanisms or differential allocation of molecular and cellular functions in the two sexes. By contrasting these findings with mouse models where females are more severely affected (MTHFR and AMBRA1), we propose a framework to approach the study of sex-specific deficits possibly leading to sex bias in NDDs. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/31664177  Title: Discovery of autism/intellectual disability somatic mutations in Alzheimer's brains: mutated ADNP cytoskeletal impairments and repair as a case study.  With Alzheimer's disease (AD) exhibiting reduced ability of neural stem cell renewal, we hypothesized that de novo mutations controlling embryonic development, in the form of brain somatic mutations instigate the disease. A leading gene presenting heterozygous dominant de novo autism-intellectual disabilities (ID) causing mutations is activity-dependent neuroprotective protein (ADNP), with intact ADNP protecting against AD-tauopathy. We discovered a genomic autism ADNP mutation (c.2188C>T) in postmortem AD olfactory bulbs and hippocampi. RNA-Seq of olfactory bulbs also identified a novel ADNP hotspot mutation, c.2187\_2188insA. Altogether, 665 mutations in 596 genes with 441 mutations in AD patients (389 genes, 38% AD-exclusive mutations) and 104 genes presenting disease-causing mutations (OMIM) were discovered. OMIM AD mutated genes converged on cytoskeletal mechanisms, autism and ID causing mutations (about 40% each). The number and average frequencies of AD-related mutations per subject were higher in AD subjects compared to controls. RNA-seq datamining (hippocampus, dorsolateral prefrontal cortex, fusiform gyrus and superior frontal gyrus-583 subjects) yielded similar results. Overlapping all tested brain areas identified unique and shared mutations, with ADNP singled out as a gene associated with autism/ID/AD and presenting several unique aging/AD mutations. The large fusiform gyrus library (117 subjects) with high sequencing coverage correlated the c.2187\_2188insA ADNP mutation frequency to Braak stage (tauopathy) and showed more ADNP mutations in AD specimens. In cell cultures, the ADNP-derived snippet NAP inhibited mutated-ADNP-microtubule (MT) toxicity and enhanced Tau-MT association. We propose a paradigm-shifting concept in the perception of AD whereby accumulating mosaic somatic mutations promote brain pathology. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/31534115  Title: The autism-mutated ADNP plays a key role in stress response.  Activity-dependent neuroprotective protein (ADNP), discovered and first characterized in our laboratory (IG), is vital for mammalian brain formation and presents one of the leading genes mutated de novo causing an autistic syndrome, namely the ADNP syndrome. Furthermore, a unique mouse model of Adnp-haploinsufficiency was developed in the laboratory (IG), with mice exhibiting cognitive and social deficiencies. ADNP is regulated by vasoactive intestinal peptide (VIP), and pituitary adenylate cyclase-activating peptide (PACAP). In this respect, PACAP was independently identified as a sexual divergent master regulator of the stress response. Here, we sought to determine the impact of the Adnp genotype and the efficacy of PACAP pre-treatment when subjecting Adnp+/- mice to stressful conditions. Significant sex differences were observed with Adnp+/- males being more susceptible to stress in the object and social recognition tests, and the females more susceptible in the open field and elevated plus maze tests. Splenic Adnp expression and plasma cortisol levels in mice were correlated with cognition (male mice) and anxiety-related behavior. These findings were further translated to humans, with observed correlations between ADNP expression and stress/cortisol content in a young men cohort. Altogether, our current results may establish ADNP as a marker of stress response. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/31035039  Title: GENYOi004-A: An induced pluripotent stem cells (iPSCs) line generated from a patient with autism-related ADNP syndrome carrying a pTyr719\* mutation.  ADNP syndrome is an intellectual disability associated with Autism spectrum disorder caused by mutations in ADNP. We generated an iPSC line from an ADNP syndrome pediatric patient harboring the mutation p.Trp719\* (GENYOi004-A). Peripheral blood mononuclear cells were reprogrammed using a non-transmissible form of Sendai viruses expressing the four Yamanaka factors (Oct3/4, SOX2, KLF4 and c-MYC). Characterization of GENYOi004-A included mutation analysis of ADNP by allele-specific PCR, genetic identity by Short Tandem Repeats polymorphism profiling, alkaline phosphatase enzymatic activity, expression of pluripotency-associated factors and pluripotency studies in vivo. GENYOi004-A will be useful to evaluate ADNP syndrome alterations at early developmental stages. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/30664622  Title: The autism/neuroprotection-linked ADNP/NAP regulate the excitatory glutamatergic synapse.  Activity-dependent neuroprotective protein (ADNP), essential for brain formation, was discovered as a leading de novo mutated gene causing the autism-like ADNP syndrome. This syndrome is phenotypically characterized by global developmental delays, intellectual disabilities, speech impediments, and motor dysfunctions. The Adnp haploinsufficient mouse mimics the human ADNP syndrome in terms of synapse density and gene expression patterns, as well as in developmental, motor, and cognitive abilities. Peripheral ADNP was also discovered as a biomarker for Alzheimer's disease and schizophrenia, with nasal administration of the ADNP snippet peptide NAP (enhancing endogenous ADNP activity) leading to partial cognitive and functional protection at the cellular, animal and clinical settings. Here, a novel formulation for effective delivery of NAP is provided with superior brain penetration capabilities. Also provided are methods for treating pertinent clinical implications such as autism, cognitive impairments, olfactory deficits, and muscle strength using the formulation in the Adnp haploinsufficient mouse. Results showed a dramatically specific increase in brain/body bioavailability with the new formulation, without breaching the blood brain barrier. Additional findings included improvements using daily intranasal treatments with NAP, at the behavioral and brain structural levels, diffusion tensor imaging (DTI), translatable to clinical practice. Significant effects on hippocampal and cerebral cortical expression of the presynaptic Slc17a7 gene encoding vesicular excitatory glutamate transporter 1 (VGLUT1) were observed at the RNA and immunohistochemical levels, explaining the DTI results. These findings tie for the first time a reduction in presynaptic glutamatergic synapses with the autism/Alzheimer's/schizophrenia-linked ADNP deficiency coupled with amelioration by NAP (CP201). |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/30659505  Title: Atypical Auditory Brainstem Response and Protein Expression Aberrations Related to ASD and Hearing Loss in the Adnp Haploinsufficient Mouse Brain.  Autism is a wide spread neurodevelopmental disorder with growing morbidity rates, affecting more boys than girls worldwide. Activity-dependent neuroprotective protein (ADNP) was recently recognized as a leading gene accounted for 0.17% of autism spectrum disorder (ASD) cases globally. Respectively, mutations in the human ADNP gene (ADNP syndrome), cause multi-system body dysfunctions with apparent ASD-related traits, commencing as early as childhood. The Adnp haploinsufficient (Adnp+/-) mouse model was researched before in relations to Alzheimer's disease and autism. Adnp+/- mice suffer from deficient social memory, vocal and motor impediments, irregular tooth eruption and short stature, all of which corresponds with reported phenotypes in patients with the ADNP syndrome. Recently, a more elaborated description of the ADNP syndrome was published, presenting impediments such as hearing disabilities in > 10% of the studied children. Irregular auditory brainstem response (ABR) has been connected to ASD-related cases and has been suggested as a potential hallmark for autism, allowing diagnosis of ASD risk and early intervention. Herein, we present detriment hearing in the Adnp+/- mice with atypical ABR and significant protein expression irregularities that coincides with ASD and hearing loss studies in the brain. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/30106381  Title: Activity-dependent neuroprotective protein deficiency models synaptic and developmental phenotypes of autism-like syndrome.  Previous findings showed that in mice, complete knockout of activity-dependent neuroprotective protein (ADNP) abolishes brain formation, while haploinsufficiency (Adnp+/-) causes cognitive impairments. We hypothesized that mutations in ADNP lead to a developmental/autistic syndrome in children. Indeed, recent phenotypic characterization of children harboring ADNP mutations (ADNP syndrome children) revealed global developmental delays and intellectual disabilities, including speech and motor dysfunctions. Mechanistically, ADNP includes a SIP motif embedded in the ADNP-derived snippet drug candidate NAP (NAPVSIPQ, also known as CP201), which binds to microtubule end-binding protein 3, essential for dendritic spine formation. Here, we established a unique neuronal membrane-tagged, GFP-expressing Adnp+/- mouse line allowing in vivo synaptic pathology quantification. We discovered that Adnp deficiency reduced dendritic spine density and altered synaptic gene expression, both of which were partly ameliorated by NAP treatment. Adnp+/-mice further exhibited global developmental delays, vocalization impediments, gait and motor dysfunctions, and social and object memory impairments, all of which were partially reversed by daily NAP administration (systemic/nasal). In conclusion, we have connected ADNP-related synaptic pathology to developmental and behavioral outcomes, establishing NAP in vivo target engagement and identifying potential biomarkers. Together, these studies pave a path toward the clinical development of NAP (CP201) for the treatment of ADNP syndrome. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/29724491  Title: Clinical Presentation of a Complex Neurodevelopmental Disorder Caused by Mutations in ADNP.  In genome-wide screening studies for de novo mutations underlying autism and intellectual disability, mutations in the ADNP gene are consistently reported among the most frequent. ADNP mutations have been identified in children with autism spectrum disorder comorbid with intellectual disability, distinctive facial features, and deficits in multiple organ systems. However, a comprehensive clinical description of the Helsmoortel-Van der Aa syndrome is lacking. We identified a worldwide cohort of 78 individuals with likely disruptive mutations in ADNP from January 2014 to October 2016 through systematic literature search, by contacting collaborators, and through direct interaction with parents. Clinicians filled in a structured questionnaire on genetic and clinical findings to enable correlations between genotype and phenotype. Clinical photographs and specialist reports were gathered. Parents were interviewed to complement the written questionnaires. We report on the detailed clinical characterization of a large cohort of individuals with an ADNP mutation and demonstrate a distinctive combination of clinical features, including mild to severe intellectual disability, autism, severe speech and motor delay, and common facial characteristics. Brain abnormalities, behavioral problems, sleep disturbance, epilepsy, hypotonia, visual problems, congenital heart defects, gastrointestinal problems, short stature, and hormonal deficiencies are common comorbidities. Strikingly, individuals with the recurrent p.Tyr719\* mutation were more severely affected. This overview defines the full clinical spectrum of individuals with ADNP mutations, a specific autism subtype. We show that individuals with mutations in ADNP have many overlapping clinical features that are distinctive from those of other autism and/or intellectual disability syndromes. In addition, our data show preliminary evidence of a correlation between genotype and phenotype. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/28940660  Title: ADNP Plays a Key Role in Autophagy: From Autism to Schizophrenia and Alzheimer's Disease.  Activity-dependent neuroprotective protein (ADNP), discovered in our laboratory in 1999, has been characterized as a master gene vital for mammalian brain formation. ADNP de novo mutations in humans result in a syndromic form of autism-like spectrum disorder (ASD), including cognitive and motor deficits, the ADNP syndrome (Helsmoortel-Van Der Aa). One of the most important cellular processes associated with ADNP is the autophagy pathway, recently discovered by us as a key player in the pathophysiology of schizophrenia. In this regard, given the link between the microtubule and autophagy systems, the ADNP microtubule end binding protein motif, namely, the neuroprotective NAP (NAPVSIPQ), was found to enhance autophagy while protecting microtubules and augmenting ADNP's association with both systems. Thus, linking autophagy and ADNP is proposed as a major target for intervention in brain diseases from autism to Alzheimer's disease (AD) and our findings introduce autophagy as a possible novel target for treating schizophrenia. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/28579975  Title: The Eight and a Half Year Journey of Undiagnosed AD: Gene Sequencing and Funding of Advanced Genetic Testing Has Led to Hope and New Beginnings.  Activity-dependent neuroprotective protein (ADNP) is one of the most prevalent de novo mutated genes in syndromic autism spectrum disorders, driving a general interest in the gene and the syndrome. The aim of this study was to provide a detailed developmental case study of ADNP p.Tyr719\* mutation toward improvements in (1) diagnostic procedures, (2) phenotypic scope, and (3) interventions. Longitudinal clinical and parental reports. AD (currently 11-year-old) had several rare congenital anomalies including imperforate anus that was surgically repaired at 2 days of age. Her findings were craniofacial asymmetries, global developmental delay, autistic behaviors (loss of smile and inability to make eye contact at the age of 15 months), and slow thriving as she gradually matures. Comprehensive diagnostic procedures at 3 years resulted in no definitive diagnosis. With parental persistence, AD began walking at 3.5 years (skipping crawling). At the age of 8.5 years, AD was subjected to whole exome sequencing, compared to the parents and diagnosed as carrying an ADNP p.Tyr719\* mutation, a causal recurring mutation in ADNP (currently ~17/80 worldwide). Brain magnetic resonance imaging demonstrated mild generalized cerebral volume loss with reduced posterior white matter. AD is non-verbal, communicating with signs and word approximations. She continues to make slow but forward developmental progress, and her case teaches newly diagnosed children within the ADNP Kids Research Foundation. This case study emphasizes the importance of diagnosis and describes, for the first time, early motor intervention therapies. Detailed developmental profile of selected cases leads to better treatments. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/28221363  Title: Premature primary tooth eruption in cognitive/motor-delayed ADNP-mutated children.  A major flaw in autism spectrum disorder (ASD) management is late diagnosis. Activity-dependent neuroprotective protein (ADNP) is a most frequent de novo mutated ASD-related gene. Functionally, ADNP protects nerve cells against electrical blockade. In mice, complete Adnp deficiency results in dysregulation of over 400 genes and failure to form a brain. Adnp haploinsufficiency results in cognitive and social deficiencies coupled to sex- and age-dependent deficits in the key microtubule and ion channel pathways. Here, collaborating with parents/caregivers globally, we discovered premature tooth eruption as a potential early diagnostic biomarker for ADNP mutation. The parents of 44/54 ADNP-mutated children reported an almost full erupted dentition by 1 year of age, including molars and only 10 of the children had teeth within the normal developmental time range. Looking at Adnp-deficient mice, by computed tomography, showed significantly smaller dental sacs and tooth buds at 5 days of age in the deficient mice compared to littermate controls. There was only trending at 2 days, implicating age-dependent dysregulation of teething in Adnp-deficient mice. Allen Atlas analysis showed Adnp expression in the jaw area. RNA sequencing (RNAseq) and gene array analysis of human ADNP-mutated lymphoblastoids, whole-mouse embryos and mouse brains identified dysregulation of bone/nervous system-controlling genes resulting from ADNP mutation/deficiency (for example, BMP1 and BMP4). AKAP6, discovered here as a major gene regulated by ADNP, also links cognition and bone maintenance. To the best of our knowledge, this is the first time that early primary (deciduous) teething is related to the ADNP syndrome, providing for early/simple diagnosis and paving the path to early intervention/specialized treatment plan. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/28115743  Title: ADNP/NAP dramatically increase microtubule end-binding protein-Tau interaction: a novel avenue for protection against tauopathy.  Activity-dependent neuroprotective protein (ADNP), vital for brain formation and cognitive function, is mutated in autism and linked to neurodegenerative/psychiatric diseases. An eight-amino-acid peptide snippet of ADNP, NAP (NAPVSIPQ), identified as a smallest active fragment, includes the SxIP microtubule (MT) end-binding protein (EB) association motif, and enhances ADNP-EB3 interaction. Depletion of EB1 or EB3 abolishes NAP protection against zinc intoxication. Furthermore, NAP enhances Tau-MT interaction, and Tau regulates the localization and function of EB1 and EB3 in developing neuronal cells. Here, we asked how NAP (ADNP) enhances Tau-MT interactions and whether this is mediated by EBs. We showed, for we believe the first time, that NAP augmented endogenous EB1 comet density in the N1E-115 neuroblastoma neuronal model. This finding was substantiated by cell transfection with fluorescent EB1 and live cell imaging. NAP increased comet amounts, length and speed. At the molecular level, NAP enhanced EB3 homodimer formation, while decreasing EB1-EB3 heterodimer content and driving EB1- and EB3-Tau interactions (dramatic 20-fold increases), leading to recruitment of EB1/EB3 and Tau to MTs under zinc intoxication. Our previous results showed that while NAP protected neuronal-like cells against oxidative stress, it did not protect NIH3T3 fibroblasts. Here, NAP did not protect NIH3T3 cells against zinc intoxication, unless these cells were transfected with Tau. Interestingly, other MT associated proteins (MAPs) may replace Tau, thus, EB-Tau (MAPs) interaction is identified as a novel target for endogenous ADNP neuroprotection, and a future target for drug development, with NAP as a prototype. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/27870441  Title: Sexual divergence in activity-dependent neuroprotective protein impacting autism, schizophrenia, and Alzheimer's disease.  Discovered in our laboratory, activity-dependent neuroprotective protein (ADNP) interacts with key regulatory proteins, including the chromatin remodeling complex SWI/SNF, proteins associated with RNA splicing, RNA translation, microtubule dynamics, and autophagy. ADNP regulates > 400 genes during mouse embryonic development and is essential for neural tube closure. ADNP key functions extend from mice to men, with mutations causing ADNP-related ID/autism syndrome, also known as the Helsmoortel-Van der Aa syndrome. ADNP mRNA increases in lymphocytes derived from schizophrenia patients and in patients suffering from mild cognitive impairment (MCI) and further increases in Alzheimer's disease patients compared with controls. Serum ADNP levels correlate with IQ. NAP (davunetide), an ADNP snippet drug candidate, protects cognition in patients suffering from amnestic MCI preceding Alzheimer's disease and significantly enhances functional daily activities in schizophrenia patients toward future development. It is important to note that ADNP is sexually regulated in the brains of birds, mice, and men and in lymphocytes of patients suffering from schizophrenia. ADNP haploinsufficiency in mice results in significantly decreased axonal transport (with male-female differences) changes in gene expression in a sex-dependent manner, including key regulatory mechanisms during brain and heart development and function and behavioral outcomes. These findings pave the path for better understanding of brain function through the prism of sex differences. © 2016 Wiley Periodicals, Inc. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/26782054  Title: Sexual divergence in microtubule function: the novel intranasal microtubule targeting SKIP normalizes axonal transport and enhances memory.  Activity-dependent neuroprotective protein (ADNP), essential for brain formation, is a frequent autism spectrum disorder (ASD)-mutated gene. ADNP associates with microtubule end-binding proteins (EBs) through its SxIP motif, to regulate dendritic spine formation and brain plasticity. Here, we reveal SKIP, a novel four-amino-acid peptide representing an EB-binding site, as a replacement therapy in an outbred Adnp-deficient mouse model. We discovered, for the first time, axonal transport deficits in Adnp(+/-) mice (measured by manganese-enhanced magnetic resonance imaging), with significant male-female differences. RNA sequencing evaluations showed major age, sex and genotype differences. Function enrichment and focus on major gene expression changes further implicated channel/transporter function and the cytoskeleton. In particular, a significant maturation change (1 month-five months) was observed in beta1 tubulin (Tubb1) mRNA, only in Adnp(+/+) males, and sex-dependent increase in calcium channel mRNA (Cacna1e) in Adnp(+/+) males compared with females. At the protein level, the Adnp(+/-) mice exhibited impaired hippocampal expression of the calcium channel (voltage-dependent calcium channel, Cacnb1) as well as other key ASD-linked genes including the serotonin transporter (Slc6a4), and the autophagy regulator, BECN1 (Beclin1), in a sex-dependent manner. Intranasal SKIP treatment normalized social memory in 8- to 9-month-old Adnp(+/-)-treated mice to placebo-control levels, while protecting axonal transport and ameliorating changes in ASD-like gene expression. The control, all d-amino analog D-SKIP, did not mimic SKIP activity. SKIP presents a novel prototype for potential ASD drug development, a prevalent unmet medical need. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/26578950  Title: ADNP: in search for molecular mechanisms and innovative therapeutic strategies for frontotemporal degeneration.  Activity-dependent neuroprotective protein (ADNP) is deregulated in Alzheimer's disease (AD) and in schizophrenia and mutated in autism. In mice, ADNP is essential for brain formation and ADNP haploinsufficiency is associated with cognitive and social deficits and tauopathy. Tauopathy, a major pathology in AD, is also found in ~45% of frontotemporal dementias (FTDs). Tau transcript, a product of a single gene, undergoes alternative splicing. Tau splicing seems to be altered in FTD brain. In transgenic mice overexpressing a mutated tau in the cerebral cortex, significant increases in ADNP transcript expression were observed in the cerebral cortex of young transgenic mice (~disease onset) and a marked decrease with aging as compared to control littermates. ADNP is a member of the SWItch/Sucrose NonFermentable (SWI/SNF) chromatin remodeling complex also associated with alternative splicing, including tau transcript splicing. Further cellular interactions of ADNP include association with microtubules, with tau being a microtubule-associated protein. NAP (davundetide), a novel drug candidate derived from ADNP, increases ADNP-microtubule association and protects against tauopathy and cognitive deficiencies in mice. Although, NAP did not provide protection in progressive supranuclear palsy (PSP), a pure tauopathy, it increased cognitive scores in amnestic mild cognitively impaired patients and protected functional activity in schizophrenia patients. This mini-review focuses on ADNP in the context of FTD and tau/microtubules and proposes NAP as a novel drug target for future clinical evaluations. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/25955282  Title: The cytoskeleton as a drug target for neuroprotection: the case of the autism- mutated ADNP.  Fifteen years ago we discovered activity-dependent neuroprotective protein (ADNP), and showed that it is essential for brain formation/function. Our protein interaction studies identified ADNP as a member of the chromatin remodeling complex, SWI/SNF also associated with alternative splicing of tau and prediction of tauopathy. Recently, we have identified cytoplasmic ADNP interactions with the autophagy regulating microtubule-associated protein 1 light chain 3 (LC3) and with microtubule end-binding (EB) proteins. The ADNP-EB-binding SIP domain is shared with the ADNP snippet drug candidate, NAPVSIPQ termed NAP (davunetide). Thus, we identified a precise target for ADNP/NAP (davunetide) neuroprotection toward improved drug development. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/25895853  Title: ADNP: A major autism mutated gene is differentially distributed (age and gender) in the songbird brain.  ADNP is a protein necessary for brain development, important for brain plasticity, cognitive and social functioning, characteristics that are all impaired in autism and in the Adnp(+/-) mouse model, in a sex-dependent manner. ADNP was originally discovered as a protein that is secreted from glial cells in response to vasoactive intestinal peptide (VIP). VIP is a major neuroprotective peptide in the CNS and PNS and was also associated with social recognition in rodents and aggression, pair-bonding and parental behaviors in birds. Comparative sequence alignment revealed high evolutionary conservation of ADNP in Chordata. Despite its importance in brain function, ADNP has never been studied in birds. Zebra finches (Taeniopygia guttata) are highly social songbirds that have a sexually dichotomous anatomical brain structure, with males demonstrating a developed song system, presenting a model to study behavior and potential sexually dependent fundamental differences. Here, using quantitative real time polymerase chain reaction (qRT-PCR), we discovered sexually dichotomous and age related differences in ADNP mRNA expression in three different regions of the song bird brain-cerebellum, cerebrum, and brain stem. Higher levels of ADNP mRNA were specifically found in young male compared to the female cerebrum, while aging caused a significant 2 and 3-fold decrease in the female and male cerebrum, respectively. Furthermore, a comparison between the three tested brain regions revealed unique sex-dependent ADNP mRNA distribution patterns, affected by aging. Future studies are aimed at deciphering the function of ADNP in birds, toward a better molecular understanding of sexual dichotomy in singing behavior in birds. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/25178163  Title: The NAP motif of activity-dependent neuroprotective protein (ADNP) regulates dendritic spines through microtubule end binding proteins.  The NAP motif of activity-dependent neuroprotective protein (ADNP) enhanced memory scores in patients suffering from mild cognitive impairment and protected activities of daily living in schizophrenia patients, while fortifying microtubule (MT)-dependent axonal transport, in mice and flies. The question is how does NAP fortify MTs? Our sequence analysis identified the MT end-binding protein (EB1)-interacting motif SxIP (SIP, Ser-Ile-Pro) in ADNP/NAP and showed specific SxIP binding sites in all members of the EB protein family (EB1-3). Others found that EB1 enhancement of neurite outgrowth is attenuated by EB2, while EB3 interacts with postsynaptic density protein 95 (PSD-95) to modulate dendritic plasticity. Here, NAP increased PSD-95 expression in dendritic spines, which was inhibited by EB3 silencing. EB1 or EB3, but not EB2 silencing inhibited NAP-mediated cell protection, which reflected NAP binding specificity. NAPVSKIPQ (SxIP=SKIP), but not NAPVAAAAQ mimicked NAP activity. ADNP, essential for neuronal differentiation and brain formation in mouse, a member of the SWI/SNF chromatin remodeling complex and a major protein mutated in autism and deregulated in schizophrenia in men, showed similar EB interactions, which were enhanced by NAP treatment. The newly identified shared MT target of NAP/ADNP is directly implicated in synaptic plasticity, explaining the breadth and efficiency of neuroprotective/neurotrophic capacities. |
| ADNP |
| Url: https://pubmed.ncbi.nlm.nih.gov/25169753  Title: The transcriptional regulator ADNP links the BAF (SWI/SNF) complexes with autism.  Mutations in ADNP were recently identified as a frequent cause of syndromic autism, characterized by deficits in social communication and interaction and restricted, repetitive behavioral patterns. Based on its functional domains, ADNP is a presumed transcription factor. The gene interacts closely with the SWI/SNF complex by direct and experimentally verified binding of its C-terminus to three of its core components. A detailed and systematic clinical assessment of the symptoms observed in our patients allows a detailed comparison with the symptoms observed in other SWI/SNF disorders. While the mutational mechanism of the first 10 patients identified suggested a gain of function mechanism, an 11th patient reported here is predicted haploinsufficient. The latter observation may raise hope for therapy, as addition of NAP, a neuroprotective octapeptide named after the first three amino acids of the sequence NAPVSPIQ, has been reported by others to ameliorate some of the cognitive abnormalities observed in a knockout mouse model. It is concluded that detailed clinical and molecular studies on larger cohorts of patients are necessary to establish a better insight in the genotype phenotype correlation and in the mutational mechanism. |
| AHDC1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34073322  Title: Focusing on Autism Spectrum Disorder in Xia-Gibbs Syndrome: Description of a Female with High Functioning Autism and Literature Review.  Xia-Gibbs syndrome (XGS) is a rare disorder caused by de novo mutations in the AT-Hook DNA binding motif Containing 1 (AHDC1) gene, which is characterised by a wide spectrum of clinical manifestations, including global developmental delay, intellectual disability, structural abnormalities of the brain, global hypotonia, feeding problems, sleep difficulties and apnoea, facial dysmorphisms, and short stature. Here, we report on a girl patient who shows a peculiar cognitive and behavioural profile including high-functioning autism spectrum disorder (ASD) without intellectual disability and provide information on her developmental trajectory with the aim of expanding knowledge of the XGS clinical spectrum. On the basis of the current clinical case and the literature review, we also attempt to deepen understanding of behavioural and psychiatric manifestations associated with XGS. In addition to the patient we described, a considerable rate of individuals with XGS display autistic symptoms or have been diagnosed with an autistic spectrum disorder. Moreover, the analysis of the few psychopathological profiles of patients with XGS described in the literature shows a frequent presence of aggressive and self-injurious behaviours that could be either an expression of autistic functioning or an additional symptom of the ASD evolution. A careful investigation of the abovementioned symptoms is therefore required, since they could represent a "red flag" for ASD. |
| AHDC1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27148574  Title: De novo truncating variants in the AHDC1 gene encoding the AT-hook DNA-binding motif-containing protein 1 are associated with intellectual disability and developmental delay.  Whole-exome sequencing (WES) represents a significant breakthrough in clinical genetics, and identifies a genetic etiology in up to 30% of cases of intellectual disability (ID). Using WES, we identified seven unrelated patients with a similar clinical phenotype of severe intellectual disability or neurodevelopmental delay who were all heterozygous for de novo truncating variants in the AT-hook DNA-binding motif-containing protein 1 (AHDC1). The patients were all minimally verbal or nonverbal and had variable neurological problems including spastic quadriplegia, ataxia, nystagmus, seizures, autism, and self-injurious behaviors. Additional common clinical features include dysmorphic facial features and feeding difficulties associated with failure to thrive and short stature. The AHDC1 gene has only one coding exon, and the protein contains conserved regions including AT-hook motifs and a PDZ binding domain. We postulate that all seven variants detected in these patients result in a truncated protein missing critical functional domains, disrupting interactions with other proteins important for brain development. Our study demonstrates that truncating variants in AHDC1 are associated with ID and are primarily associated with a neurodevelopmental phenotype. |
| AKT3, PIK3CA |
| Url: https://pubmed.ncbi.nlm.nih.gov/35355055  Title: Profiling PI3K-AKT-MTOR variants in focal brain malformations reveals new insights for diagnostic care.  Focal malformations of cortical development including focal cortical dysplasia, hemimegalencephaly and megalencephaly, are a spectrum of neurodevelopmental disorders associated with brain overgrowth, cellular and architectural dysplasia, intractable epilepsy, autism and intellectual disability. Importantly, focal cortical dysplasia is the most common cause of focal intractable paediatric epilepsy. Gain and loss of function variants in the PI3K-AKT-MTOR pathway have been identified in this spectrum, with variable levels of mosaicism and tissue distribution. In this study, we performed deep molecular profiling of common PI3K-AKT-MTOR pathway variants in surgically resected tissues using droplet digital polymerase chain reaction (ddPCR), combined with analysis of key phenotype data. A total of 159 samples, including 124 brain tissue samples, were collected from 58 children with focal malformations of cortical development. We designed an ultra-sensitive and highly targeted molecular diagnostic panel using ddPCR for six mutational hotspots in three PI3K-AKT-MTOR pathway genes, namely PIK3CA (p.E542K, p.E545K, p.H1047R), AKT3 (p.E17K) and MTOR (p.S2215F, p.S2215Y). We quantified the level of mosaicism across all samples and correlated genotypes with key clinical, neuroimaging and histopathological data. Pathogenic variants were identified in 17 individuals, with an overall molecular solve rate of 29.31%. Variant allele fractions ranged from 0.14 to 22.67% across all mutation-positive samples. Our data show that pathogenic MTOR variants are mostly associated with focal cortical dysplasia, whereas pathogenic PIK3CA variants are more frequent in hemimegalencephaly. Further, the presence of one of these hotspot mutations correlated with earlier onset of epilepsy. However, levels of mosaicism did not correlate with the severity of the cortical malformation by neuroimaging or histopathology. Importantly, we could not identify these mutational hotspots in other types of surgically resected epileptic lesions (e.g. polymicrogyria or mesial temporal sclerosis) suggesting that PI3K-AKT-MTOR mutations are specifically causal in the focal cortical dysplasia-hemimegalencephaly spectrum. Finally, our data suggest that ultra-sensitive molecular profiling of the most common PI3K-AKT-MTOR mutations by targeted sequencing droplet digital polymerase chain reaction is an effective molecular approach for these disorders with a good diagnostic yield when paired with neuroimaging and histopathology. |
| AKT3, PIK3CA |
| Url: https://pubmed.ncbi.nlm.nih.gov/31441589  Title: Megalencephaly syndromes associated with mutations of core components of the PI3K-AKT-MTOR pathway: PIK3CA, PIK3R2, AKT3, and MTOR.  Megalencephaly (MEG) is a developmental abnormality of brain growth characterized by early onset, often progressive, brain overgrowth. Focal forms of megalencephaly associated with cortical dysplasia, such as hemimegalencephaly and focal cortical dysplasia, are common causes of focal intractable epilepsy in children. The increasing use of high throughput sequencing methods, including high depth sequencing to more accurately detect and quantify mosaic mutations, has allowed us to identify the molecular etiologies of many MEG syndromes, including most notably the PI3K-AKT-MTOR related MEG disorders. Thorough molecular and clinical characterization of affected individuals further allow us to derive preliminary genotype-phenotype correlations depending on the gene, mutation, level of mosaicism, and tissue distribution. Our review of published data on these disorders so far shows that mildly activating variants (that are typically constitutional or germline) are associated with diffuse megalencephaly with intellectual disability and/or autism spectrum disorder; moderately activating variants (that are typically high-level mosaic) are associated with megalencephaly with pigmentary abnormalities of the skin; and strongly activating variants (that are usually very low-level mosaic) are associated with focal brain malformations including hemimegalencephaly and focal cortical dysplasia. Accurate molecular diagnosis of these disorders is undoubtedly crucial to more optimally treat children with these disorders using PI3K-AKT-MTOR pathway inhibitors. |
| AKT3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28969385  Title: Mutations of AKT3 are associated with a wide spectrum of developmental disorders including extreme megalencephaly.  Mutations of genes within the phosphatidylinositol-3-kinase (PI3K)-AKT-MTOR pathway are well known causes of brain overgrowth (megalencephaly) as well as segmental cortical dysplasia (such as hemimegalencephaly, focal cortical dysplasia and polymicrogyria). Mutations of the AKT3 gene have been reported in a few individuals with brain malformations, to date. Therefore, our understanding regarding the clinical and molecular spectrum associated with mutations of this critical gene is limited, with no clear genotype-phenotype correlations. We sought to further delineate this spectrum, study levels of mosaicism and identify genotype-phenotype correlations of AKT3-related disorders. We performed targeted sequencing of AKT3 on individuals with these phenotypes by molecular inversion probes and/or Sanger sequencing to determine the type and level of mosaicism of mutations. We analysed all clinical and brain imaging data of mutation-positive individuals including neuropathological analysis in one instance. We performed ex vivo kinase assays on AKT3 engineered with the patient mutations and examined the phospholipid binding profile of pleckstrin homology domain localizing mutations. We identified 14 new individuals with AKT3 mutations with several phenotypes dependent on the type of mutation and level of mosaicism. Our comprehensive clinical characterization, and review of all previously published patients, broadly segregates individuals with AKT3 mutations into two groups: patients with highly asymmetric cortical dysplasia caused by the common p.E17K mutation, and patients with constitutional AKT3 mutations exhibiting more variable phenotypes including bilateral cortical malformations, polymicrogyria, periventricular nodular heterotopia and diffuse megalencephaly without cortical dysplasia. All mutations increased kinase activity, and pleckstrin homology domain mutants exhibited enhanced phospholipid binding. Overall, our study shows that activating mutations of the critical AKT3 gene are associated with a wide spectrum of brain involvement ranging from focal or segmental brain malformations (such as hemimegalencephaly and polymicrogyria) predominantly due to mosaic AKT3 mutations, to diffuse bilateral cortical malformations, megalencephaly and heterotopia due to constitutional AKT3 mutations. We also provide the first detailed neuropathological examination of a child with extreme megalencephaly due to a constitutional AKT3 mutation. This child has one of the largest documented paediatric brain sizes, to our knowledge. Finally, our data show that constitutional AKT3 mutations are associated with megalencephaly, with or without autism, similar to PTEN-related disorders. Recognition of this broad clinical and molecular spectrum of AKT3 mutations is important for providing early diagnosis and appropriate management of affected individuals, and will facilitate targeted design of future human clinical trials using PI3K-AKT pathway inhibitors. |
| AKT3, EN2, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23885228  Title: Towards identification of individual etiologies by resolving genomic and biological conundrums in patients with autism spectrum disorders.  Recent genomic research into autism spectrum disorders (ASD) has revealed a remarkably complex genetic architecture. Large numbers of common variants, copy number variations and single nucleotide variants have been identified, yet each of them individually afforded only a small phenotypic impact. A polygenic model in which multiple genes interact either in an additive or a synergistic way appears the most plausible for the majority of ASD patients. Based on recently identified ASD candidate genes, transgenic mouse models for neuroligins/neurorexins and genes such as Cntnap2, Cntn5, Tsc1, Tsc2, Akt3, Cyfip1, Scn1a, En2, Slc6a4, and Bckdk have been generated and studied with respect to behavioral and neuroanatomical phenotypes and sensitivity to drug treatments. From these models, a few clues for potential pharmacologic intervention emerged. The Fmr1, Shank2 and Cntn5 knockout mice exhibited alterations of glutamate receptors, which may become a target for pharmacologic modulation. Some of the phenotypes of Mecp2 knockout mice can be ameliorated by administering IGF1. In the near future, comprehensive genotyping of individual patients and siblings combined with the novel insights generated from the transgenic animal studies may provide us with personalized treatment options. Eventually, autism may indeed turn out to be a phenotypically heterogeneous group of disorders ('autisms') caused by combinations of changes in multiple possible candidate genes, being different in each patient and requiring for each combination of mutations a distinct, individually tailored treatment. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/33248253  Title: Characterization of mice bearing humanized androgen receptor genes (h/mAr) varying in polymorphism length.  The androgen receptor (AR) is known for masculinization of behavior and brain. To better understand the role that AR plays, mice bearing humanized Ar genes with varying lengths of a polymorphic N-terminal glutamine (Q) tract were created (Albertelli et al., 2006). The length of the Q tract is inversely proporitional to AR activity. Biological studies of the Q tract length may also provide a window into potential AR contributions to sex-biases in disease risk. Here we take a multi-pronged approach to characterizing AR signaling effects on brain and behavior in mice using the humanized Ar Q tract model. We first map effects of Q tract length on regional brain anatomy, and consider if these are modified by gonadal sex. We then test the notion that spatial patterns of anatomical variation related to Q tract length could be organized by intrinsic spatiotemporal patterning of AR gene expression in the mouse brain. Finally, we test influences of Q tract length on four behavioral tests.Altering Q tract length led to neuroanatomical differences in a non-linear dosage-dependent fashion. Gene expression analyses indicated that adult neu- roanatomical changes due to Q tract length are only associated with neurode- velopment (as opposed to adulthood). No significant effect of Q tract length was found on the behavior of the three mouse models. These results indicate that AR activity differentially mediates neuroanatomy and behavior, that AR activity alone does not mediate sex differences, and that neurodevelopmen- tal processes are associated with spatial patterns of volume changes due to Q tract length in adulthood. They also indicate that androgen sensitivity in adulthood is not likely to lead to autism-related behaviors or neuroanatomy, although neurodevelopmental processes may play a role earlier. Further study into sex differences, development, other behaviors, and other sex-specific mech- anisms are needed to better understand AR sensitivity, neurodevelopmental disorders, and the sex difference in their prevalence. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/30319350  Title: Sex Hormones Regulate SHANK Expression.  Autism spectrum disorders (ASD) have a higher prevalence in male individuals compared to females, with a ratio of affected boys compared to girls of 4:1 for ASD and 11:1 for Asperger syndrome. Mutations in the SHANK genes (comprising SHANK1, SHANK2 and SHANK3) coding for postsynaptic scaffolding proteins have been tightly associated with ASD. As early brain development is strongly influenced by sex hormones, we investigated the effect of dihydrotestosterone (DHT) and 17β-estradiol on SHANK expression in a human neuroblastoma cell model. Both sex hormones had a significant impact on the expression of all three SHANK genes, which could be effectively blocked by androgen and estrogen receptor antagonists. In neuron-specific androgen receptor knock-out mice (Ar NesCre), we found a nominal significant reduction of all Shank genes at postnatal day 7.5 in the cortex. In the developing cortex of wild-type (WT) CD1 mice, a sex-differential protein expression was identified for all Shanks at embryonic day 17.5 and postnatal day 7.5 with significantly higher protein levels in male compared to female mice. Together, we could show that SHANK expression is influenced by sex hormones leading to a sex-differential expression, thus providing novel insights into the sex bias in ASD. |
| Androgen Receptor, MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/30104728  Title: Sex-specific impact of prenatal androgens on social brain default mode subsystems.  Early-onset neurodevelopmental conditions (e.g., autism) affect males more frequently than females. Androgens may play a role in this male-bias by sex-differentially impacting early prenatal brain development, particularly neural circuits that later develop specialized roles in social cognition. Here, we find that increasing prenatal testosterone in humans is associated with later reduction of functional connectivity between social brain default mode (DMN) subsystems in adolescent males, but has no effect in females. Since testosterone can work directly via the androgen receptor (AR) or indirectly via the estrogen receptor through aromatase conversion to estradiol, we further examined how a potent non-aromatizable androgen, dihydrotestosterone (DHT), acts via the AR to influence gene expression in human neural stem cells (hNSC)-particularly for genes of high-relevance for DMN circuitry. DHT dysregulates a number of genes enriched for syndromic causes of autism and intellectual disability and for genes that in later development are expressed in anatomical patterns that highly correspond to the cortical midline DMN subsystem. DMN-related and DHT-affected genes (e.g., MEF2C) are involved in a number of synaptic processes, many of which impact excitation-inhibition balance. Androgens have male-specific prenatal influence over social brain circuitry in humans and may be relevant towards explaining some component of male-bias in early-onset neurodevelopmental conditions. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/29428674  Title: Genes and Pathways Regulated by Androgens in Human Neural Cells, Potential Candidates for the Male Excess in Autism Spectrum Disorder.  Prenatal exposure to androgens during brain development in male individuals may participate to increase their susceptibility to develop neurodevelopmental disorders such as autism spectrum disorder (ASD) and intellectual disability. However, little is known about the action of androgens in human neural cells. We used human neural stem cells differentiated from embryonic stem cells to investigate targets of androgens. RNA sequencing revealed that treatment with dihydrotestosterone (DHT) leads to subtle but significant changes in the expression of about 200 genes, encoding proteins of extracellular matrix or involved in signal transduction of growth factors (e.g., insulin/insulin growth factor 1). We showed that the most differentially expressed genes (DEGs), RGCC, RNF144B, NRCAM, TRIM22, FAM107A, IGFBP5, and LAMA2, are reproducibly regulated by different androgens in different genetic backgrounds. We showed, by overexpressing the androgen receptor in neuroblastoma cells SH-SY5Y or knocking it down in human neural stem cells, that this regulation involves the androgen receptor. A chromatin immunoprecipitation combined with direct sequencing analysis identified androgen receptor-bound sequences in nearly half of the DHT-DEGs and in numerous other genes. DHT-DEGs appear enriched in genes involved in ASD (ASXL3, NLGN4X, etc.), associated with ASD (NRCAM), or differentially expressed in patients with ASD (FAM107A, IGFBP5). Androgens increase human neural stem cell proliferation and survival in nutrient-deprived culture conditions, with no detectable effect on regulation of neurite outgrowth. We characterized androgen action in neural progenitor cells, identifying DHT-DEGs that appear to be enriched in genes related to ASD. We also showed that androgens increase proliferation of neuronal precursors and protect them from death during their differentiation in nutrient-deprived conditions. |
| Androgen Receptor, FOXP1, FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28204507  Title: Foxp1 expression is essential for sex-specific murine neonatal ultrasonic vocalization.  Autism and speech and language deficits are predominantly found in boys, however the causative mechanisms for this sex bias are unknown. Human FOXP1 is associated with autism, intellectual disability and speech and language deficits. Its closely related family member FOXP2 is involved in speech and language disorder and Foxp2 deficient mice have demonstrated an absence of ultrasonic vocalizations (USVs). Since Foxp1 and Foxp2 form heterodimers for transcriptional regulation, we investigated USV in neonatal brain-specific Foxp1 KO mice. Foxp1 KO pups had strongly reduced USV and lacked the sex-specific call rate from WT pups, indicating that Foxp1 is essential for normal USV. As expression differences of Foxp1 or Foxp2 could explain the sex-dimorphic vocalization in WT animals, we quantified both proteins in the striatum and cortex at P7.5 and detected a sex-specific expression of Foxp2 in the striatum. We further analyzed Foxp1 and Foxp2 expression in the striatum and cortex of CD1 mice at different embryonic and postnatal stages and observed sex differences in both genes at E17.5 and P7.5. Sex hormones, especially androgens are known to play a crucial role in the sexual differentiation of vocalizations in many vertebrates. We show that Foxp1 and the androgen receptor are co-expressed in striatal medium spiny neurons and that brain-specific androgen receptor KO (ArNesCre) mice exhibit reduced Foxp1 expression in the striatum at E17.5 and P7.5 and an increased Foxp2 level in the cortex at P7.5. Thus, androgens may contribute to sex-specific differences in Foxp1 and Foxp2 expression and USV. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/27423376  Title: Prenatal exposure to sodium valproate alters androgen receptor expression in the developing cerebellum in a region and age specific manner in male and female rats.  Valproic acid (VPA) is an anti-epileptic drug with teratogenicity activity that has been related to autism. In rodents, exposure to VPA in utero leads to brain abnormalities similar than those reported in the autistic brain. Particularly, VPA reduces the number of Purkinje neurons in the rat cerebellum parallel to cerebellar abnormalities found in autism. Thus, we injected pregnant females on embryonic day 12 either with VPA (600mg/kg, i.p.) or 0.9% saline solution and obtained the cerebellum from their offspring at different postnatal time points. Testosterone has been linked to autism and plays an important role during brain development. Therefore, we identified and analyzed the androgen receptor (AR) by immunohistochemistry and densitometry, respectively. We found VPA decreases AR density in the superficial Purkinje layer only in cerebellar lobule 8 at PN7, but increased it at PN14 compared to control in males. In females, VPA decreased AR density in the superficial Purkinje layer in cerebellar lobule 6 at PN14, but increased it in lobule 9 at the same time point. No differences were found in the deep Purkinje layer of any cerebellar lobule in terms of AR density neither in males nor females. We additionally found a particular AR density decreasing in both superficial and deep regions across development in the majority of cerebellar lobules in males, but in all cerebellar lobules in females. Thus, our results indicate that VPA disrupts the AR ontogeny in the developing cerebellum in an age and region specific manner in male and female rats. Future epigenetic studies including the evaluation of histone deacetylases (HDAC's) might shed light these results as HDAC's are expressed by Purkinje neurons, interact with the AR and are VPA targets. This work contributes to the understanding of the cerebellar development and it might help to understand the role of the cerebellum in neurodevelopmental disorders such as autism. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/26781567  Title: Developmental neurogenetics and multimodal neuroimaging of sex differences in autism.  Examining sex differences in the brain has been historically contentious but is nonetheless important for advancing mental health for both girls and boys. Unfortunately, females in biomedical research remain underrepresented in most mental health conditions including autism spectrum disorders (ASD), even though equal inclusion of females would improve treatment for girls and yield benefits to boys. This review examines sex differences in the relationship between neuroanatomy and neurogenetics of ASD. Recent findings reveal that girls diagnosed with ASD exhibit more intellectual and behavioral problems compared to their male counterparts, suggesting that girls may be less likely diagnosed in the absence of such problems or that they require a higher mutational load to meet the diagnostic criteria. Thus far, the female biased effect of chromosome 4, 5p15.33, 8p, 9p24.1, 11p12-13, 15q, and Xp22.3 and the male biased effect of 1p31.3, 5q12.3, 7q, 9q33.3, 11q13.4, 13q33.3, 16p11.2, 17q11-21, Xp22.33/Yp11.31, DRD1, NLGN3, MAOA, and SHANK1 deletion have been discovered in ASD. The SNPs of genes such as RYR2, UPP2, and the androgen receptor gene have been shown to have sex-biasing factors in both girls and boys diagnosed with ASD. These sex-related genetic factors may drive sex differences in the neuroanatomy of these girls and boys, including abnormal enlargement in temporal gray and white matter volumes, and atypical reduction in cerebellar gray matter volumes and corpus callosum fibers projecting to the anterior frontal cortex in ASD girls relative to boys. Such factors may also be responsible for the attenuation of brain sexual differentiation in adult men and women with ASD; however, much remains to be uncovered or replicated. Future research should leverage further the association between neuroanatomy and genetics in girls for an integrated and interdisciplinary understanding of ASD. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/24865607  Title: Neuropsychology and brain morphology in Klinefelter syndrome - the impact of genetics.  Klinefelter syndrome (KS, 47,XXY) is associated with increased psychiatric morbidity and cognitive disabilities, although the neuropsychological phenotype shows great variability. Androgen receptor polymorphism (CAG repeat length), skewed X-chromosome inactivation and parent-of-origin of the extra X-chromosome have been suggested to influence cognitive function and psychological traits. These issues have not been clarified for KS patients. We studied X-chromosome inactivation pattern, CAG repeat length and parent-of-origin in relation to educational and cohabitation status, personality and autism traits, psychological distress, cognitive function and brain volumes in 73 KS patients and 73 controls. Grey matter (GM) volume of left insula was significantly decreased in KS patients with skewed X-inactivation (z = 5.78) and we observed a borderline significant difference in global brain matter volume where KS patients with skewed X-chromosome inactivation tended to have smaller brains. Skewed X-inactivation, CAG repeat length and parent-of-origin were not correlated with educational and marital status, personality traits, autism traits, and psychological distress, prevalence of depression and anxiety or cognitive function. Interestingly our results regarding brain volumes indicate that X-inactivation has an influence on GM volume in left insula and might also be related to global GM volume, indicating a possible effect of X-linked genes on the development of GM volume in KS patient. Skewed X-inactivation, CAG repeat length and parent-of-origin have no impact on the neuropsychological phenotype in KS (http://www.clinicaltrials.gov (Clinical trial NCT00999310)). |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/24119295  Title: Differential recruitment of coregulators to the RORA promoter adds another layer of complexity to gene (dys) regulation by sex hormones in autism.  Our independent cohort studies have consistently shown the reduction of the nuclear receptor RORA (retinoic acid-related orphan receptor-alpha) in lymphoblasts as well as in brain tissues from individuals with autism spectrum disorder (ASD). Moreover, we have found that RORA regulates the gene for aromatase, which converts androgen to estrogen, and that male and female hormones regulate RORA in opposite directions, with androgen suppressing RORA, suggesting that the sexually dimorphic regulation of RORA may contribute to the male bias in ASD. However, the molecular mechanisms through which androgen and estrogen differentially regulate RORA are still unknown. Here we use functional knockdown of hormone receptors and coregulators with small interfering RNA (siRNA) to investigate their involvement in sex hormone regulation of RORA in human neuronal cells. Luciferase assays using a vector containing various RORA promoter constructs were first performed to identify the promoter regions required for inverse regulation of RORA by male and female hormones. Sequential chromatin immunoprecipitation methods followed by quantitative reverse transcriptase-polymerase chain reaction (qRT-PCR) analyses of RORA expression in hormone-treated SH-SY5Y cells were then utilized to identify coregulators that associate with hormone receptors on the RORA promoter. siRNA-mediated knockdown of interacting coregulators was performed followed by qRT-PCR analyses to confirm the functional requirement of each coregulator in hormone-regulated RORA expression. Our studies demonstrate the direct involvement of androgen receptor (AR) and estrogen receptor (ER) in the regulation of RORA by male and female hormones, respectively, and that the promoter region between -10055 bp and -2344 bp from the transcription start site of RORA is required for the inverse hormonal regulation. We further show that AR interacts with SUMO1, a reported suppressor of AR transcriptional activity, whereas ERα interacts with the coactivator NCOA5 on the RORA promoter. siRNA-mediated knockdown of SUMO1 and NCOA5 attenuate the sex hormone effects on RORA expression. AR and SUMO1 are involved in the suppression RORA expression by androgen, while ERα and NCOA5 collaborate in the up-regulation of RORA by estrogen. While this study offers a better understanding of molecular mechanisms involved in sex hormone regulation of RORA, it also reveals another layer of complexity with regard to gene regulation in ASD. Inasmuch as coregulators are capable of interacting with a multitude of transcription factors, aberrant expression of coregulator proteins, as we have seen previously in lymphoblasts from individuals with ASD, may contribute to the polygenic nature of gene dysregulation in ASD. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/23801657  Title: Stem cells as a good tool to investigate dysregulated biological systems in autism spectrum disorders.  Identification of the causes of autism spectrum disorders (ASDs) is hampered by their genetic heterogeneity; however, the different genetic alterations leading to ASD seem to be implicated in the disturbance of common molecular pathways or biological processes. In this scenario, the search for differentially expressed genes (DEGs) between ASD patients and controls is a good alternative to identify the molecular etiology of such disorders. Here, we employed genome-wide expression analysis to compare the transcriptome of stem cells of human exfoliated deciduous teeth (SHEDs) of idiopathic autistic patients (n = 7) and control samples (n = 6). Nearly half of the 683 identified DEGs are expressed in the brain (P = 0.003), and a significant number of them are involved in mechanisms previously associated with ASD such as protein synthesis, cytoskeleton regulation, cellular adhesion and alternative splicing, which validate the use of SHEDs to disentangle the causes of autism. Autistic patients also presented overexpression of genes regulated by androgen receptor (AR), and AR itself, which in turn interacts with CHD8 (chromodomain helicase DNA binding protein 8), a gene recently shown to be associated with the cause of autism and found to be upregulated in some patients tested here. These data provide a rationale for the mechanisms through which CHD8 leads to these diseases. In summary, our results suggest that ASD share deregulated pathways and revealed that SHEDs represent an alternative cell source to be used in the understanding of the biological mechanisms involved in the etiology of ASD. |
| Androgen Receptor |
| Url: https://pubmed.ncbi.nlm.nih.gov/19167832  Title: Possible association between the androgen receptor gene and autism spectrum disorder.  Autism is a highly heritable disorder but the specific genes involved remain largely unknown. The higher prevalence of autism in men than in women, in conjunction with a number of other observations, has led to the suggestion that prenatal brain exposure to androgens may be of importance for the development of this condition. Prompted by this hypothesis, we investigated the potential influence of variation in the androgen receptor (AR) gene on the susceptibility for autism. To this end, 267 subjects with autism spectrum disorder and 617 controls were genotyped for three polymorphisms in exon 1 of the AR gene: the CAG repeat, the GGN repeat and the rs6152 SNP. In addition, parents and affected siblings were genotyped for 118 and 32 of the cases, respectively. Case-control comparisons revealed higher prevalence of short CAG alleles as well as of the A allele of the rs6152 SNP in female cases than in controls, but revealed no significant differences with respect to the GGN repeat. Analysis of the 118 families using transmission disequilibrium test, on the other hand, suggested an association with the GGN polymorphism, the rare 20-repeat allele being undertransmitted to male cases and the 23-repeat allele being overtransmitted to female cases. Sequencing of the AR gene in 46 patients revealed no mutations or rare variants. The results lend some support for an influence of the studied polymorphisms on the susceptibility for autism, but argue against the possibility that mutations in the AR gene are common in subjects with this condition. |
| Androgen Receptor, CTCF, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19132145  Title: MECP2 promoter methylation and X chromosome inactivation in autism.  Epigenetic mechanisms have been proposed to play a role in the etiology of autism. This hypothesis is supported by the discovery of increased MECP2 promoter methylation associated with decreased MeCP2 protein expression in autism male brain. To further understand the influence of female X chromosome inactivation (XCI) and neighboring methylation patterns on aberrant MECP2 promoter methylation in autism, multiple methylation analyses were peformed on brain and blood samples from individuals with autism. Bisulfite sequencing analyses of a region 0.6 kb upstream of MECP2 in brain DNA samples revealed an abrupt transition from a highly methylated region in both sexes to a region unmethylated in males and subject to XCI in females. Chromatin immunoprecipitation analysis demonstrated that the CCTC-binding factor (CTCF) bound to this transition region in neuronal cells, consistent with a chromatin boundary at the methylation transition. Male autism brain DNA samples displayed a slight increase in methylation in this transition region, suggesting a possible aberrant spreading of methylation into the MECP2 promoter in autism males across this boundary element. In addition, autistic female brain DNA samples showed evidence for aberrant MECP2 promoter methylation as an increase in the number of bisulfite sequenced clones with undefined XCI status for MECP2 but not androgen receptor (AR). To further investigate the specificity of MECP2 methylation alterations in autism, blood DNA samples from females and mothers of males with autism were also examined for XCI skewing at AR, but no significant increase in XCI skewing was observed compared to controls. These results suggest that the aberrant MECP2 methylation in autism brain DNA samples is due to locus-specific rather than global X chromosome methylation changes. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/36743289  Title: Dendritic spine and synapse pathology in chromatin modifier-associated autism spectrum disorders and intellectual disability.  Formation of dendritic spine and synapse is an essential final step of brain wiring to establish functional communication in the developing brain. Recent findings have displayed altered dendritic spine and synapse morphogenesis, plasticity, and related molecular mechanisms in animal models and post-mortem human brains of autism spectrum disorders (ASD) and intellectual disability (ID). Many genes and proteins are shown to be associated with spines and synapse development, and therefore neurodevelopmental disorders. In this review, however, particular attention will be given to chromatin modifiers such as AT-Rich Interactive Domain 1B (ARID1B), KAT8 regulatory non-specific lethal (NSL) complex subunit 1 (KANSL1), and WD Repeat Domain 5 (WDR5) which are among strong susceptibility factors for ASD and ID. Emerging evidence highlights the critical status of these chromatin remodeling molecules in dendritic spine morphogenesis and synaptic functions. Molecular and cellular insights of ARID1B, KANSL1, and WDR5 will integrate into our current knowledge in understanding and interpreting the pathogenesis of ASD and ID. Modulation of their activities or levels may be an option for potential therapeutic treatment strategies for these neurodevelopmental conditions. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/35445787  Title: Genetic mouse models of autism spectrum disorder present subtle heterogenous cardiac abnormalities.  Autism spectrum disorder (ASD) and congenital heart disease (CHD) are linked on a functional and genetic level. Most work has investigated CHD-related neurodevelopmental abnormalities. Cardiac abnormalities in ASD have been less studied. We investigated the prevalence of cardiac comorbidities relative to ASD genetic contributors. Using high frequency ultrasound imaging, we screened 9 ASD-related genetic mouse models (Arid1b(+/-) , Chd8(+/-) , 16p11.2 (deletion), Sgsh(+/-) , Sgsh(-/-) , Shank3 Δexon 4-9(+/-) , Shank3 Δexon 4-9(-/-) , Fmr1(-/-) , Vps13b(+/-) ), and pooled wild-type littermates (WTs). We measured heart rate (HR), aorta diameter (AoD), thickness and thickening of the left-ventricular (LV) anterior and posterior walls, LV chamber diameter, fractional shortening, stroke volume and cardiac output, mitral inflow Peak E and A velocity ratio, ascending aorta velocity time integral (VTI). Mutant groups presented small-scale alterations in cardiac structure and function compared to WTs (LV anterior wall thickness and thickening, chamber diameter and fractional shortening, HR). A greater number of significant differences was observed among mutant groups than between mutant groups and WTs. Mutant groups differed primarily in structural measures (LV chamber diameter and anterior wall thickness, HR, AoD). The mutant groups with most differences to WTs were 16p11.2 (deletion), Fmr1(-/-) , Arid1b(+/-) . The mutant groups with most differences from other mutant groups were 16p11.2 (deletion), Sgsh(+/-) , Fmr1(-/-) . Our results recapitulate the associated clinical findings. The characteristic ASD heterogeneity was recapitulated in the cardiac phenotype. The type of abnormal measures (morphological, functional) can highlight common underlying mechanisms. Clinically, knowledge of cardiac abnormalities in ASD can be essential as even non-lethal abnormalities impact normal development. LAY SUMMARY: Autism spectrum disorder (ASD) and congenital heart disease (CHD) are linked functionally and genetically. ASD cardiac phenotyping is limited. We assessed the cardiac phenotype of 9 ASD-related mouse models. We found subtle heterogenous cardiac abnormalities compared to controls, with more differences within ASD than between ASD and controls, mirroring clinical findings. Clinically, knowing the cardiac abnormalities in ASD is vital as even non-lethal cardiac abnormalities can impact development. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/35110736  Title: Autism genes converge on asynchronous development of shared neuron classes.  Genetic risk for autism spectrum disorder (ASD) is associated with hundreds of genes spanning a wide range of biological functions1-6. The alterations in the human brain resulting from mutations in these genes remain unclear. Furthermore, their phenotypic manifestation varies across individuals7,8. Here we used organoid models of the human cerebral cortex to identify cell-type-specific developmental abnormalities that result from haploinsufficiency in three ASD risk genes-SUV420H1 (also known as KMT5B), ARID1B and CHD8-in multiple cell lines from different donors, using single-cell RNA-sequencing (scRNA-seq) analysis of more than 745,000 cells and proteomic analysis of individual organoids, to identify phenotypic convergence. Each of the three mutations confers asynchronous development of two main cortical neuronal lineages-γ-aminobutyric-acid-releasing (GABAergic) neurons and deep-layer excitatory projection neurons-but acts through largely distinct molecular pathways. Although these phenotypes are consistent across cell lines, their expressivity is influenced by the individual genomic context, in a manner that is dependent on both the risk gene and the developmental defect. Calcium imaging in intact organoids shows that these early-stage developmental changes are followed by abnormal circuit activity. This research uncovers cell-type-specific neurodevelopmental abnormalities that are shared across ASD risk genes and are finely modulated by human genomic context, finding convergence in the neurobiological basis of how different risk genes contribute to ASD pathology. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/34426966  Title: A systematic review of brain MRI findings in monogenic disorders strongly associated with autism spectrum disorder.  Research on monogenic forms of autism spectrum disorder (autism) can inform our understanding of genetic contributions to the autism phenotype; yet, there is much to be learned about the pathways from gene to brain structure to behavior. This systematic review summarizes and evaluates research on brain magnetic resonance imaging (MRI) findings in monogenic conditions that have strong association with autism. This will improve understanding of the impact of genetic variability on brain structure and related behavioral traits in autism. The search strategy for this systematic review followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines. Risk of bias (ROB) assessment was completed on included studies using the Newcastle-Ottawa Scales. Of 4,287 studies screened, 69 were included pertaining to 13 of the top 20 genes with the strongest association with autism. The greatest number of studies related to individuals with PTEN variants and autism. Brain MRI abnormalities were reported for 12 of the 13 genes studied, and in 51.7% of participants across all 13 genes, including 100% of participants with ARID1B variants. Specific MRI findings were highly variable, with no clear patterns emerging within or between the 13 genes, although white matter abnormalities were the most common. Few studies reported specific details about methods for acquisition and processing of brain MRI, and descriptors for brain abnormalities were variable. ROB assessment indicated high ROB for all studies, largely due to small sample sizes and lack of comparison groups. Brain abnormalities are common in this population of individuals, in particular, children; however, a range of different brain abnormalities were reported within and between genes. Directions for future neuroimaging research in monogenic autism are suggested. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/33982067  Title: Exposure to sevoflurane results in changes of transcription factor occupancy in sperm and inheritance of autism†.  One in 54 children in the United States is diagnosed with autism spectrum disorder. De novo germline and somatic mutations cannot account for all cases of autism spectrum disorder, suggesting that epigenetic alterations triggered by environmental exposures may be responsible for a subset of autism spectrum disorder cases. Human and animal studies have shown that exposure of the developing brain to general anesthetic agents can trigger neurodegeneration and neurobehavioral abnormalities, but the effects of general anesthetics on the germline have not been explored in detail. We exposed pregnant mice to sevoflurane during the time of embryonic development when the germ cells undergo epigenetic reprogramming and found that more than 38% of the directly exposed F1 animals exhibit impairments in anxiety and social interactions. Strikingly, 44-47% of the F2 and F3 animals, which were not directly exposed to sevoflurane, show the same behavioral problems. We performed ATAC-seq and identified more than 1200 differentially accessible sites in the sperm of F1 animals, 69 of which are also present in the sperm of F2 animals. These sites are located in regulatory regions of genes strongly associated with autism spectrum disorder, including Arid1b, Ntrk2, and Stmn2. These findings suggest that epimutations caused by exposing germ cells to sevoflurane can lead to autism spectrum disorder in the offspring, and this effect can be transmitted through the male germline inter- and transgenerationally. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/33757588  Title: Neuroanatomy and behavior in mice with a haploinsufficiency of AT-rich interactive domain 1B (ARID1B) throughout development.  One of the causal mechanisms underlying neurodevelopmental disorders (NDDs) is chromatin modification and the genes that regulate chromatin. AT-rich interactive domain 1B (ARID1B), a chromatin modifier, has been linked to autism spectrum disorder and to affect rare and inherited genetic variation in a broad set of NDDs. A novel preclinical mouse model of Arid1b deficiency was created and validated to characterize and define neuroanatomical, behavioral and transcriptional phenotypes. Neuroanatomy was assessed ex vivo in adult animals and in vivo longitudinally from birth to adulthood. Behavioral testing was also performed throughout development and tested all aspects of motor, learning, sociability, repetitive behaviors, seizure susceptibility, and general milestones delays. We validated decreased Arid1b mRNA and protein in Arid1b+/- mice, with signatures of increased axonal and synaptic gene expression, decreased transcriptional regulator and RNA processing expression in adult Arid1b+/- cerebellum. During neonatal development, Arid1b+/- mice exhibited robust impairments in ultrasonic vocalizations (USVs) and metrics of developmental growth. In addition, a striking sex effect was observed neuroanatomically throughout development. Behaviorally, as adults, Arid1b+/- mice showed low motor skills in open field exploration and normal three-chambered approach. Arid1b+/- mice had learning and memory deficits in novel object recognition but not in visual discrimination and reversal touchscreen tasks. Social interactions in the male-female social dyad with USVs revealed social deficits on some but not all parameters. No repetitive behaviors were observed. Brains of adult Arid1b+/- mice had a smaller cerebellum and a larger hippocampus and corpus callosum. The corpus callosum increase seen here contrasts previous reports which highlight losses in corpus callosum volume in mice and humans. The behavior and neuroimaging analyses were done on separate cohorts of mice, which did not allow a direct correlation between the imaging and behavioral findings, and the transcriptomic analysis was exploratory, with no validation of altered expression beyond Arid1b. This study represents a full validation and investigation of a novel model of Arid1b+/- haploinsufficiency throughout development and highlights the importance of examining both sexes throughout development in NDDs. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/33686214  Title: Neurobiology of ARID1B haploinsufficiency related to neurodevelopmental and psychiatric disorders.  ARID1B haploinsufficiency is a frequent cause of intellectual disability (ID) and autism spectrum disorder (ASD), and also leads to emotional disturbances. In this review, we examine past and present clinical and preclinical research into the neurobiological function of ARID1B. The presentation of ARID1B-related disorders (ARID1B-RD) is highly heterogeneous, including varying degrees of ID, ASD, and physical features. Recent research includes the development of suitable clinical readiness assessments for the treatment of ARID1B-RD, as well as similar neurodevelopmental disorders. Recently developed mouse models of Arid1b haploinsufficiency successfully mirror many of the behavioral phenotypes of ASD and ID. These animal models have helped to solidify the molecular mechanisms by which ARID1B regulates brain development and function, including epigenetic regulation of the Pvalb gene and promotion of Wnt/β-catenin signaling in neural progenitors in the ventral telencephalon. Finally, preclinical studies have identified the use of a positive allosteric modulator of the GABAA receptor as an effective treatment for some Arid1b haploinsufficiency-related behavioral phenotypes, and there is potential for the refinement of this therapy in order to translate it into clinical use. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/33594090  Title: Differential roles of ARID1B in excitatory and inhibitory neural progenitors in the developing cortex.  Genetic evidence indicates that haploinsufficiency of ARID1B causes intellectual disability (ID) and autism spectrum disorder (ASD), but the neural function of ARID1B is largely unknown. Using both conditional and global Arid1b knockout mouse strains, we examined the role of ARID1B in neural progenitors. We detected an overall decrease in the proliferation of cortical and ventral neural progenitors following homozygous deletion of Arid1b, as well as altered cell cycle regulation and increased cell death. Each of these phenotypes was more pronounced in ventral neural progenitors. Furthermore, we observed decreased nuclear localization of β-catenin in Arid1b-deficient neurons. Conditional homozygous deletion of Arid1b in ventral neural progenitors led to pronounced ID- and ASD-like behaviors in mice, whereas the deletion in cortical neural progenitors resulted in minor cognitive deficits. This study suggests an essential role for ARID1B in forebrain neurogenesis and clarifies its more pronounced role in inhibitory neural progenitors. Our findings also provide insights into the pathogenesis of ID and ASD. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/33262327  Title: Autism-associated miR-873 regulates ARID1B, SHANK3 and NRXN2 involved in neurodevelopment.  Autism spectrum disorders (ASD) are highly heritable neurodevelopmental disorders with significant genetic heterogeneity. Noncoding microRNAs (miRNAs) are recognised as playing key roles in development of ASD albeit the function of these regulatory genes remains unclear. We previously conducted whole-exome sequencing of Australian families with ASD and identified four novel single nucleotide variations in mature miRNA sequences. A pull-down transcriptome analysis using transfected SH-SY5Y cells proposed a mechanistic model to examine changes in binding affinity associated with a unique mutation found in the conserved 'seed' region of miR-873-5p (rs777143952: T > A). Results suggested several ASD-risk genes were differentially targeted by wild-type and mutant miR-873 variants. In the current study, a dual-luciferase reporter assay confirmed miR-873 variants have a 20-30% inhibition/dysregulation effect on candidate autism risk genes ARID1B, SHANK3 and NRXN2 and also confirmed the affected expression with qPCR. In vitro mouse hippocampal neurons transfected with mutant miR-873 showed less morphological complexity and enhanced sodium currents and excitatory neurotransmission compared to cells transfected with wild-type miR-873. A second in vitro study showed CRISPR/Cas9 miR-873 disrupted SH-SY5Y neuroblastoma cells acquired a neuronal-like morphology and increased expression of ASD important genes ARID1B, SHANK3, ADNP2, ANK2 and CHD8. These results represent the first functional evidence that miR-873 regulates key neural genes involved in development and cell differentiation. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/32398858  Title: Arid1b haploinsufficiency in parvalbumin- or somatostatin-expressing interneurons leads to distinct ASD-like and ID-like behavior.  Inhibitory interneurons are essential for proper brain development and function. Dysfunction of interneurons is implicated in several neurodevelopmental disorders, including autism spectrum disorder (ASD) and intellectual disability (ID). We have previously shown that Arid1b haploinsufficiency interferes with interneuron development and leads to social, cognitive, and emotional impairments consistent with ASD and ID. It is unclear, however, whether interneurons play a major role for the behavioral deficits in Arid1b haploinsufficiency. Furthermore, it is critical to determine which interneuron subtypes contribute to distinct behavioral phenotypes. In the present study, we generated Arid1b haploinsufficient mice in which a copy of the Arid1b gene is deleted in either parvalbumin (PV) or somatostatin (SST) interneurons, and examined their ASD- and ID-like behaviors. We found that Arid1b haploinsufficiency in PV or SST interneurons resulted in distinct features that do not overlap with one another. Arid1b haploinsufficiency in PV neurons contributed to social and emotional impairments, while the gene deletion in the SST population caused stereotypies as well as learning and memory dysfunction. These findings demonstrate a critical role of interneurons in Arid1b haploinsufficient pathology and suggest that PV and SST interneurons may have distinct roles in modulating neurological phenotypes in Arid1b haploinsufficiency-induced ASD and ID. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/30933046  Title: Corpus callosum metrics predict severity of visuospatial and neuromotor dysfunctions in ARID1B mutations with Coffin-Siris syndrome.  ARID1B mutations in Coffin-Siris syndrome are a cause of intellectual disability (0.5-1%), with various degrees of autism and agenesis of the corpus callosum (10%). Little is known regarding the cognitive and motor consequences of ARID1B mutations in humans and no link has been made between corpus callosum anomalies and visuospatial and neuromotor dysfunctions. We have investigated the visuospatial and neuromotor phenotype in eight patients with ARID1B mutations. A paramedian sagittal section of the brain MRI was selected, and corpus callosum was measured in anteroposterior length, genu and trunk width. Spearman's rank order coefficients were used to explore correlations between visuospatial and social cognitive variables and dimensions of the corpus callosum. A significant correlation between genu width size and visual cognition was observed. Retrocerebellar cysts were associated with corpus callosum anomalies. Here, we show that corpus callosum anomalies caused in ARID1B mutations may be predictive of the visuospatial and motor phenotype in Coffin-Siris syndrome. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/30149092  Title: The role of ARID1B, a BAF chromatin remodeling complex subunit, in neural development and behavior.  Haploinsufficiency of the chromatin remodeling factor ARID1B leads to autism spectrum disorder and intellectual disability. Several independent research groups, including our own, recently examined the effects of heterozygous deletion of Arid1b in mice and reported severe behavioral abnormalities reminiscent of autism spectrum disorders and intellectual disability as well as marked changes in gene expression and decreased body size. Arid1b heterozygous mice also display significant cortical excitatory/inhibitory imbalance due to altered GABAergic neuron numbers and impaired inhibitory synaptic transmission. Abnormal epigenetic modifications, including histone acetylation and methylation, are additionally associated with Arid1b haploinsufficiency in the brain. Treating adult Arid1b mutant mice with a positive GABA allosteric modulator, however, rescues multiple behavioral abnormalities, such as cognitive and social impairments, as well as elevated anxiety. While treating Arid1b haploinsufficient mice with recombinant mouse growth hormone successfully increases body size, it has no effect on aberrant behavior. Here we summarize the recent findings regarding the role of ARID1B in brain development and behavior and discuss the utility of the Arid1b heterozygous mouse model in neurodevelopmental and psychiatric research. We also discuss some of the opportunities and potential challenges in developing translational applications for humans and possible avenues for further research into the mechanisms of ARID1B pathology in the brain. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/29184203  Title: Arid1b haploinsufficiency disrupts cortical interneuron development and mouse behavior.  Haploinsufficiency of the AT-rich interactive domain 1B (ARID1B) gene causes autism spectrum disorder and intellectual disability; however, the neurobiological basis for this is unknown. Here we generated Arid1b-knockout mice and examined heterozygotes to model human patients. Arid1b-heterozygous mice showed a decreased number of cortical GABAergic interneurons and reduced proliferation of interneuron progenitors in the ganglionic eminence. Arid1b haploinsufficiency also led to an imbalance between excitatory and inhibitory synapses in the cerebral cortex. Furthermore, we found that Arid1b haploinsufficiency suppressed histone H3 lysine 9 acetylation (H3K9ac) overall and particularly reduced H3K9ac of the Pvalb promoter, resulting in decreased transcription. Arid1b-heterozygous mice exhibited abnormal cognitive and social behaviors, which were rescued by treatment with a positive allosteric GABAA receptor modulator. Our results demonstrate a critical role for Arid1b in interneuron development and behavior and provide insight into the pathogenesis of autism spectrum disorder and intellectual disability. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/28867767  Title: Arid1b Haploinsufficiency Causes Abnormal Brain Gene Expression and Autism-Related Behaviors in Mice.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder with core symptoms that include poor social communication, restricted interests, and repetitive behaviors. Several ASD mouse models exhibit impaired social interaction, anxiety-like behavior, and elevated perseveration. Large-scale whole exome sequencing studies identified many genes putatively associated with ASD. Like chromodomain helicase DNA binding protein 8 (CHD8), the most frequently mutated gene in individuals with ASD, the candidate gene AT-rich interaction domain 1B (ARID1B) encodes a chromatin remodeling factor. Arid1b heterozygous knockout (hKO) mice exhibited ASD-like traits related to social behavior, anxiety, and perseveration, in addition to associated features reported in some cases of ASD, such as reduced weight, impaired motor coordination, and hydrocephalus. Hydrocephalus was present in 5 of 91 hKO mice, while it was not observed in wild-type littermates (0 of 188). Genome-wide gene expression patterns in Arid1b hKO mice were similar to those in ASD patients and Chd8-haploinsufficient mice, an ASD model, and to developmental changes in gene expression in fast-spiking cells in the mouse brain. Our results suggest that Arid1b haploinsufficiency causes ASD-like phenotypes in mice. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/28695822  Title: Arid1b haploinsufficient mice reveal neuropsychiatric phenotypes and reversible causes of growth impairment.  Sequencing studies have implicated haploinsufficiency of ARID1B, a SWI/SNF chromatin-remodeling subunit, in short stature (Yu et al., 2015), autism spectrum disorder (O'Roak et al., 2012), intellectual disability (Deciphering Developmental Disorders Study, 2015), and corpus callosum agenesis (Halgren et al., 2012). In addition, ARID1B is the most common cause of Coffin-Siris syndrome, a developmental delay syndrome characterized by some of the above abnormalities (Santen et al., 2012; Tsurusaki et al., 2012; Wieczorek et al., 2013). We generated Arid1b heterozygous mice, which showed social behavior impairment, altered vocalization, anxiety-like behavior, neuroanatomical abnormalities, and growth impairment. In the brain, Arid1b haploinsufficiency resulted in changes in the expression of SWI/SNF-regulated genes implicated in neuropsychiatric disorders. A focus on reversible mechanisms identified Insulin-like growth factor (IGF1) deficiency with inadequate compensation by Growth hormone-releasing hormone (GHRH) and Growth hormone (GH), underappreciated findings in ARID1B patients. Therapeutically, GH supplementation was able to correct growth retardation and muscle weakness. This model functionally validates the involvement of ARID1B in human disorders, and allows mechanistic dissection of neurodevelopmental diseases linked to chromatin-remodeling. |
| ARID1B, MECP2, TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28584888  Title: Autism spectrum disorder: neuropathology and animal models.  Autism spectrum disorder (ASD) has a major impact on the development and social integration of affected individuals and is the most heritable of psychiatric disorders. An increase in the incidence of ASD cases has prompted a surge in research efforts on the underlying neuropathologic processes. We present an overview of current findings in neuropathology studies of ASD using two investigational approaches, postmortem human brains and ASD animal models, and discuss the overlap, limitations, and significance of each. Postmortem examination of ASD brains has revealed global changes including disorganized gray and white matter, increased number of neurons, decreased volume of neuronal soma, and increased neuropil, the last reflecting changes in densities of dendritic spines, cerebral vasculature and glia. Both cortical and non-cortical areas show region-specific abnormalities in neuronal morphology and cytoarchitectural organization, with consistent findings reported from the prefrontal cortex, fusiform gyrus, frontoinsular cortex, cingulate cortex, hippocampus, amygdala, cerebellum and brainstem. The paucity of postmortem human studies linking neuropathology to the underlying etiology has been partly addressed using animal models to explore the impact of genetic and non-genetic factors clinically relevant for the ASD phenotype. Genetically modified models include those based on well-studied monogenic ASD genes (NLGN3, NLGN4, NRXN1, CNTNAP2, SHANK3, MECP2, FMR1, TSC1/2), emerging risk genes (CHD8, SCN2A, SYNGAP1, ARID1B, GRIN2B, DSCAM, TBR1), and copy number variants (15q11-q13 deletion, 15q13.3 microdeletion, 15q11-13 duplication, 16p11.2 deletion and duplication, 22q11.2 deletion). Models of idiopathic ASD include inbred rodent strains that mimic ASD behaviors as well as models developed by environmental interventions such as prenatal exposure to sodium valproate, maternal autoantibodies, and maternal immune activation. In addition to replicating some of the neuropathologic features seen in postmortem studies, a common finding in several animal models of ASD is altered density of dendritic spines, with the direction of the change depending on the specific genetic modification, age and brain region. Overall, postmortem neuropathologic studies with larger sample sizes representative of the various ASD risk genes and diverse clinical phenotypes are warranted to clarify putative etiopathogenic pathways further and to promote the emergence of clinically relevant diagnostic and therapeutic tools. In addition, as genetic alterations may render certain individuals more vulnerable to developing the pathological changes at the synapse underlying the behavioral manifestations of ASD, neuropathologic investigation using genetically modified animal models will help to improve our understanding of the disease mechanisms and enhance the development of targeted treatments. |
| ARID1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/21801163  Title: Corpus callosum abnormalities, intellectual disability, speech impairment, and autism in patients with haploinsufficiency of ARID1B.  Corpus callosum abnormalities, intellectual disability, speech impairment, and autism in patients with haploinsufficiency of ARID1B. Corpus callosum abnormalities are common brain malformations with a wide clinical spectrum ranging from severe intellectual disability to normal cognitive function. The etiology is expected to be genetic in as much as 30-50% of the cases, but the underlying genetic cause remains unknown in the majority of cases. By next-generation mate-pair sequencing we mapped the chromosomal breakpoints of a patient with a de novo balanced translocation, t(1;6)(p31;q25), agenesis of corpus callosum (CC), intellectual disability, severe speech impairment, and autism. The chromosome 6 breakpoint truncated ARID1B which was also truncated in a recently published translocation patient with a similar phenotype. Quantitative polymerase chain reaction (Q-PCR) data showed that a primer set proximal to the translocation showed increased expression of ARID1B, whereas primer sets spanning or distal to the translocation showed decreased expression in the patient relative to a non-related control set. Phenotype-genotype comparison of the translocation patient to seven unpublished patients with various sized deletions encompassing ARID1B confirms that haploinsufficiency of ARID1B is associated with CC abnormalities, intellectual disability, severe speech impairment, and autism. Our findings emphasize that ARID1B is important in human brain development and function in general, and in the development of CC and in speech development in particular. |
| ARNT2, ZNF804A |
| Url: https://pubmed.ncbi.nlm.nih.gov/24736721  Title: Heat shock alters the expression of schizophrenia and autism candidate genes in an induced pluripotent stem cell model of the human telencephalon.  Schizophrenia (SZ) and autism spectrum disorders (ASD) are highly heritable neuropsychiatric disorders, although environmental factors, such as maternal immune activation (MIA), play a role as well. Cytokines mediate the effects of MIA on neurogenesis and behavior in animal models. However, MIA stimulators can also induce a febrile reaction, which could have independent effects on neurogenesis through heat shock (HS)-regulated cellular stress pathways. However, this has not been well-studied. To help understand the role of fever in MIA, we used a recently described model of human brain development in which induced pluripotent stem cells (iPSCs) differentiate into 3-dimensional neuronal aggregates that resemble a first trimester telencephalon. RNA-seq was carried out on aggregates that were heat shocked at 39°C for 24 hours, along with their control partners maintained at 37°C. 186 genes showed significant differences in expression following HS (p<0.05), including known HS-inducible genes, as expected, as well as those coding for NGFR and a number of SZ and ASD candidates, including SMARCA2, DPP10, ARNT2, AHI1 and ZNF804A. The degree to which the expression of these genes decrease or increase during HS is similar to that found in copy loss and copy gain copy number variants (CNVs), although the effects of HS are likely to be transient. The dramatic effect on the expression of some SZ and ASD genes places HS, and perhaps other cellular stressors, into a common conceptual framework with disease-causing genetic variants. The findings also suggest that some candidate genes that are assumed to have a relatively limited impact on SZ and ASD pathogenesis based on a small number of positive genetic findings, such as SMARCA2 and ARNT2, may in fact have a much more substantial role in these disorders - as targets of common environmental stressors. |
| ARNT2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24465693  Title: Human variants in the neuronal basic helix-loop-helix/Per-Arnt-Sim (bHLH/PAS) transcription factor complex NPAS4/ARNT2 disrupt function.  Neuronal Per-Arnt-Sim homology (PAS) Factor 4 (NPAS4) is a neuronal activity-dependent transcription factor which heterodimerises with ARNT2 to regulate genes involved in inhibitory synapse formation. NPAS4 functions to maintain excitatory/inhibitory balance in neurons, while mouse models have shown it to play roles in memory formation, social interaction and neurodegeneration. NPAS4 has therefore been implicated in a number of neuropsychiatric or neurodegenerative diseases which are underpinned by defects in excitatory/inhibitory balance. Here we have explored a broad set of non-synonymous human variants in NPAS4 and ARNT2 for disruption of NPAS4 function. We found two variants in NPAS4 (F147S and E257K) and two variants in ARNT2 (R46W and R107H) which significantly reduced transcriptional activity of the heterodimer on a luciferase reporter gene. Furthermore, we found that NPAS4.F147S was unable to activate expression of the NPAS4 target gene BDNF due to reduced dimerisation with ARNT2. Homology modelling predicts F147 in NPAS4 to lie at the dimer interface, where it appears to directly contribute to protein/protein interaction. We also found that reduced transcriptional activation by ARNT2 R46W was due to disruption of nuclear localisation. These results provide insight into the mechanisms of NPAS4/ARNT dimerisation and transcriptional activation and have potential implications for cognitive phenotypic variation and diseases such as autism, schizophrenia and dementia. |
| ARNTL |
| Url: https://pubmed.ncbi.nlm.nih.gov/35682995  Title: Haploinsufficiency of a Circadian Clock Gene Bmal1 (Arntl or Mop3) Causes Brain-Wide mTOR Hyperactivation and Autism-like Behavioral Phenotypes in Mice.  Approximately 50-80% of children with autism spectrum disorders (ASDs) exhibit sleep problems, but the contribution of circadian clock dysfunction to the development of ASDs remains largely unknown. The essential clock gene Bmal1 (Arntl or Mop3) has been associated with human sociability, and its missense mutation is found in ASD. Our recent study found that Bmal1-null mice exhibit a variety of autism-like phenotypes. Here, we further investigated whether an incomplete loss of Bmal1 function could cause significant autism-like behavioral changes in mice. Our results demonstrated that heterozygous Bmal1 deletion (Bmal1+/-) reduced the Bmal1 protein levels by ~50-75%. Reduced Bmal1 expression led to decreased levels of clock proteins, including Per1, Per2, Cry 1, and Clock but increased mTOR activities in the brain. Accordingly, Bmal1+/- mice exhibited aberrant ultrasonic vocalizations during maternal separation, deficits in sociability and social novelty, excessive repetitive behaviors, impairments in motor coordination, as well as increased anxiety-like behavior. The novel object recognition memory remained intact. Together, these results demonstrate that haploinsufficiency of Bmal1 can cause autism-like behavioral changes in mice, akin to those identified in Bmal1-null mice. This study provides further experimental evidence supporting a potential role for disrupted clock gene expression in the development of ASD. |
| ARNTL |
| Url: https://pubmed.ncbi.nlm.nih.gov/34054130  Title: Genetic underpinnings of sociability in the general population.  Levels of sociability are continuously distributed in the general population, and decreased sociability represents an early manifestation of several brain disorders. Here, we investigated the genetic underpinnings of sociability in the population. We performed a genome-wide association study (GWAS) of a sociability score based on four social functioning-related self-report questions from 342,461 adults in the UK Biobank. Subsequently we performed gene-wide and functional follow-up analyses. Robustness analyses were performed in the form of GWAS split-half validation analyses, as well as analyses excluding neuropsychiatric cases. Using genetic correlation analyses as well as polygenic risk score analyses we investigated genetic links of our sociability score to brain disorders and social behavior outcomes. Individuals with autism spectrum disorders, bipolar disorder, depression, and schizophrenia had a lower sociability score. The score was significantly heritable (SNP h2 of 6%). We identified 18 independent loci and 56 gene-wide significant genes, including genes like ARNTL, DRD2, and ELAVL2. Many associated variants are thought to have deleterious effects on gene products and our results were robust. The sociability score showed negative genetic correlations with autism spectrum, disorders, depression, schizophrenia, and two sociability-related traits-loneliness and social anxiety-but not with bipolar disorder or Alzheimer's disease. Polygenic risk scores of our sociability GWAS were associated with social behavior outcomes within individuals with bipolar disorder and with major depressive disorder. Variation in population sociability scores has a genetic component, which is relevant to several psychiatric disorders. Our findings provide clues towards biological pathways underlying sociability. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/36845779  Title: Phenotypes of a female patient with novel de novo frameshift ARX variant identified by whole-exome sequencing: a case report.  Variants in the aristaless-related homeobox (ARX) gene cause a diverse spectrum of phenotypes of neurodevelopmental disorders (NDD) in male patients. This article describes the role of genetic testing using whole-exome sequencing (WES) in detecting a novel de novo frameshift variant in the ARX gene in a female patient with autism, seizure, and global developmental delay. A 2-year-old girl with frequent seizures, global developmental delay, and autistic features was referred to our hospital. She was the second child of consanguineous non-affected parents. She had a high forehead, mildly prominent ears, and prominent nasal root. A generalized epileptiform discharge was noted in her electroencephalography. Brain MRI revealed corpus callosum agenesis, cerebral atrophy, and a left parafalcine cyst. The WES result showed a likely pathogenic variant identified as a novel de novo deletion in exon 4 of the ARX gene, which creates a frameshift variant. The patient is on dual therapy of antiepilepsy drugs, physiotherapy, speech therapy, occupational therapy, and oral motor exercises. Variants in the ARX gene can result in various phenotypes in males transmitted from asymptomatic carrier females. However, several reports showed that the ARX variants might cause phenotypes in females with milder symptoms than affected males. We report a novel de novo ARX variant in an affected female with a NDD. Our study confirms that the ARX variant might cause remarkable pleiotropy phenotypes in females. Moreover, WES could help to identify the pathogenic variant in NDD patients with diverse phenotypes. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/35656878  Title: Generation of FLAG-tagged Arx knock-in mouse model.  The Aristaless-related homeobox (ARX) is a paired-like homeodomain transcription factor playing important roles in brain development. Patients with mutations in ARX have a spectrum of neurodevelopmental disorders such as epilepsy, intellectual disability, and autism spectrum disorder, with or without structural abnormalities of the brain such as lissencephaly (smooth brain), microcephaly (small brain), and/or agenesis of the corpus callosum. Mouse models have provided important clues on the pathophysiologic roles of ARX in these disorders. However, successfully isolating specific in vivo complexes of ARX, with DNA and proteins, has remained as a challenge. To facilitate in vivo detection of ARX complexes, we generated a mouse line containing one epitope of FLAG-tag (1 × FLAG) targeted at the translational start site of the endogenous Arx gene using CRSPR/Cas9 strategy. Homozygous Flag-Arx mice are viable and fertile without gross abnormality, suggesting that the FLAG-tag does not perturb the normal function of ARX. Using a FLAG antibody, we successfully detected ARX with immunofluorescent staining and pulled down ARX in embryonic brain tissues. This Flag-Arx mouse line will be a useful tool to isolate ARX complexes from mouse tissues for many applications. |
| ARX, KDM5C |
| Url: https://pubmed.ncbi.nlm.nih.gov/35328505  Title: Further Delineation of Duplications of ARX Locus Detected in Male Patients with Varying Degrees of Intellectual Disability.  The X-linked gene encoding aristaless-related homeobox (ARX) is a bi-functional transcription factor capable of activating or repressing gene transcription, whose mutations have been found in a wide spectrum of neurodevelopmental disorders (NDDs); these include cortical malformations, paediatric epilepsy, intellectual disability (ID) and autism. In addition to point mutations, duplications of the ARX locus have been detected in male patients with ID. These rearrangements include telencephalon ultraconserved enhancers, whose structural alterations can interfere with the control of ARX expression in the developing brain. Here, we review the structural features of 15 gain copy-number variants (CNVs) of the ARX locus found in patients presenting wide-ranging phenotypic variations including ID, speech delay, hypotonia and psychiatric abnormalities. We also report on a further novel Xp21.3 duplication detected in a male patient with moderate ID and carrying a fully duplicated copy of the ARX locus and the ultraconserved enhancers. As consequences of this rearrangement, the patient-derived lymphoblastoid cell line shows abnormal activity of the ARX-KDM5C-SYN1 regulatory axis. Moreover, the three-dimensional (3D) structure of the Arx locus, both in mouse embryonic stem cells and cortical neurons, provides new insight for the functional consequences of ARX duplications. Finally, by comparing the clinical features of the 16 CNVs affecting the ARX locus, we conclude that-depending on the involvement of tissue-specific enhancers-the ARX duplications are ID-associated risk CNVs with variable expressivity and penetrance. |
| ARX, KDM5C, ZNF711 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34356104  Title: Analysis of a Set of KDM5C Regulatory Genes Mutated in Neurodevelopmental Disorders Identifies Temporal Coexpression Brain Signatures.  Dysregulation of transcriptional pathways is observed in multiple forms of neurodevelopmental disorders (NDDs), such as intellectual disability (ID), epilepsy and autism spectrum disorder (ASD). We previously demonstrated that the NDD genes encoding lysine-specific demethylase 5C (KDM5C) and its transcriptional regulators Aristaless related-homeobox (ARX), PHD Finger Protein 8 (PHF8) and Zinc Finger Protein 711 (ZNF711) are functionally connected. Here, we show their relation to each other with respect to the expression levels in human and mouse datasets and in vivo mouse analysis indicating that the coexpression of these syntenic X-chromosomal genes is temporally regulated in brain areas and cellular sub-types. In co-immunoprecipitation assays, we found that the homeotic transcription factor ARX interacts with the histone demethylase PHF8, indicating that this transcriptional axis is highly intersected. Furthermore, the functional impact of pathogenic mutations of ARX, KDM5C, PHF8 and ZNF711 was tested in lymphoblastoid cell lines (LCLs) derived from children with varying levels of syndromic ID establishing the direct correlation between defects in the KDM5C-H3K4me3 pathway and ID severity. These findings reveal novel insights into epigenetic processes underpinning NDD pathogenesis and provide new avenues for assessing developmental timing and critical windows for potential treatments. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/33659799  Title: [Co-expression of glutamatergic and autism-related genes in the hippocampus of male mice with disturbances of social behavior].  There is a hypothesis of the involvement of the glutamatergic system in the development of autism. It has been shown that the chronic experience in daily intermale confrontations leads to disturbances in social behavior: a decrease in communicativeness, disturbances of socialization, emergence of stereotypical behaviors that can be considered as symptoms of the autistic spectrum disorders. So, the aim of this study was to investigate changes in the expression of glutamatergic (GG) and autism-related (GA) genes in the hippocampus of animals with impaired social behavior caused by repeated experience of social defeat or aggression in daily agonistic confrontations. To form groups of animals with contrasting behaviors, a model of sensory contact (chronic social stress) was used. The collected brain samples were sequenced at JSC Genoanalytica (http://genoanalytica.ru/ , Moscow, Russia). Transcriptomic analysis revealed a down-regulation of autism-related (Shank3, Auts2, Ctnnd2, Nrxn2) and glutamatergic (Grm4) genes in aggressive mice. At the same time, the expression of GA-related genes (Shank2, Nlgn2, Ptcdh10, Reln, Arx) and GG genes (Grik3, Grm2, Grm4, Slc17a7, Slc1a4, Slc25a22) excluding Grin2a was increased in defeated mice. Correlative analysis revealed a statistically significant association between GG and GA expression. These results can serve as a confirmation of the participation of the glutamatergic system in the pathophysiology of the autistic spectrum disorder. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/32383243  Title: Constraint and conservation of paired-type homeodomains predicts the clinical outcome of missense variants of uncertain significance.  The need to interpret the pathogenicity of novel missense variants of unknown significance identified in the homeodomain of X-chromosome aristaless-related homeobox (ARX) gene prompted us to assess the utility of conservation and constraint across these domains in multiple genes compared to conventional in vitro functional analysis. Pathogenic missense variants clustered in the homeodomain of ARX contribute to intellectual disability (ID) and epilepsy, with and without brain malformation in affected males. Here we report novel c.1112G>A, p.Arg371Gln and c.1150C>T, p.Arg384Cys variants in male patients with ID and severe seizures. The third case of a male patient with a c.1109C>T, p.Ala370Val variant is perhaps the first example of ID and autism spectrum disorder (ASD), without seizures or brain malformation. We compiled data sets of pathogenic variants from ClinVar and presumed benign variation from gnomAD and demonstrated that the high levels of sequence conservation and constraint of benign variation within the homeodomain impacts upon the ability of publicly available in silico prediction tools to accurately discern likely benign from likely pathogenic variants in these data sets. Despite this, considering the inheritance patterns of the genes and disease variants with the conservation and constraint of disease variants affecting the homeodomain in conjunction with current clinical assessments may assist in predicting the pathogenicity of missense variants, particularly for genes with autosomal recessive and X-linked patterns of disease inheritance, such as ARX. In vitro functional analysis demonstrates that the transcriptional activity of all three variants was diminished compared to ARX-Wt. We review the associated phenotypes of the published cases of patients with ARX homeodomain variants and propose expansion of the ARX-related phenotype to include severe ID and ASD without brain malformations or seizures. We propose that the use of the constraint and conservation data in conjunction with consideration of the patient phenotype and inheritance pattern may negate the need for the experimental functional validation currently required to achieve a diagnosis. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/29062978  Title: INTERNEURONOPATHIES AND THEIR ROLE IN EARLY LIFE EPILEPSIES AND NEURODEVELOPMENTAL DISORDERS.  GABAergic interneurons control the neural circuitry and network activity in the brain. The advances in genetics have identified genes that control the development, maturation and integration of GABAergic interneurons and implicated them in the pathogenesis of epileptic encephalopathies or neurodevelopmental disorders. For example, mutations of the Aristaless-Related homeobox X-linked gene (ARX) may result in defective GABAergic interneuronal migration in infants with epileptic encephalopathies like West syndrome (WS), Ohtahara syndrome or X-linked lissencephaly with abnormal genitalia (XLAG). The concept of "interneuronopathy", i.e. impaired development, migration or function of interneurons, has emerged as a possible etiopathogenic mechanism for epileptic encephalopathies. Treatments that enhance GABA levels, may help seizure control but do not necessarily show disease modifying effect. On the other hand, interneuronopathies can be seen in other conditions in which epilepsy may not be the primary manifestation, such as autism. In this review, we plan to outline briefly the current state of knowledge on the origin, development, and migration and integration of GABAergic interneurons, present neurodevelopmental conditions, with or without epilepsy, that have been associated with interneuronopathies and discuss the evidence linking certain types of interneuronal dysfunction with epilepsy and/or cognitive or behavioral deficits. |
| ARX, HDAC4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27798109  Title: Embryonic forebrain transcriptome of mice with polyalanine expansion mutations in the ARX homeobox gene.  The Aristaless-related homeobox (ARX) gene encodes a paired-type homeodomain transcription factor with critical roles in embryonic development. Mutations in ARX give rise to intellectual disability (ID), epilepsy and brain malformation syndromes. To capture the genetics and molecular disruptions that underpin the ARX-associated clinical phenotypes, we undertook a transcriptome wide RNASeq approach to analyse developing (12.5 dpc) telencephalon of mice modelling two recurrent polyalanine expansion mutations with different phenotypic severities in the ARX gene. Here we report 238 genes significantly deregulated (Log2FC  > +/-1.1, P-value <0.05) when both mutations are compared to wild-type (WT) animals. When each mutation is considered separately, a greater number of genes were deregulated in the severe PA1 mice (825) than in the PA2 animals (78). Analysing genes deregulated in either or both mutant strains, we identified 12% as implicated in ID, epilepsy and autism (99/858), with ∼5% of them as putative or known direct targets of ARX transcriptional regulation. We propose a core pathway of transcription regulators, including Hdac4, involved in chromatin condensation and transcriptional repression, and one of its targets, the transcription factor Twist1, as potential drivers of the ID and infantile spasms in patients with ARX polyalanine expansion mutations. We predict that the subsequent disturbance to this pathway is a consequence of ARX protein reduction with a broader and more significant level of disruption in the PA1 in comparison to the PA2 mice. Identifying early triggers of ARX-associated phenotypes contributes to our understanding of particular clusters/pathways underpinning comorbid phenotypes that are shared by many neurodevelopmental disorders. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/26337422  Title: Copy number variants in patients with intellectual disability affect the regulation of ARX transcription factor gene.  Protein-coding mutations in the transcription factor-encoding gene ARX cause various forms of intellectual disability (ID) and epilepsy. In contrast, variations in surrounding non-coding sequences are correlated with milder forms of non-syndromic ID and autism and had suggested the importance of ARX gene regulation in the etiology of these disorders. We compile data on several novel and some already identified patients with or without ID that carry duplications of ARX genomic region and consider likely genetic mechanisms underlying the neurodevelopmental defects. We establish the long-range regulatory domain of ARX and identify its brain region-specific autoregulation. We conclude that neurodevelopmental disturbances in the patients may not simply arise from increased dosage due to ARX duplication. This is further exemplified by a small duplication involving a non-functional ARX copy, but with duplicated enhancers. ARX enhancers are located within a 504-kb region and regulate expression specifically in the forebrain in developing and adult zebrafish. Transgenic enhancer-reporter lines were used as in vivo tools to delineate a brain region-specific negative and positive autoregulation of ARX. We find autorepression of ARX in the telencephalon and autoactivation in the ventral thalamus. Fluorescently labeled brain regions in the transgenic lines facilitated the identification of neuronal outgrowth and pathfinding disturbances in the ventral thalamus and telencephalon that occur when arxa dosage is diminished. In summary, we have established a model for how breakpoints in long-range gene regulation alter the expression levels of a target gene brain region-specifically, and how this can cause subtle neuronal phenotypes relating to the etiology of associated neuropsychiatric disease. |
| ARX, KDM5A, KDM5C, KMT2A, KMT2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/25135975  Title: Regulation of histone H3K4 methylation in brain development and disease.  The growing list of mutations implicated in monogenic disorders of the developing brain includes at least seven genes (ARX, CUL4B, KDM5A, KDM5C, KMT2A, KMT2C, KMT2D) with loss-of-function mutations affecting proper regulation of histone H3 lysine 4 methylation, a chromatin mark which on a genome-wide scale is broadly associated with active gene expression, with its mono-, di- and trimethylated forms differentially enriched at promoter and enhancer and other regulatory sequences. In addition to these rare genetic syndromes, dysregulated H3K4 methylation could also play a role in the pathophysiology of some cases diagnosed with autism or schizophrenia, two conditions which on a genome-wide scale are associated with H3K4 methylation changes at hundreds of loci in a subject-specific manner. Importantly, the reported alterations for some of the diseased brain specimens included a widespread broadening of H3K4 methylation profiles at gene promoters, a process that could be regulated by the UpSET(KMT2E/MLL5)-histone deacetylase complex. Furthermore, preclinical studies identified maternal immune activation, parental care and monoaminergic drugs as environmental determinants for brain-specific H3K4 methylation. These novel insights into the epigenetic risk architectures of neurodevelopmental disease will be highly relevant for efforts aimed at improved prevention and treatment of autism and psychosis spectrum disorders. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/19587282  Title: A triplet repeat expansion genetic mouse model of infantile spasms syndrome, Arx(GCG)10+7, with interneuronopathy, spasms in infancy, persistent seizures, and adult cognitive and behavioral impairment.  Infantile spasms syndrome (ISS) is a catastrophic pediatric epilepsy with motor spasms, persistent seizures, mental retardation, and in some cases, autism. One of its monogenic causes is an insertion mutation [c.304ins (GCG)(7)] on the X chromosome, expanding the first polyalanine tract of the interneuron-specific transcription factor Aristaless-related homeobox (ARX) from 16 to 23 alanine codons. Null mutation of the Arx gene impairs GABA and cholinergic interneuronal migration but results in a neonatal lethal phenotype. We developed the first viable genetic mouse model of ISS that spontaneously recapitulates salient phenotypic features of the human triplet repeat expansion mutation. Arx((GCG)10+7) ("Arx plus 7") pups display abnormal spasm-like myoclonus and other key EEG features, including multifocal spikes, electrodecremental episodes, and spontaneous seizures persisting into maturity. The neurobehavioral profile of Arx mutants was remarkable for lowered anxiety, impaired associative learning, and abnormal social interaction. Laminar decreases of Arx+ cortical interneurons and a selective reduction of calbindin-, but not parvalbumin- or calretinin-expressing interneurons in neocortical layers and hippocampus indicate that specific classes of synaptic inhibition are missing from the adult forebrain, providing a basis for the seizures and cognitive disorder. A significant reduction of calbindin-, NPY (neuropeptide Y)-expressing, and cholinergic interneurons in the mutant striatum suggest that dysinhibition within this network may contribute to the dyskinetic motor spasms. This mouse model narrows the range of critical pathogenic elements within brain inhibitory networks essential to recreate this complex neurodevelopmental syndrome. |
| ARX, DLX2, EN2, FOXP2, OTX1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19018235  Title: Polymorphisms of coding trinucleotide repeats of homeogenes in neurodevelopmental psychiatric disorders.  Autism (MIM#209850) and schizophrenia (MIM#181500) are both neurodevelopmental psychiatric disorders characterized by a highly genetic component. Homeogenes and forkhead genes encode transcription factors, which have been involved in brain development and cell differentiation. Thus, they are relevant candidate genes for psychiatric disorders. Genetic studies have reported an association between autism and DLX2, HOXA1, EN2, ARX, and FOXP2 genes whereas only three studies of EN2, OTX2, and FOXP2 were performed on schizophrenia. Interestingly, most of these candidate genes contain trinucleotide repeats coding for polyamino acid stretch in which instability can be the cause of neurodevelopmental disorders. Our goal was to identify variations of coding trinucleotide repeats in schizophrenia, autism, and idiopathic mental retardation. We screened the coding trinucleotide repeats of OTX1, EN1, DLX2, HOXA1, and FOXP2 genes in populations suffering from schizophrenia (247 patients), autism (98 patients), and idiopathic mental retardation (56 patients), and compared them with control populations (112 super controls and 202 healthy controls). Novel deletions and insertions of coding trinucleotide repeats were found in the DLX2, HOXA1, and FOXP2 genes. Most of these variations were detected in controls and no difference in their distribution was observed between patient and control groups. Two different polymorphisms in FOXP2 were, however, found only in autistic patients and the functional consequences of these variations of repeats have to be characterized and correlated to particular clinical features. This study did not identify specific disease risk variants of trinucleotide repeats in OTX1, EN1, DLX2, HOXA1, and FOXP2 candidate genes in neurodevelopmental psychiatric disorders. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/17331656  Title: Aristaless-related homeobox gene, the gene responsible for West syndrome and related disorders, is a Groucho/transducin-like enhancer of split dependent transcriptional repressor.  Aristaless-related homeobox gene (ARX) is an important paired-type homeobox gene involved in the development of human brain. The ARX gene mutations are a significant contributor to various forms of X-chromosome-linked mental retardation with and without additional features including epilepsy, lissencephaly with abnormal genitalia, hand dystonia or autism. Here we demonstrate that the human ARX protein is a potent transcriptional repressor, which binds to Groucho/transducin-like enhancer of split (TLE) co-factor proteins and the TLE1 in particular through its octapeptide (Engrailed homology repressor domain (eh-1) homology) domain. We show that the transcription repression activity of ARX is modulated by two strong repression domains, one located within the octapeptide domain and the second in the region of the polyalanine tract 4, and one activator domain, the aristaless domain. Importantly, we show that the transcription repression activity of ARX is affected by various naturally occurring mutations. The introduction of the c.98T>C (p.L33P) mutation results in the lack of binding to TLE1 protein and relaxed transcription repression. The introduction of the two most frequent ARX polyalanine tract expansion mutations increases the repression activity in a manner dependent on the number of extra alanines. Interestingly, deletions of alanine residues within polyalanine tracts 1 and 2 show low or no effect. In summary we demonstrate that the ARX protein is a strong transcription repressor, we identify novel ARX interacting proteins (TLE) and offer an explanation of a molecular pathogenesis of some ARX mutations, including the most frequent ARX mutations, the polyalanine tract expansion mutations, c.304ins(GCG)7 and c.428\_451dup. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/17044103  Title: Mutation screening of the ARX gene in patients with autism.  Mutations in the Aristaless related homeobox (ARX) gene are associated with a broad spectrum of disorders, including nonsyndromic X-linked mental retardation, sometimes associated with epilepsy, as well as syndromic forms with brain abnormalities and abnormal genitalia. Furthermore, ARX mutations have been described in a few patients with autism or autistic features. In this study, we screened the ARX gene in 226 male patients with autism spectrum disorders and mental retardation; 42 of the patients had epilepsy. The mutation analysis was performed by direct sequencing of all exons and flanking regions. No ARX mutations were identified in any of the patients tested. These findings indicate that mutations in the ARX gene are very rare in autism. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/14722918  Title: Mutations of ARX are associated with striking pleiotropy and consistent genotype-phenotype correlation.  We recently identified mutations of ARX in nine genotypic males with X-linked lissencephaly with abnormal genitalia (XLAG), and in several female relatives with isolated agenesis of the corpus callosum (ACC). We now report 13 novel and two recurrent mutations of ARX, and one nucleotide change of uncertain significance in 20 genotypic males from 16 families. Most had XLAG, but two had hydranencephaly and abnormal genitalia, and three males from one family had Proud syndrome or ACC with abnormal genitalia. We obtained detailed clinical information on all 29 affected males, including the nine previously reported subjects. Premature termination mutations consisting of large deletions, frameshifts, nonsense mutations, and splice site mutations in exons 1 to 4 caused XLAG or hydranencephaly with abnormal genitalia. Nonconservative missense mutations within the homeobox caused less severe XLAG, while conservative substitution in the homeodomain caused Proud syndrome. A nonconservative missense mutation near the C-terminal aristaless domain caused unusually severe XLAG with microcephaly and mild cerebellar hypoplasia. In addition, several less severe phenotypes without malformations have been reported, including mental retardation with cryptogenic infantile spasms (West syndrome), other seizure types, dystonia or autism, and nonsyndromic mental retardation. The ARX mutations associated with these phenotypes have included polyalanine expansions or duplications, missense mutations, and one deletion of exon 5. Together, the group of phenotypes associated with ARX mutations demonstrates remarkable pleiotropy, but also comprises a nearly continuous series of developmental disorders that begins with hydranencephaly, lissencephaly, and agenesis of the corpus callosum, and ends with a series of overlapping syndromes with apparently normal brain structure. |
| ARX |
| Url: https://pubmed.ncbi.nlm.nih.gov/14631200  Title: The ARX story (epilepsy, mental retardation, autism, and cerebral malformations): one gene leads to many phenotypes.  Infantile spasms, mental retardation, autism, and dystonia represent disabling diseases for which little etiologic information is available. Mutations in the Aristaless related homeobox gene (ARX) have been found in patients with these conditions. This discovery provides important genetic information and may ultimately offer treatment options for these patients. Recent work has demonstrated that mutations in ARX cause X-linked West syndrome, X-linked myoclonic epilepsy with spasticity and intellectual disability, Partington syndrome (mental retardation, ataxia, and dystonia), as well as nonsyndromic forms of mental retardation. Patients with these aforementioned diseases and ARX mutations were not reported to have brain imaging abnormalities. In contrast, mutations in ARX mutations have also been found in X-linked lissencephaly with abnormal genitalia, which typically includes severe brain malformations (lissencephaly, agenesis of the corpus callosum, and midbrain malformations), intractable seizures, and a severely shortened lifespan. ARX knockout mice manifest defects in overall neuroblast proliferation as well as selective abnormalities in gamma-aminobutyric acid-ergic interneuron migration. Consistent with these findings in mice, phenotype/genotype studies in humans suggest that truncating mutations cause X-linked lissencephaly with abnormal genitalia, and insertion/missense mutations result in epilepsy and mental retardation without cortical dysplasia. Mutations in the homeobox gene, ARX, cause a diverse spectrum of disease that includes cognitive impairment, epilepsy, and in another group of patients severe cortical malformations. Although the precise prevalence of ARX mutations is unclear, ARX may rival Fragile X as a cause of mental retardation and epilepsy in males. |
| ASH1L, CREBBP, NSD1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36104286  Title: Identification of a transcriptional signature found in multiple models of ASD and related disorders.  Epigenetic regulation plays a critical role in many neurodevelopmental disorders (NDDs), including autism spectrum disorder (ASD). In particular, many such disorders are the result of mutations in genes that encode chromatin-modifying proteins. However, although these disorders share many features, it is unclear whether they also share gene expression disruptions resulting from the aberrant regulation of chromatin. We examined five chromatin modifiers that are all linked to ASD despite their different roles in regulating chromatin. Specifically, we depleted ASH1L, CHD8, CREBBP, EHMT1, and NSD1 in parallel in a highly controlled neuronal culture system. We then identified sets of shared genes, or transcriptional signatures, that are differentially expressed following loss of multiple ASD-linked chromatin modifiers. We examined the functions of genes within the transcriptional signatures and found an enrichment in many neurotransmitter transport genes and activity-dependent genes. In addition, these genes are enriched for specific chromatin features such as bivalent domains that allow for highly dynamic regulation of gene expression. The down-regulated transcriptional signature is also observed within multiple mouse models of NDDs that result in ASD, but not those only associated with intellectual disability. Finally, the down-regulated transcriptional signature can distinguish between control and idiopathic ASD patient iPSC-derived neurons as well as postmortem tissue, demonstrating that this gene set is relevant to the human disorder. This work identifies a transcriptional signature that is found within many neurodevelopmental syndromes, helping to elucidate the link between epigenetic regulation and the underlying cellular mechanisms that result in ASD. |
| ASH1L, CHD2, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36061363  Title: Disease similarity network analysis of Autism Spectrum Disorder and comorbid brain disorders.  Autism Spectrum Disorder (ASD) is a neurodevelopmental disorder with heterogeneous clinical presentation, variable severity, and multiple comorbidities. A complex underlying genetic architecture matches the clinical heterogeneity, and evidence indicates that several co-occurring brain disorders share a genetic component with ASD. In this study, we established a genetic similarity disease network approach to explore the shared genetics between ASD and frequent comorbid brain diseases (and subtypes), namely Intellectual Disability, Attention-Deficit/Hyperactivity Disorder, and Epilepsy, as well as other rarely co-occurring neuropsychiatric conditions in the Schizophrenia and Bipolar Disease spectrum. Using sets of disease-associated genes curated by the DisGeNET database, disease genetic similarity was estimated from the Jaccard coefficient between disease pairs, and the Leiden detection algorithm was used to identify network disease communities and define shared biological pathways. We identified a heterogeneous brain disease community that is genetically more similar to ASD, and that includes Epilepsy, Bipolar Disorder, Attention-Deficit/Hyperactivity Disorder combined type, and some disorders in the Schizophrenia Spectrum. To identify loss-of-function rare de novo variants within shared genes underlying the disease communities, we analyzed a large ASD whole-genome sequencing dataset, showing that ASD shares genes with multiple brain disorders from other, less genetically similar, communities. Some genes (e.g., SHANK3, ASH1L, SCN2A, CHD2, and MECP2) were previously implicated in ASD and these disorders. This approach enabled further clarification of genetic sharing between ASD and brain disorders, with a finer granularity in disease classification and multi-level evidence from DisGeNET. Understanding genetic sharing across disorders has important implications for disease nosology, pathophysiology, and personalized treatment. |
| ASH1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/35449559  Title: Neural Hyperactivity Is a Core Pathophysiological Change Induced by Deletion of a High Autism Risk Gene Ash1L in the Mouse Brain.  ASH1L is one of the highest risk genes associated with autism spectrum disorder (ASD) and intellectual disability (ID). Our recent studies demonstrate that loss of Ash1l in the mouse brain is sufficient to induce ASD/ID-like behavioral and cognitive deficits, suggesting that disruptive ASH1L mutations are likely to have a positive correlation with ASD/ID genesis. However, the core pathophysiological changes in the Ash1l-deficient brain remain largely unknown. Here we show that loss of Ash1l in the mouse brain causes locomotor hyperactivity, high metabolic activity, and hyperactivity-related disturbed sleep and lipid metabolic changes. In addition, the mutant mice display lower thresholds for the convulsant reagent-induced epilepsy and increased neuronal activities in multiple brain regions. Thus, our current study reveals that neural hyperactivity is a core pathophysiological change in the Ash1l-deficient mouse brain, which may function as a brain-level mechanism leading to the Ash1l-deletion-induced brain functional abnormalities and autistic-like behavioral deficits. |
| ASH1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/35210569  Title: Counteracting epigenetic mechanisms regulate the structural development of neuronal circuitry in human neurons.  Autism spectrum disorders (ASD) are associated with defects in neuronal connectivity and are highly heritable. Genetic findings suggest that there is an overrepresentation of chromatin regulatory genes among the genes associated with ASD. ASH1 like histone lysine methyltransferase (ASH1L) was identified as a major risk factor for ASD. ASH1L methylates Histone H3 on Lysine 36, which is proposed to result primarily in transcriptional activation. However, how mutations in ASH1L lead to deficits in neuronal connectivity associated with ASD pathogenesis is not known. We report that ASH1L regulates neuronal morphogenesis by counteracting the catalytic activity of Polycomb Repressive complex 2 group (PRC2) in stem cell-derived human neurons. Depletion of ASH1L decreases neurite outgrowth and decreases expression of the gene encoding the neurotrophin receptor TrkB whose signaling pathway is linked to neuronal morphogenesis. The neuronal morphogenesis defect is overcome by inhibition of PRC2 activity, indicating that a balance between the Trithorax group protein ASH1L and PRC2 activity determines neuronal morphology. Thus, our work suggests that ASH1L may epigenetically regulate neuronal morphogenesis by modulating pathways like the BDNF-TrkB signaling pathway. Defects in neuronal morphogenesis could potentially impair the establishment of neuronal connections which could contribute to the neurodevelopmental pathogenesis associated with ASD in patients with ASH1L mutations. |
| ASH1L, EZH2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35081333  Title: ASH1L haploinsufficiency results in autistic-like phenotypes in mice and links Eph receptor gene to autism spectrum disorder.  ASD-associated genes are enriched for synaptic proteins and epigenetic regulators. How those chromatin modulators establish ASD traits have remained unknown. We find haploinsufficiency of Ash1l causally induces anxiety and autistic-like behavior, including repetitive behavior, and alters social behavior. Specific depletion of Ash1l in forebrain induces similar ASD-associated behavioral defects. While the learning ability remains intact, the discrimination ability of Ash1l mutant mice is reduced. Mechanistically, deletion of Ash1l in neurons induces excessive synapses due to the synapse pruning deficits, especially during the post-learning period. Dysregulation of synaptic genes is detected in Ash1l mutant brain. Specifically, Eph receptor A7 is downregulated in Ash1l+/- mice through accumulating EZH2-mediated H3K27me3 in its gene body. Importantly, increasing activation of EphA7 in Ash1l+/- mice by supplying its ligand, ephrin-A5, strongly promotes synapse pruning and rescues discrimination deficits. Our results suggest that Ash1l haploinsufficiency is a highly penetrant risk factor for ASD, resulting from synapse pruning deficits. |
| ASH1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/34782621  Title: Deficiency of autism risk factor ASH1L in prefrontal cortex induces epigenetic aberrations and seizures.  ASH1L, a histone methyltransferase, is identified as a top-ranking risk factor for autism spectrum disorder (ASD), however, little is known about the biological mechanisms underlying the link of ASH1L haploinsufficiency to ASD. Here we show that ASH1L expression and H3K4me3 level are significantly decreased in the prefrontal cortex (PFC) of postmortem tissues from ASD patients. Knockdown of Ash1L in PFC of juvenile mice induces the downregulation of risk genes associated with ASD, intellectual disability (ID) and epilepsy. These downregulated genes are enriched in excitatory and inhibitory synaptic function and have decreased H3K4me3 occupancy at their promoters. Furthermore, Ash1L deficiency in PFC causes the diminished GABAergic inhibition, enhanced glutamatergic transmission, and elevated PFC pyramidal neuronal excitability, which is associated with severe seizures and early mortality. Chemogenetic inhibition of PFC pyramidal neuronal activity, combined with the administration of GABA enhancer diazepam, rescues PFC synaptic imbalance and seizures, but not autistic social deficits or anxiety-like behaviors. These results have revealed the critical role of ASH1L in regulating synaptic gene expression and seizures, which provides insights into treatment strategies for ASH1L-associated brain diseases. |
| ASH1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/34509565  Title: Vorinostat, a histone deacetylase inhibitor, ameliorates the sociability and cognitive memory in an Ash1L-deletion-induced ASD/ID mouse model.  Autism spectrum disorder (ASD) and intellectual disability (ID) are neurodevelopmental diseases associated with various gene mutations. Previous genetic and clinical studies reported that ASH1L is a high ASD risk gene identified in human patients. Our recent study used a mouse model to demonstrate that loss of ASH1L in the developing mouse brain was sufficient to cause multiple developmental defects, core autistic-like behaviors, and impaired cognitive memory, suggesting that the disruptive ASH1L mutations are the causative drivers leading the human ASD/ID genesis. Using this Ash1L-deletion-induced ASD/ID mouse model, here we showed that postnatal administration of vorinostat (SAHA), a histone deacetylase inhibitor (HDACi), significantly ameliorated both ASD-like behaviors and ID-like cognitive memory deficit. Thus, our study demonstrates that SAHA is a promising reagent for the pharmacological treatment of core ASD/ID behavioral and memory deficits caused by disruptive ASH1L mutations. |
| ASH1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/34145365  Title: Loss of histone methyltransferase ASH1L in the developing mouse brain causes autistic-like behaviors.  Autism spectrum disorder (ASD) is a neurodevelopmental disease associated with various gene mutations. Recent genetic and clinical studies report that mutations of the epigenetic gene ASH1L are highly associated with human ASD and intellectual disability (ID). However, the causality and underlying molecular mechanisms linking ASH1L mutations to genesis of ASD/ID remain undetermined. Here we show loss of ASH1L in the developing mouse brain is sufficient to cause multiple developmental defects, core autistic-like behaviors, and impaired cognitive memory. Gene expression analyses uncover critical roles of ASH1L in regulating gene expression during neural cell development. Thus, our study establishes an ASD/ID mouse model revealing the critical function of an epigenetic factor ASH1L in normal brain development, a causality between Ash1L mutations and ASD/ID-like behaviors in mice, and potential molecular mechanisms linking Ash1L mutations to brain functional abnormalities. |
| ASH1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/33258273  Title: Role of Ash1l in Tourette syndrome and other neurodevelopmental disorders.  Ash1l potentially contributes to neurodevelopmental diseases. Although specific Ash1l mutations are rare, they have led to informative studies in animal models that may bring therapeutic advances. Ash1l is highly expressed in the brain and correlates with the neuropathology of Tourette syndrome (TS), autism spectrum disorder, and intellectual disability during development, implicating shared epigenetic factors and overlapping neuropathological mechanisms. Functional convergence of Ash1l generated several significant signaling pathways: chromatin remodeling and transcriptional regulation, protein synthesis and cellular metabolism, and synapse development and function. Here, we systematically review the literature on Ash1l, including its discovery, expression, function, regulation, implication in the nervous system, signaling pathway, mutations, and putative involvement in TS and other neurodevelopmental traits. Such findings highlight Ash1l pleiotropy and the necessity of transcending a single gene to complicated mechanisms of network convergence underlying these diseases. With the progress in functional genomic analysis (highlighted in this review), and although the importance and necessity of Ash1l becomes increasingly apparent in the medical field, further research is required to discover the precise function and molecular regulatory mechanisms related to Ash1l. Thus, a new perspective is proposed for basic scientific research and clinical interventions for cross-disorder diseases. |
| ASH1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/32553196  Title: De Novo Variants in CNOT1, a Central Component of the CCR4-NOT Complex Involved in Gene Expression and RNA and Protein Stability, Cause Neurodevelopmental Delay.  CNOT1 is a member of the CCR4-NOT complex, which is a master regulator, orchestrating gene expression, RNA deadenylation, and protein ubiquitination. We report on 39 individuals with heterozygous de novo CNOT1 variants, including missense, splice site, and nonsense variants, who present with a clinical spectrum of intellectual disability, motor delay, speech delay, seizures, hypotonia, and behavioral problems. To link CNOT1 dysfunction to the neurodevelopmental phenotype observed, we generated variant-specific Drosophila models, which showed learning and memory defects upon CNOT1 knockdown. Introduction of human wild-type CNOT1 was able to rescue this phenotype, whereas mutants could not or only partially, supporting our hypothesis that CNOT1 impairment results in neurodevelopmental delay. Furthermore, the genetic interaction with autism-spectrum genes, such as ASH1L, DYRK1A, MED13, and SHANK3, was impaired in our Drosophila models. Molecular characterization of CNOT1 variants revealed normal CNOT1 expression levels, with both mutant and wild-type alleles expressed at similar levels. Analysis of protein-protein interactions with other members indicated that the CCR4-NOT complex remained intact. An integrated omics approach of patient-derived genomics and transcriptomics data suggested only minimal effects on endonucleolytic nonsense-mediated mRNA decay components, suggesting that de novo CNOT1 variants are likely haploinsufficient hypomorph or neomorph, rather than dominant negative. In summary, we provide strong evidence that de novo CNOT1 variants cause neurodevelopmental delay with a wide range of additional co-morbidities. Whereas the underlying pathophysiological mechanism warrants further analysis, our data demonstrate an essential and central role of the CCR4-NOT complex in human brain development. |
| ATRX, CREBBP, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33880059  Title: Whole-Exome Sequencing for Identifying Genetic Causes of Intellectual Developmental Disorders.  Intellectual developmental disorders (IDD) generally refers to the persistent impairment of cognitive activities and mental retardation caused by physical damage to the brain or incomplete brain development. We aimed to explore its genetic causes. In this study, 21 IDD patients were recruited. The Gesell developmental scales (GDS) and Wechsler intelligence scale for children (WISC) were used to assess the impaired level of intellectual development for all probands. A superconducting MRI scanner (Philips AcsNT 3.0 T Philips, Best, The Netherlands) was used to perform a plain MRI scan of the skull on the probands. The whole-exome sequencing was carried out using next-generation sequencing in all probands and their families. Eight had seizures and four had typical characteristics of autism. Pregnancy and delivery were uneventful except for three patients. Moderate IDD (52.4%) accounted for the majority. The abnormal MRI results included ventriculomegaly, pachygyria, broadening external cerebral space, abnormal signal change and agenesis of corpus callosum. Eleven variants were identified, including the variant in CREBBP, MECP2, HCFC1, ATRX, RAB39B, CLCN4, DYRK1A and CASKgenes. The function areas result of gene-positive group were compared to that of gene-negative group. Not significant (p>0.05) items were revealed after this analysis. Eleven variants were identified, including the variant in CREBBP, MECP2, HCFC1, ATRX, RAB39B, CLCN4, DYRK1A and CASK genes. The function areas result of gene-positive group were not significantly different from the gene-negative group. |
| ATRX |
| Url: https://pubmed.ncbi.nlm.nih.gov/31846713  Title: Conserved and divergent expression dynamics during early patterning of the telencephalon in mouse and chick embryos.  The mammalian and the avian telencephalon are nearly indistinguishable at early embryonic vesicle stages but differ substantially in form and function at their adult stage. We sequenced and analyzed RNA populations present in mouse and chick during the early stages of embryonic telencephalon to understand conserved and lineage-specific developmental differences. We found approximately 3000 genes that orchestrate telencephalon development. Many chromatin-associated epigenetic and transcription regulators show high expression in both species and some show species-specific expression dynamics. Interestingly, previous studies associated them to autism, intellectual disabilities, and mental retardation supporting a causal link between their impaired functions during telencephalon development and brain dysfunction. Strikingly, the conserved up-regulated genes were differentially enriched in ontologies related to development or functions of the adult brain. Moreover, a differential enrichment of distinct repertoires of transcription factor binding motifs in their upstream promoter regions suggest a species-specific regulation of the various gene groups identified. Overall, our results reveal that the ontogenetic divergences between the mouse and chick telencephalon result from subtle differences in the regulation of common patterning signaling cascades and regulatory networks unique to each species at their very early stages of development. |
| ATRX |
| Url: https://pubmed.ncbi.nlm.nih.gov/30392976  Title: Complete Disruption of Autism-Susceptibility Genes by Gene Editing Predominantly Reduces Functional Connectivity of Isogenic Human Neurons.  Autism spectrum disorder (ASD) is phenotypically and genetically heterogeneous. We present a CRISPR gene editing strategy to insert a protein tag and premature termination sites creating an induced pluripotent stem cell (iPSC) knockout resource for functional studies of ten ASD-relevant genes (AFF2/FMR2, ANOS1, ASTN2, ATRX, CACNA1C, CHD8, DLGAP2, KCNQ2, SCN2A, TENM1). Neurogenin 2 (NGN2)-directed induction of iPSCs allowed production of excitatory neurons, and mutant proteins were not detectable. RNA sequencing revealed convergence of several neuronal networks. Using both patch-clamp and multi-electrode array approaches, the electrophysiological deficits measured were distinct for different mutations. However, they culminated in a consistent reduction in synaptic activity, including reduced spontaneous excitatory postsynaptic current frequencies in AFF2/FMR2-, ASTN2-, ATRX-, KCNQ2-, and SCN2A-null neurons. Despite ASD susceptibility genes belonging to different gene ontologies, isogenic stem cell resources can reveal common functional phenotypes, such as reduced functional connectivity. |
| ATRX |
| Url: https://pubmed.ncbi.nlm.nih.gov/23246909  Title: An epigenetic framework for neurodevelopmental disorders: from pathogenesis to potential therapy.  Neurodevelopmental disorders (NDDs) are characterized by aberrant and delayed early-life development of the brain, leading to deficits in language, cognition, motor behaviour and other functional domains, often accompanied by somatic symptoms. Environmental factors like perinatal infection, malnutrition and trauma can increase the risk of the heterogeneous, multifactorial and polygenic disorders, autism and schizophrenia. Conversely, discrete genetic anomalies are involved in Down, Rett and Fragile X syndromes, tuberous sclerosis and neurofibromatosis, the less familiar Phelan-McDermid, Sotos, Kleefstra, Coffin-Lowry and "ATRX" syndromes, and the disorders of imprinting, Angelman and Prader-Willi syndromes. NDDs have been termed "synaptopathies" in reference to structural and functional disturbance of synaptic plasticity, several involve abnormal Ras-Kinase signalling ("rasopathies"), and many are characterized by disrupted cerebral connectivity and an imbalance between excitatory and inhibitory transmission. However, at a different level of integration, NDDs are accompanied by aberrant "epigenetic" regulation of processes critical for normal and orderly development of the brain. Epigenetics refers to potentially-heritable (by mitosis and/or meiosis) mechanisms controlling gene expression without changes in DNA sequence. In certain NDDs, prototypical epigenetic processes of DNA methylation and covalent histone marking are impacted. Conversely, others involve anomalies in chromatin-modelling, mRNA splicing/editing, mRNA translation, ribosome biogenesis and/or the regulatory actions of small nucleolar RNAs and micro-RNAs. Since epigenetic mechanisms are modifiable, this raises the hope of novel therapy, though questions remain concerning efficacy and safety. The above issues are critically surveyed in this review, which advocates a broad-based epigenetic framework for understanding and ultimately treating a diverse assemblage of NDDs ("epigenopathies") lying at the interface of genetic, developmental and environmental processes. This article is part of the Special Issue entitled 'Neurodevelopmental Disorders'. |
| BAZ2B |
| Url: https://pubmed.ncbi.nlm.nih.gov/35353793  Title: Genome-wide DNA methylation profiles of autism spectrum disorder.  We aimed to identify differentially methylated genes and related signaling pathways in autism spectrum disorder (ASD). First, the DNA methylation profile in the brain samples (GSE131706 and GSE80017) and peripheral blood samples (GSE109905) was downloaded from the Gene Expression Omnibus database (GEO) dataset, followed by identification of differentially methylated genes and functional analysis. Second, the GSE109905 data set was used to further validate the methylation state and test the ability to diagnose disease of identified differentially methylated genes. Third, expression measurement of selected differentially methylated genes was performed in whole blood from an independent sample. Finally, protein-protein interaction (PPI) network of core differentially methylated genes was constructed. Totally, 74 differentially methylated genes were identified in ASD, including 38 hypermethylated genes and 36 hypomethylated genes. 15 differentially methylated genes were further identified after validation in the GSE109905 data set. Among these, major histocompatibility complex (HLA)-DQA1 was involved in the molecular function of myosin heavy chain class II receptor activity; HLA-DRB5 was involved in the signaling pathways of cell adhesion molecules, Epstein-Barr virus infection and antigen processing and presentation. In the PPI analysis, the interaction pairs of HLA-DQA1 and HLA-DRB5, FMN2 and ACTR3, and CALCOCO2 and BAZ2B were identified. Interestingly, FMN2, BAZ2B, HLA-DRB5, CALCOCO2 and DUSP22 had a potential diagnostic value for patients with ASD. The expression result of four differentially methylated genes (HLA-DRB5, NTM, IL16 and COL5A3) in the independent sample was consistent with the integrated analysis. Identified differentially methylated genes and enriched signaling pathway could be associated with ASD. |
| BCL11A, FOXP1, TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36579832  Title: Characterisation of the TBR1 interactome: variants associated with neurodevelopmental disorders disrupt novel protein interactions.  TBR1 is a neuron-specific transcription factor involved in brain development and implicated in a neurodevelopmental disorder (NDD) combining features of autism spectrum disorder (ASD), intellectual disability (ID) and speech delay. TBR1 has been previously shown to interact with a small number of transcription factors and co-factors also involved in NDDs (including CASK, FOXP1/2/4 and BCL11A), suggesting that the wider TBR1 interactome may have a significant bearing on normal and abnormal brain development. Here we have identified approximately 250 putative TBR1-interaction partners by affinity purification coupled to mass spectrometry. As well as known TBR1-interactors such as CASK, the identified partners include transcription factors and chromatin modifiers, along with ASD- and ID-related proteins. Five interaction candidates were independently validated using bioluminescence resonance energy transfer assays. We went on to test the interaction of these candidates with TBR1 protein variants implicated in cases of NDD. The assays uncovered disturbed interactions for NDD-associated variants and identified two distinct protein-binding domains of TBR1 that have essential roles in protein-protein interaction. |
| BCL11A, CTNNB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33997709  Title: Cell-type and fetal-sex-specific targets of prenatal alcohol exposure in developing mouse cerebral cortex.  Prenatal alcohol exposure (PAE) results in cerebral cortical dysgenesis. Single-cell RNA sequencing was performed on murine fetal cerebral cortical cells from six timed pregnancies, to decipher persistent cell- and sex-specific effects of an episode of PAE during early neurogenesis. We found, in an analysis of 38 distinct neural subpopulations across 8 lineage subtypes, that PAE altered neural maturation and cell cycle and disrupted gene co-expression networks. Whereas most differentially regulated genes were inhibited, particularly in females, PAE also induced sex-independent neural expression of fetal hemoglobin, a presumptive epigenetic stress adaptation. PAE inhibited Bcl11a, Htt, Ctnnb1, and other upstream regulators of differentially expressed genes and inhibited several autism-linked genes, suggesting that neurodevelopmental disorders share underlying mechanisms. PAE females exhibited neural loss of X-inactivation, with correlated activation of autosomal genes and evidence for spliceosome dysfunction. Thus, episodic PAE persistently alters the developing neural transcriptome, contributing to sex- and cell-type-specific teratology. |
| BCL11A, FEZF2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32610082  Title: A Chromatin Accessibility Atlas of the Developing Human Telencephalon.  To discover regulatory elements driving the specificity of gene expression in different cell types and regions of the developing human brain, we generated an atlas of open chromatin from nine dissected regions of the mid-gestation human telencephalon, as well as microdissected upper and deep layers of the prefrontal cortex. We identified a subset of open chromatin regions (OCRs), termed predicted regulatory elements (pREs), that are likely to function as developmental brain enhancers. pREs showed temporal, regional, and laminar differences in chromatin accessibility and were correlated with gene expression differences across regions and gestational ages. We identified two functional de novo variants in a pRE for autism risk gene SLC6A1, and using CRISPRa, demonstrated that this pRE regulates SCL6A1. Additionally, mouse transgenic experiments validated enhancer activity for pREs proximal to FEZF2 and BCL11A. Thus, this atlas serves as a resource for decoding neurodevelopmental gene regulation in health and disease. |
| BCL11A |
| Url: https://pubmed.ncbi.nlm.nih.gov/32322190  Title: Bcl11 Transcription Factors Regulate Cortical Development and Function.  Transcription factors regulate multiple processes during brain development and in the adult brain, from brain patterning to differentiation and maturation of highly specialized neurons as well as establishing and maintaining the functional neuronal connectivity. The members of the zinc-finger transcription factor family Bcl11 are mainly expressed in the hematopoietic and central nervous systems regulating the expression of numerous genes involved in a wide range of pathways. In the brain Bcl11 proteins are required to regulate progenitor cell proliferation as well as differentiation, migration, and functional integration of neural cells. Mutations of the human Bcl11 genes lead to anomalies in multiple systems including neurodevelopmental impairments like intellectual disabilities and autism spectrum disorders. |
| BCL11A |
| Url: https://pubmed.ncbi.nlm.nih.gov/28694195  Title: Hemoglobins emerging roles in mental disorders. Metabolical, genetical and immunological aspects.  Hemoglobin (Hb) expression in the central nervous system is recently shown. Cooccurences of mental disorders (mainly bipolar disorder (BD) and tic disorders) with β- or α-thalassemia trait or erythrocytosis were witnessed, which may be due to peripheral or central hypoxia/hyperoxia or haplotypal gene interactions. β-Globin genes reside at 11p15.5 close to tyrosine hydroxylase, dopamine receptor DRD4 and Brain Derived Neurotrophic Factor, which involve in psychiatric diseases. α-Globin genes reside at 16p13.3 which associates with BD, tic disorders, ATR-16 Syndrome and Rubinstein Taybi Syndrome (RTS). CREB-Binding Protein (CEBBP)-gene is mutated in RTS, which commonly associates with mood disorders. 16p13.3 region also contains GRIN2A gene encoding N-methyl-d-aspartate receptor-2A and SSTR5 (Somatostatin Receptor-5), again involving in mental disorders. We demonstrated a protective role of minor HbA2 against post-partum episodes in BD and association of higher minor HbF (fetal hemoglobin) levels with family history of psychosis in a BD-patient cohort. HbA2 increases in cardiac ischemia and in mountain dwellers indicating its likely protection against ischemia/hypoxia. HMGIY, a repressive transcription factor of δ-globin chain of HbA2 is increased in lymphocytes of schizophrenics. In autism, deletional mutations were found in BCL11A gene, which cause persistence of HbF at high levels in adulthood. Also, certain polymorphisms in BCL11A strongly associate with schizophrenia. Further, many drugs from anabolic steroids to antimalarial agents elevate HbF and may cause mania. We ascribe a protective role to HbA2 and a maladaptive detrimental role to HbF in psychopathology. We believe that future studies on hemoglobins may pave to discover novel pathogenesis mechanisms in mental disorders. |
| BCL11A |
| Url: https://pubmed.ncbi.nlm.nih.gov/28573701  Title: Molecular and clinical delineation of 2p15p16.1 microdeletion syndrome.  Interstitial 2p15p16.1 microdeletion is a rare chromosomal syndrome previously reported in 33 patients. It is characterized by intellectual disability, developmental delay, autism spectrum disorders, microcephaly, short stature, dysmorphic features, and multiple congenital organ defects. It is defined as a contiguous gene syndrome and two critical regions have been proposed at 2p15 and 2p16.1 loci. Nevertheless, patients with deletion of both critical regions shared similar features of the phenotype and the correlation genotype-phenotype is still unclear. We review all published cases and describe three additional patients, to define the phenotype-genotype correlation more precisely. We reported on two patients including the first prenatal case described so far, carrying a 2p15 deletion affecting two genes: XPO1 and part of USP34. Both patients shared similar features including facial dysmorphism and cerebral abnormalities. We considered the genes involved in the deleted segment to further understand the abnormal phenotype. The third case we described here was a 4-year-old boy with a heterozygous de novo 427 kb deletion encompassing BCL11A and PAPOLG at 2p16.1. He displayed speech delay, autistic traits, and motor stereotypies associated with brain structure abnormalities. We discuss the contribution of the genes included in the deletion to the abnormal phenotype. Our three new patients compared to previous cases, highlighted that despite two critical regions, both distal deletion at 2p16.1 and proximal deletion at 2p15 are associated with phenotypes that are very close to each other. Finally, we also discuss the genetic counseling of this microdeletion syndrome particularly in the course of prenatal diagnosis. |
| BTRC |
| Url: https://pubmed.ncbi.nlm.nih.gov/34469739  Title: Full-length isoform transcriptome of the developing human brain provides further insights into autism.  Alternative splicing plays an important role in brain development, but its global contribution to human neurodevelopmental diseases (NDDs) requires further investigation. Here we examine the relationships between splicing isoform expression in the brain and de novo loss-of-function mutations from individuals with NDDs. We analyze the full-length isoform transcriptome of the developing human brain and observe differentially expressed isoforms and isoform co-expression modules undetectable by gene-level analyses. These isoforms are enriched in loss-of-function mutations and microexons, are co-expressed with a unique set of partners, and have higher prenatal expression. We experimentally test the effect of splice-site mutations and demonstrate exon skipping in five NDD risk genes, including SCN2A, DYRK1A, and BTRC. Our results suggest that the splice site mutation in BTRC reduces translational efficiency, likely affecting Wnt signaling through impaired degradation of β-catenin. We propose that functional effects of mutations should be investigated at the isoform- rather than gene-level resolution. |
| CAMK2A, TSHZ3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34349780  Title: Camk2a-Cre and Tshz3 Expression in Mouse Striatal Cholinergic Interneurons: Implications for Autism Spectrum Disorder.  Camk2a-Cre mice have been widely used to study the postnatal function of several genes in forebrain projection neurons, including cortical projection neurons (CPNs) and striatal medium-sized spiny neurons (MSNs). We linked heterozygous deletion of TSHZ3/Tshz3 gene to autism spectrum disorder (ASD) and used Camk2a-Cre mice to investigate the postnatal function of Tshz3, which is expressed by CPNs but not MSNs. Recently, single-cell transcriptomics of the adult mouse striatum revealed the expression of Camk2a in interneurons and showed Tshz3 expression in striatal cholinergic interneurons (SCINs), which are attracting increasing interest in the field of ASD. These data and the phenotypic similarity between the mice with Tshz3 haploinsufficiency and Camk2a-Cre-dependent conditional deletion of Tshz3 (Camk2a-cKO) prompted us to better characterize the expression of Tshz3 and the activity of Camk2a-Cre transgene in the striatum. Here, we show that the great majority of Tshz3-expressing cells are SCINs and that all SCINs express Tshz3. Using lineage tracing, we demonstrate that the Camk2a-Cre transgene is expressed in the SCIN lineage where it can efficiently elicit the deletion of the Tshz3-floxed allele. Moreover, transcriptomic and bioinformatic analysis in Camk2a-cKO mice showed dysregulated striatal expression of a number of genes, including genes whose human orthologues are associated with ASD and synaptic signaling. These findings identifying the expression of the Camk2a-Cre transgene in SCINs lineage lead to a reappraisal of the interpretation of experiments using Camk2a-Cre-dependent gene manipulations. They are also useful to decipher the cellular and molecular substrates of the ASD-related behavioral abnormalities observed in Tshz3 mouse models. |
| CAMK2A |
| Url: https://pubmed.ncbi.nlm.nih.gov/29935919  Title: Increased expression of BDNF mRNA in the frontal cortex of autistic patients.  Autistic spectrum disorders (ASDs) are neurodevelopmental disorders for which genetic components have been well defined. However, specific gene deregulations related to synapse function in the autistic brain have not been as extensively described. Based on a candidate genes approach, we present in this study the expression data of 4 transcripts of interest (BDNF, CAMK2a, NR-CAM and RIMS1) located at the synapse in two regions of interest in the context of the ASDs; the lobule VI of cerebellum and the Brodmann area 46. We have also genotyped in our cohort the coding single nucleotide polymorphism rs6265, located in the BDNF gene. After correction for age and sex, whereas no change was observed in the lobule VI between controls and autistic patients, we found a significant increase of BDNF expression level in the BA46 from autistic patients. No significant interaction between the rs6265 genotype and autism was observed for the BDNF expression. However, "A" allele carriers are more likely to have increased BDNF levels. Finally, we found a significant positive correlation between BDNF and RIMS1 expression levels. Our data suggest that these two molecules which are involved in cell signalling at the synapse, might have coordinated expressions and, that BDNF regulation in the brain has to be investigated further in the context of ASDs. |
| CAMK2A |
| Url: https://pubmed.ncbi.nlm.nih.gov/25929186  Title: Camk2a-Cre-mediated conditional deletion of chromatin remodeler Brg1 causes perinatal hydrocephalus.  Mammalian SWI/SNF-like BAF chromatin remodeling complexes are essential for many aspects of neural development. Mutations in the genes encoding the core subunit Brg1/SmarcA4 or other complex components cause neurodevelopmental diseases and are associated with autism. Congenital hydrocephalus is a serious brain disorder often experienced by these patients. We report a role of Brg1 in the pathogenesis of hydrocephalus disorder. We discovered an unexpected early activity of mouse Camk2a-Cre transgene, which mediates Brg1 deletion in a subset of forebrain neurons beginning in the late embryonic stage. Brg1 deletion in these neurons led to severe congenital hydrocephalus with enlargement of the lateral ventricles and attenuation of the cerebral cortex. The Brg1-deficient mice had significantly smaller subcommissural organs and narrower Sylvian aqueducts than mice that express normal levels of Brg1. Effects were non-cell autonomous and may be responsible for the development of the congenital hydrocephalus phenotype. Our study provides evidence indicating that abnormalities in Brg1 function result in defects associated with neurodevelopmental disorders and autism. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/36837929  Title: Prenatal SSRI Exposure Increases the Risk of Autism in Rodents via Aggravated Oxidative Stress and Neurochemical Changes in the Brain.  The mechanisms underlying selective serotonin reuptake inhibitor (SSRI) use during pregnancy as a major autism risk factor are unclear. Here, brain neurochemical changes following fluoxetine exposure and in an autism model were compared to determine the effects on autism risk. The study was performed on neonatal male western albino rats which were divided into Groups one (control), two (propionic acid [PPA]-induced autism model), and three (prenatal SSRI-exposed newborn rats whose mothers were exposed to 5 mg/kg of fluoxetine over gestation days 10-20). SSRI (fluoxetine) induced significant neurochemical abnormalities in the rat brain by increasing lipid peroxide (MDA), Interferon-gamma (IFN-γ), and caspase-3 levels and by depleting Glutathione (GSH), Glutathione S-transferases (GST), Catalase, potassium (K+), and Creatine kinase (CK) levels, similarly to what has been discovered in the PPA model of autism when compared with control. Prenatal fluoxetine exposure plays a significant role in asset brain damage in newborns; further investigation of fluoxetine as an autism risk factor is thus warranted. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/34984596  Title: Centella asiatica Alleviates AlCl3-induced Cognitive Impairment, Oxidative Stress, and Neurodegeneration by Modulating Cholinergic Activity and Oxidative Burden in Rat Brain.  Aluminum (Al) is linked to the development of many neurological disorders such as Alzheimer's disease (AD), Parkinson's disease, and autism. Centella asiatica (CA) is a regenerating herb traditionally used to stimulate memory. This study was designed to assess the neuroprotective role of ethanolic extract of CA (CAE) in AlCl3-induced neurological conditions in rats. Adult rats were chronically treated with AlCl3 (100 mg/kg b.w./day) for 60 days to establish the dementia model, and co-administration of CAE was evaluated for its ability to attenuate the toxic effect of AlCl3. CAE was given orally at a dose of 150 and 300 mg/kg b.w./day, for 60 days. The behavioral performances of rats were tested through Y-maze and open field tests. Lipid peroxidation, superoxide dismutase, and catalase activity were evaluated to measure oxidative stress; and acetylcholinesterase (AChE) activity was assessed to evaluate cholinergic dysfunction in the rat brain. H&E staining was used to assess structural abnormalities in the cortex and hippocampus. The result showed that AlCl3 induces cognitive dysfunction (impaired learning and memory, anxiety, diminished locomotor activity), oxidative stress, cholinergic impairment, and histopathological alteration in the rat brain. Co-administration of CAE with AlCl3 markedly protects the brain from AlCl3-induced cognitive dysfunction, oxidative stress, AChE activity, and cytoarchitectural alterations. Furthermore, 15 days CAE treatment after 45 days AlCl3 administration markedly ameliorates the AlCl3-induced neurotoxicity indicating its potential for therapeutic use. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/34528217  Title: Lead (Pb)-induced oxidative stress mediates sex-specific autistic-like behaviour in Drosophila melanogaster.  Autism spectrum disorder (ASD) is a highly prevalent neurodevelopmental disorder characterised by three main behavioural symptoms: abnormal social interaction, verbal and non-verbal communication impairments, and repetitive and restricted activities or interests. Even though the exact aetiology of ASD remains unknown, studies have shown a link between genetics and environmental pollutants. Heavy metal lead (Pb), the environmental pollutant, is associated with ASD. Pb may also exhibit sex-specific ASD behaviour, as has been demonstrated in the global human populations. Drosophila melanogaster as a model has been used in the present study to understand the involvement of Pb-induced oxidative stress in developing ASD behaviour. The larval feeding technique has been employed to administer different Pb concentrations (0.2-0.8 mM) to Oregon-R (ORR), superoxide dismutase (Sod), or catalase (Cat) antioxidants overexpressed or knockdown flies. Adult Drosophila (5-day old) were used for Pb content, biochemical, and behavioural analysis.Pb accumulated in the Drosophila brain induces oxidative stress and exhibited a human autistic-like behaviour such as reduced climbing, increased grooming, increased social spacing, and decreased learning and memory in a sex-specific manner.Pb-induced autistic-like behaviour was intensified in Sod or Cat-knockdown flies, whereas Sod or Cat-overexpressed flies overcome that behavioural alterations. These results unequivocally proved that Pb-induced oxidative stress causes ASD behaviour of humans in Drosophila. Thus, Drosophila is used as a model organism to analyse ASD-like human behaviour and underlines the importance of using antioxidant therapy in alleviating ASD symptoms in children. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/34298871  Title: Curcumin Potentiates α7 Nicotinic Acetylcholine Receptors and Alleviates Autistic-Like Social Deficits and Brain Oxidative Stress Status in Mice.  Autistic spectrum disorder (ASD) refers to a group of neurodevelopmental disorders characterized by impaired social interaction and cognitive deficit, restricted repetitive behaviors, altered immune responses, and imbalanced oxidative stress status. In recent years, there has been a growing interest in studying the role of nicotinic acetylcholine receptors (nAChRs), specifically α7-nAChRs, in the CNS. Influence of agonists for α7-nAChRs on the cognitive behavior, learning, and memory formation has been demonstrated in neuro-pathological condition such as ASD and attention-deficit hyperactivity disorder (ADHD). Curcumin (CUR), the active compound of the spice turmeric, has been shown to act as a positive allosteric modulator of α7-nAChRs. Here we hypothesize that CUR, acting through α7-nAChRs, influences the neuropathology of ASD. In patch clamp studies, fast inward currents activated by choline, a selective agonist of α7-nAChRs, were significantly potentiated by CUR. Moreover, choline induced enhancement of spontaneous inhibitory postsynaptic currents was markedly increased in the presence of CUR. Furthermore, CUR (25, 50, and 100 mg/kg, i.p.) ameliorated dose-dependent social deficits without affecting locomotor activity or anxiety-like behaviors of tested male Black and Tan BRachyury (BTBR) mice. In addition, CUR (50 and 100 mg/kg, i.p.) mitigated oxidative stress status by restoring the decreased levels of superoxide dismutase (SOD) and catalase (CAT) in the hippocampus and the cerebellum of treated mice. Collectively, the observed results indicate that CUR potentiates α7-nAChRs in native central nervous system neurons, mitigates disturbed oxidative stress, and alleviates ASD-like features in BTBR mice used as an idiopathic rodent model of ASD, and may represent a promising novel pharmacological strategy for ASD treatment. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/33785420  Title: Astrocyte-mediated disruption of ROS homeostasis in Fragile X mouse model.  Astrocytes, glial cells within the brain, work to protect neurons during high levels of activity by maintaining oxidative homeostasis via regulation of energy supply and antioxidant systems. In recent years, mitochondrial dysfunction has been highlighted as an underlying factor of pathology in many neurological disorders. In animal studies of Fragile X Syndrome (FXS), the leading genetic cause of autism, higher levels of reactive oxygen species, lipid peroxidation, and protein oxidation within the brain indicates that mitochondria function is also altered in FXS. Despite their integral contribution to redox homeostasis within the CNS, the role of astrocytes on the occurrence or progression of neurodevelopmental disorders in this way is rarely considered. This study specifically examines changes to astrocyte mitochondrial function and antioxidant expression that may occur in FXS. Using the Fmr1 knockout (KO) mouse model, mitochondrial respiration and reactive oxygen species (ROS) emission were analyzed in primary cortical astrocytes. While mitochondrial respiration was similar between genotypes, ROS emission was significantly elevated in Fmr1 KO astrocytes. Notably, NADPH-oxidase 2 expression in Fmr1 KO astrocytes was also enhanced but only changes in catalase antioxidant enzyme expression were noted. Characterization of astrocyte factors involved in redox imbalance is invaluable to uncovering potential sources of oxidative stress in neurodevelopmental disorders and more specifically, the intercellular mechanisms that contribute to dysfunction in FXS. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/33669336  Title: The Multi-Targeting Ligand ST-2223 with Histamine H3 Receptor and Dopamine D2/D3 Receptor Antagonist Properties Mitigates Autism-Like Repetitive Behaviors and Brain Oxidative Stress in Mice.  Autism spectrum disorder (ASD) is a complex heterogeneous neurodevelopmental disorder characterized by social and communicative impairments, as well as repetitive and restricted behaviors (RRBs). With the limited effectiveness of current pharmacotherapies in treating repetitive behaviors, the present study determined the effects of acute systemic treatment of the novel multi-targeting ligand ST-2223, with incorporated histamine H3 receptor (H3R) and dopamine D2/D3 receptor affinity properties, on ASD-related RRBs in a male Black and Tan BRachyury (BTBR) mouse model of ASD. ST-2223 (2.5, 5, and 10 mg/kg, i.p.) significantly mitigated the increase in marble burying and self-grooming, and improved reduced spontaneous alternation in BTBR mice (all p p p p < 0.05). These preliminary in vivo findings demonstrate the ameliorative effects of ST-2223 on RRBs in a mouse model of ASD, suggesting its pharmacological prospective to rescue core ASD-related behaviors. Further confirmatory investigations on its effects on various brain neurotransmitters, e.g., dopamine and histamine, in different brain regions are still warranted to corroborate and expand these initial data. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/33220538  Title: Glyphosate targets fish monoaminergic systems leading to oxidative stress and anxiety.  Glyphosate is the active ingredient of some of the most highly produced and used herbicides worldwide. The intensive applications of glyphosate-based herbicides and its half-life in water lead to its presence in many aquatic ecosystems. Whereas recent studies have reported neurotoxic effects of glyphosate including autism-related effects, most of them used extremely high (mg/L to g/L) concentrations, so it is still unclear if chronic, low environmentally relevant concentrations of this compound (ng/L to μg/L) can induce neurotoxicity. In this study we analyzed the neurotoxicity of glyphosate in adult zebrafish after waterborne exposure to environmentally relevant concentrations (0.3 and 3 μg/L) for two weeks. Our data showed that exposed fish presented a significant impairment of exploratory and social behaviors consistent with increased anxiety. The anterior brain of the exposed fish presented a significant increase in dopamine and serotonin levels, as well as in the DOPAC/dopamine and homovanillic acid/dopamine turnover ratios. Moreover, the expression of genes involved in the dopaminergic system, as th1, th2, comtb, and scl6a3 was downregulated. Finally, the brain of exposed fish presented a significant increase in the catalase and superoxide dismutase activities, with a concomitant decrease of glutathione stores. These changes in the antioxidant defense system are consistent with the observed increase in oxidative stress, reflected by the increase in the levels of lipid peroxidation in the brain. The presented results show that current glyphosate concentrations commonly found in many aquatic ecosystems may have detrimental consequences on fish survival by decreasing exploration of the environment or altering social interactions. Furthermore, as zebrafish is also a vertebrate model widely used in human neurobehavioral studies, these results are relevant not only for environmental risk assessment, but also for understanding the risk of chronic low-dose exposures on human health. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/32994972  Title: Antioxidant and hepatorenal protective effects of bee pollen fractions against propionic acid-induced autistic feature in rats.  In the brain, propionic acid (PA) can cross cell membranes and accumulate within cells, leading to intracellular acidification, which may alter neurotransmitter release (NT), communication between neurons, and behavior. Such elevation in levels of PA constitutes a neurodevelopmental metabolic disorder called propionic acidemia, which could clinically manifest as autism. The purpose of this study was to investigate the protective effects of different fractions of bee pollen (BP) on PA-induced autism in rats, and to evaluate their effects on the expression of liver and renal biomarkers. Groups of rats received treatments of different fractions of BP at a dose of 250 mg/kg of body weight/day for a period of 1 month. Normal control group I and group II were orally administered with phosphate-buffered saline and propionic acid, respectively, for 3 days. BP contains various health-promoting phenolic components. Different fractions of BP administered pre- and post-treatment with PA showed significant reduction in the levels of liver and renal biomarkers (p < .05). Also, a significant enhancement in the levels of glutathione S-transferase (GST), catalase CAT), and ascorbic acid (VIT C) was observed. Supplementation with BP significantly reduced biochemical changes in the liver, kidneys, and brain of rats with PA-induced toxicity. It exhibited protective effects against oxidative damage and reactive oxygen species produced by PA-induced adverse reactions in rats. Taken together, our study shows that BP possesses protective effects in PA-induced liver and kidney damage. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/32876824  Title: Exposure to a high dose of amoxicillin causes behavioral changes and oxidative stress in young zebrafish.  Autistic spectrum disorder (ASD) is a group of early-onset neurodevelopmental disorders characterized by impaired social and communication skills. Autism is widely described as a behavioral syndrome with multiple etiologies where may exhibit neurobiological, genetic, and psychological deficits. Studies have indicated that long term use of antibiotics can alter the intestinal flora followed by neuroendocrine changes, leading to behavioral changes. Indeed, previous studies demonstrate that a high dose of amoxicillin can change behavioral parameters in murine animal models. The objective was to evaluate behavioral and oxidative stress parameters in zebrafish exposed to a high dose of amoxicillin for 7 days. Young zebrafish were exposed to a daily concentration of amoxicillin (100 mg/L) for 7 days. Subsequently, the behavioral analysis was performed, and the brain content was dissected for the evaluation of oxidative stress parameters. Zebrafish exposed to a high dose of amoxicillin showed locomotor alteration and decreased social interaction behavior. In addition, besides the significant decrease of sulfhydryl content, there was a marked decrease in catalase activity, as well as an increased superoxide dismutase activity in brain tissue. Thus, through the zebrafish model was possible to note a central effect related to the exposition of amoxicillin, the same as observed in murine models. Further, the present data reinforce the relation of the gut-brain-axis and the use of zebrafish as a useful tool to investigate new therapies for autistic traits. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/32503208  Title: The Dual-Active Histamine H3 Receptor Antagonist and Acetylcholine Esterase Inhibitor E100 Alleviates Autistic-Like Behaviors and Oxidative Stress in Valproic Acid Induced Autism in Mice.  The histamine H3 receptor (H3R) functions as auto- and hetero-receptors, regulating the release of brain histamine (HA) and acetylcholine (ACh), respectively. The enzyme acetylcholine esterase (AChE) is involved in the metabolism of brain ACh. Both brain HA and ACh are implicated in several cognitive disorders like Alzheimer's disease, schizophrenia, anxiety, and narcolepsy, all of which are comorbid with autistic spectrum disorder (ASD). Therefore, the novel dual-active ligand E100 with high H3R antagonist affinity (hH3R: Ki = 203 nM) and balanced AChE inhibitory effect (EeAChE: IC50 = 2 µM and EqBuChE: IC50 = 2 µM) was investigated on autistic-like sociability, repetitive/compulsive behaviour, anxiety, and oxidative stress in male C57BL/6 mice model of ASD induced by prenatal exposure to valproic acid (VPA, 500 mg/kg, intraperitoneal (i.p.)). Subchronic systemic administration with E100 (5, 10, and 15 mg/kg, i.p.) significantly and dose-dependently attenuated sociability deficits of autistic (VPA) mice in three-chamber behaviour (TCB) test (all p p p p p < 0.05). These results demonstrate the promising effects of E100 on in-vivo VPA-induced ASD-like features in mice, and provide evidence that a potent dual-active H3R antagonist and AChE inhibitor (AChEI) is a potential drug candidate for future therapeutic management of autistic-like behaviours. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/31226268  Title: Maternal glyphosate-based herbicide exposure alters antioxidant-related genes in the brain and serum metabolites of male rat offspring.  In response to the rapid development of genetically engineered glyphosate-tolerant crops, the use of glyphosate-based herbicides (GBHs), in agriculture, has increased substantially. Currently, it is estimated that 747 million kg of GBHs are applied per year. Although several epidemiological studies have demonstrated that there are health risks associated with GBH exposure, the effects these chemicals have on the oxidative and inflammatory response in the brain are still unclear. In fact, alterations in these processes could contribute to the development of neurological diseases, such as Alzheimer's disease and autism spectrum disorders. The present study exposed pregnant rats to GBH and evaluated changes in the expression of genes related to oxidnte defense and inflammation response and monitored the serum metabolome in the adult male offspring. Pregnant Wistar rats were administered distilled water or Roundup®, at either 5 and 50 mg/kg/day, (p.o.) from gestational day (GD) 18 to postnatal day (PND) 5. There was a significant increase in the gene expression levels of Neuroglobin (Ngb - oxygen storage and tissue protection) (105%, p = 0.031), Glutathione Peroxidase 1 (Gpx1 - oxidative stress) (95%, p = 0.005), Prostaglandin-Endoperoxidase Synthase 1 (Ptgs1 - inflammation) (109%, p = 0.033) and Hypoxia inducible factor 1 subunit alpha (Hif1α - oxygen sensor) (73%, p = 0.017), in the cerebellum of PND90 rats perinatally exposed to 50 mg GBH/kg/day. Moreover, both GBH-exposed groups displayed a significant decrease in the expression of Catalase (Cat - oxidative stress) (49%, p = 0.003; and 31% p = 0.050, respectively) expression, in the cortex. Serum metabolites analyses, from the same animals of each group, demonstrated that there were significant changes in the concentrations of lysophosphatidylcholine and phosphatidylcholine, which have been associated with neurodegenerative diseases. The results of the present study suggest GBH exposure during pregnancy alters the expression of genes associated with oxidant defense, inflammation and lipid metabolism. It is plausible that maternal GBH exposure could have lasting neuronal effects on the offspring later in life. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/27034117  Title: Memantine ameliorates autistic behavior, biochemistry & blood brain barrier impairments in rats.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder, commonly characterized by altered social behavior, communication, biochemistry and pathological conditions. One percent of the worldwide population suffers from autism and males suffer more than females. NMDA receptors have the important role in neurodevelopment, neuropsychiatric and neurodegenerative disorders. This study has been designed to investigate the role of memantine, a NMDA receptor modulator, in prenatal valproic acid-induced autism in rats. Animals with prenatal valproic acid have shown the reduction in social interaction (three-chamber social behavior apparatus), spontaneous alternation (Y-Maze), exploratory activity (Hole board test), intestinal motility, serotonin levels (both in prefrontal cortex and ileum) and prefrontal cortex mitochondrial complex activity (complex I, II, IV). Furthermore, prenatal valproic acid-treated animals have shown an increase in locomotion (actophotometer), anxiety (elevated plus maze), brain oxidative stress (thiobarbituric acid reactive species, glutathione, catalase), nitrosative stress (nitrite/nitrate), inflammation (both in brain and ileum myeloperoxidase activity), calcium and blood-brain barrier permeability. Treatment with memantine has significantly attenuated prenatal valproic acid-induced reduction in social interaction, spontaneous alteration, exploratory activity intestinal motility, serotonin levels and prefrontal cortex mitochondrial complex activity. Furthermore, memantine has also attenuated the prenatal valproic acid-induced increase in locomotion, anxiety, brain oxidative and nitrosative stress, inflammation, calcium and blood-brain barrier permeability. Thus, it may be concluded that prenatal valproic acid has induced autistic behavior, biochemistry and blood-brain barrier impairment in animals, which were significantly attenuated by memantine. NMDA receptor modulators like memantine should be explored further for the therapeutic benefits in autism. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/26551768  Title: Minocycline ameliorates prenatal valproic acid induced autistic behaviour, biochemistry and blood brain barrier impairments in rats.  Autism is a neurodevelopment disorder. One percent worldwide population suffers with autism and males suffer more than females. Microglia plays an important role in neurodevelopment, neuropsychiatric and neurodegenerative disorders. The present study has been designed to investigate the role of minocycline in prenatal valproic acid induced autism in rats. Animals with prenatal valproic acid have reduced social interaction (three chamber social behaviour apparatus), spontaneous alteration (Y-Maze), exploratory activity (Hole board test), intestinal motility, serotonin levels (both in prefrontal cortex and ileum) and prefrontal cortex mitochondrial complex activity (complexes I, II, IV). Furthermore, prenatal valproic acid treated animals have shown an increase in locomotion (actophotometer), anxiety (elevated plus maze), brain oxidative stress (thiobarbituric acid reactive species, glutathione, catalase), nitrosative stress (nitrite/nitrate), inflammation (both in brain and ileum myeloperoxidase activity), calcium and blood brain barrier permeability. Treatment with minocycline significantly attenuated prenatal valproic acid induced reduction in social interaction, spontaneous alteration, exploratory activity intestinal motility, serotonin levels and prefrontal cortex mitochondrial complex activity. Furthermore, minocycline has also attenuated prenatal valproic acid induced increase in locomotion, anxiety, brain oxidative and nitrosative stress, inflammation, calcium and blood brain barrier permeability. Thus, it may be concluded that prenatal valproic acid has induced autistic behaviour, biochemistry and blood brain barrier impairment in animals, which were significantly attenuated by minocycline. Minocycline should be explored further for its therapeutic benefits in autism. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/26498253  Title: Benefits of agomelatine in behavioral, neurochemical and blood brain barrier alterations in prenatal valproic acid induced autism spectrum disorder.  Valproic acid administration during gestational period causes behavior and biochemical deficits similar to those observed in humans with autism spectrum disorder. Although worldwide prevalence of autism spectrum disorder has been increased continuously, therapeutic agents to ameliorate the social impairment are very limited. The present study has been structured to investigate the therapeutic potential of melatonin receptor agonist, agomelatine in prenatal valproic acid (Pre-VPA) induced autism spectrum disorder in animals. Pre-VPA has produced reduction in social interaction (three chamber social behavior apparatus), spontaneous alteration (Y-Maze), exploratory activity (Hole board test), intestinal motility, serotonin levels (prefrontal cortex and ileum) and prefrontal cortex mitochondrial complex activity (complex I, II, IV). Furthermore, Pre-VPA has increased locomotor activity (actophotometer), anxiety, brain oxidative stress (thiobarbituric acid reactive species, glutathione, and catalase), nitrosative stress (nitrite/nitrate), inflammation (brain and ileum myeloperoxidase activity), calcium levels and blood brain barrier leakage in animals. Treatment with agomelatine has significantly attenuated Pre-VPA induced reduction in social interaction, spontaneous alteration, exploratory activity intestinal motility, serotonin levels and prefrontal cortex mitochondrial complex activity. Furthermore, agomelatine also attenuated Pre-VPA induced increase in locomotion, anxiety, brain oxidative stress, nitrosative stress, inflammation, calcium levels and blood brain barrier leakage. It is concluded that, Pre-VPA has induced autism spectrum disorder, which was attenuated by agomelatine. Agomelatine has shown ameliorative effect on behavioral, neurochemical and blood brain barrier alteration in Pre-VPA exposed animals. Thus melatonin receptor agonists may provide beneficial therapeutic strategy for managing autism spectrum disorder. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/26276454  Title: Laser Acupuncture Improves Behavioral Disorders and Brain Oxidative Stress Status in the Valproic Acid Rat Model of Autism.  The therapeutic strategy against autism, a severe neurological development disorder, is one of the challenges of this decade. Recent findings show that oxidative stress plays a crucial role on the pathophysiology of autism, and laser acupuncture at Shenmen (HT7) can improve oxidative status in many neurological disorders. Therefore, we aimed to assess the effect of laser acupuncture at HT7 on behavior disorders and oxidative stress status in the cortex, striatum, and hippocampus of the valproic acid rat model of autism. Laser acupuncture was performed once daily during postnatal day (PND) 14-PND 40. Behavioral tests including rotarod, open-field, learning and memory, and social behavior tests were performed during PND 14-PND 40. At the end of study, brain oxidative status including malondialdehyde levels and the activities of superoxide dismutase, catalase, and glutathione peroxidase were determined in the cortex, striatum, and hippocampus. Laser acupuncture at HT7 significantly improved autistic-like behaviors. Decreased malondialdehyde levels were observed in all areas mentioned above, however, increased glutathione peroxidase activity was observed only in the striatum and hippocampus. No changes in superoxide dismutase and catalase activities were observed in any investigated area of the brain. Therefore, our study suggests that laser acupuncture at HT7 partly mitigates autistic-like symptoms via improved oxidative status. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/26264039  Title: Neuroprotective effect of Tagara, an Ayurvedic drug against methyl mercury induced oxidative stress using rat brain mitochondrial fractions.  Methyl mercury (MeHg), an important environmental toxicant is implicated in neurological disorders such as Hunter-Russell syndrome and Autism. Therefore, the present work is in search of new drugs that can alleviate MeHg toxicity. In this connection, Tagara, an ayurvedic drug is used for assessing its neuro protective effect against MeHg toxicity. In the present study, we assessed the phytochemical contents of Tagara by colorimetric and HPLC analyses. The neuroprotective effect of Tagara on MeHg induced neurotoxicity was measured in terms of viability by MTT assay and oxidative stress in terms of catalase activity, glutathione and thiobarbituric acid reactive substance levels. Further, the chelating effect of Tagara towards MeHg was performed to identify the molecular mechanism. Statistical analysis was done by statistical package for social sciences (SPSS) version 16.0. The results demonstrated that Tagara contains significant amounts of phenols and flavonoids. Also, HPLC analysis of Tagara revealed the presence of essential oils such as hydroxyvalerenic and valerenic acids. Our results demonstrated that exposure of rat brain mitochondrial fractions to MeHg resulted in a dose dependent death in MTT assay and IC50 value was found to be 10 μM. However, a 250 μg dose of Tagara effectively prevented MeHg induced mitochondrial damage. The oxidative stress caused by MeHg results in elevated levels of reactive oxygen species as evidenced by elevated TBARS (Thiobarbituric acid-reactive substances) levels and diminished catalase enzyme activity and glutathione content. However, Tagara at 250 μg concentration offsets these alterations caused by MeHg. Further, Tagara also diminished GSH oxidation caused by MeHg, confirming its chelating effect, one of the molecular mechanisms that triggers protection against oxidative damage. Our results revealed that MeHg induced toxicity is predominantly mediated through oxidative stress mechanism and the propensity of Tagara to abolish such reactions. Hence, we propose that Tagara with a source of potential neuroprotectants may be a useful approach to alleviate MeHg associated neurotoxicity. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/25852770  Title: The neurotoxic effects of ampicillin-associated gut bacterial imbalances compared to those of orally administered propionic acid in the etiology of persistent autistic features in rat pups: effects of various dietary regimens.  A healthy gut with normal intestinal microflora is completely disrupted by oral antibiotics. The byproducts of harmful gut bacteria can interfere with brain development and may contribute to autism. Strategies to improve the gut microflora profile through dietary modification may help to alleviate gut disorders in autistic patients. Sixty young male western albino rats were divided into six equal groups. The first group served as the control; the second group was given an oral neurotoxic dose of propionic (PPA) (250 mg/kg body weight/day) for three days. The third group received an orogastric dose of ampicillin (50 mg/kg for three weeks) with a standard diet. Groups 4, 5 and 6 were given an orogastric dose of ampicillin and fed high-carbohydrate, high-protein and high-lipid diets, respectively, for 10 weeks. Biochemical parameters related to oxidative stress were investigated in brain homogenates from each group. The microbiology results revealed descriptive changes in the fecal microbiota of rats treated with ampicillin either alone or with the three dietary regimens. The results of PPA acid and ampicillin treatment showed significant increases in lipid peroxidation and catalase with decreases in glutathione and potassium compared with levels in the control group. A protein-rich diet was effective at restoring the glutathione level, while the carbohydrate-rich diet recovered lipid peroxidation and catalase activity. In addition, the three dietary regimens significantly increase the potassium level in the brain tissue of the test animals. Lactate dehydrogenase was remarkably elevated in all groups relative to the control. No outstanding effects were observed in glutathione S-transferase and creatine kinase. The changes observed in the measured parameters reflect the neurotoxic effects of PPA and ampicillin. Lipid peroxide and catalase activity and the levels of glutathione and potassium are satisfactory biomarkers of PPA and ampicillin neurotoxicity. Based on the effects of the three dietary regimens, a balanced diet can protect against PPA or ampicillin-induced neurotoxicity that might induce autistic traits. These outcomes will help efforts directed at controlling the prevalence of autism, a disorder that has recently been associated with PPA neurotoxicity. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/25732953  Title: Astaxanthin improves behavioral disorder and oxidative stress in prenatal valproic acid-induced mice model of autism.  Prenatal exposure to valproic acid on gestational day 12.5 may lead to the impaired behavior in the offspring, which is similar to the human autistic symptoms. To the contrary, astaxanthin shows neuroprotective effect by its antioxidant mechanism. We aimed to (i) develop mice model of autism and (ii) investigate the effect of astaxanthin on such model animals. Valproic acid (600 mg/kg) was administered intraperitoneally to the pregnant mice on gestational day 12.5. Prenatal valproic acid-exposed mice were divided into 2 groups on postnatal day 25 and astaxanthin (2mg/kg) was given to the experimental group (VPA\_AST, n=10) while saline was given to the control group (VPA, n=10) for 4 weeks. Behavioral test including social interaction, open field and hot-plate were conducted on postnatal day 25 and oxidative stress markers such as lipid peroxidation, advanced protein oxidation product, nitric oxide, glutathione, and activity of superoxide dismutase and catalase were estimated on postnatal day 26 to confirm mice model of autism and on postnatal day 56 to assess the effect of astaxanthin. On postnatal day 25, prenatal valproic acid-exposed mice exhibited (i) delayed eye opening (ii) longer latency to respond painful stimuli, (iii) poor sociability and social novelty and (iv) high level of anxiety. In addition, an increased level of oxidative stress was found by determining different oxidative stress markers. Treatment with astaxanthin significantly (p<0.05) improved the behavioral disorder and reduced the oxidative stress in brain and liver. In conclusion, prenatal exposure to valproic day in pregnant mice leads to the development of autism-like features. Astaxanthin improves the impaired behavior in animal model of autism presumably by its antioxidant activity. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/24007566  Title: Evidence for differential alternative splicing in blood of young boys with autism spectrum disorders.  Since RNA expression differences have been reported in autism spectrum disorder (ASD) for blood and brain, and differential alternative splicing (DAS) has been reported in ASD brains, we determined if there was DAS in blood mRNA of ASD subjects compared to typically developing (TD) controls, as well as in ASD subgroups related to cerebral volume. RNA from blood was processed on whole genome exon arrays for 2-4-year-old ASD and TD boys. An ANCOVA with age and batch as covariates was used to predict DAS for ALL ASD (n=30), ASD with normal total cerebral volumes (NTCV), and ASD with large total cerebral volumes (LTCV) compared to TD controls (n=20). A total of 53 genes were predicted to have DAS for ALL ASD versus TD, 169 genes for ASD\_NTCV versus TD, 1 gene for ASD\_LTCV versus TD, and 27 genes for ASD\_LTCV versus ASD\_NTCV. These differences were significant at P <0.05 after false discovery rate corrections for multiple comparisons (FDR <5% false positives). A number of the genes predicted to have DAS in ASD are known to regulate DAS (SFPQ, SRPK1, SRSF11, SRSF2IP, FUS, LSM14A). In addition, a number of genes with predicted DAS are involved in pathways implicated in previous ASD studies, such as ROS monocyte/macrophage, Natural Killer Cell, mTOR, and NGF signaling. The only pathways significant after multiple comparison corrections (FDR <0.05) were the Nrf2-mediated reactive oxygen species (ROS) oxidative response (superoxide dismutase 2, catalase, peroxiredoxin 1, PIK3C3, DNAJC17, microsomal glutathione S-transferase 3) and superoxide radical degradation (SOD2, CAT). These data support differences in alternative splicing of mRNA in blood of ASD subjects compared to TD controls that differ related to head size. The findings are preliminary, need to be replicated in independent cohorts, and predicted alternative splicing differences need to be confirmed using direct analytical methods. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/22708999  Title: Glutathione-related factors and oxidative stress in autism, a review.  Autism spectrum disorders are complex neuro-developmental disorders whose neurobiology is proposed to be associated with oxidative stress which is induced by reactive oxygen species. The process of oxidative stress can be a target for therapeutic interventions. In this study, we aimed to review the role of oxidative stress, plasma glutathione (GSH), and related factors as the potential sources of damage to the brain as well as the possible related factors which reduce the oxidative stress. Methylation capacity, sulfates level, and the total glutathione level are decreased in autism. On the other hand, both oxidized glutathione and the ratio of oxidized to reduced glutathione are increased in autism. In addition, the activity of glutathione peroxidase, superoxide dismutase, and catalase, as a part of the antioxidative stress system are decreased. The current literature suggests an imbalance of oxidative and anti-oxidative stress systems in autism. Glutathione is involved in neuro-protection against oxidative stress and neuro-inflammation in autism by improving the anti-oxidative stress system. Decreasing the oxidative stress might be a potential treatment for autism. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/22531301  Title: Etiology of autistic features: the persisting neurotoxic effects of propionic acid.  Recent clinical observations suggest that certain gut and dietary factors may transiently worsen symptoms in autism. Propionic acid (PA) is a short chain fatty acid and an important intermediate of cellular metabolism. Although PA has several beneficial biological effects, its accumulation is neurotoxic. Two groups of young Western albino male rats weighing about 45 to 60 grams (approximately 21 days old) were used in the present study. The first group consisted of oral buffered PA-treated rats that were given a neurotoxic dose of 250 mg/kg body weight/day for three days, n = eight; the second group of rats were given only phosphate buffered saline and used as a control. Biochemical parameters representing oxidative stress, energy metabolism, neuroinflammation, neurotransmission, and apoptosis were investigated in brain homogenates of both groups. Biochemical analyses of brain homogenates from PA-treated rats showed an increase in oxidative stress markers (for example, lipid peroxidation), coupled with a decrease in glutathione (GSH) and glutathione peroxidase (GPX) and catalase activities. Impaired energy metabolism was ascertained through the decrease of lactate dehydrogenase and activation of creatine kinase (CK). Elevated IL-6, TNFα, IFNγ and heat shock protein 70 (HSP70) confirmed the neuroinflammatory effect of PA. Moreover, elevation of caspase3 and DNA fragmentation proved the pro-apoptotic and neurotoxic effect of PA to rat pups By comparing the results obtained with those from animal models of autism or with clinical data on the biochemical profile of autistic patients, this study showed that the neurotoxicity of PA as an environmental factor could play a central role in the etiology of autistic biochemical features. |
| Catalase |
| Url: https://pubmed.ncbi.nlm.nih.gov/22322665  Title: Bacopa monniera (L.) Wettst ameliorates behavioral alterations and oxidative markers in sodium valproate induced autism in rats.  Early prenatal or post natal exposure to environmental insults such as valproic acid (VPA), thalidomide and ethanol could induce behavioral alterations similar to autistic symptoms. Bacopa monniera, a renowned plant in ayurvedic medicine is useful in several neurological disorders. The purpose of the present study was to evaluate the effect of B. monniera on VPA induced autism. On 12.5 day of gestation the female pregnant rats were divided into control and VPA treated groups. They were administered saline/VPA (600 mg/kg, i.p.) respectively and allowed to raise their own litters. Group I-male pups of saline treated mothers. On postnatal day (PND) 21 VPA induced autistic male pups were divided into two groups (n = 6); Group II-received saline and Group III-received B. monniera (300 mg/kg/p.o.) from PND 21-35. Behavioral tests (nociception, locomotor activity, exploratory activity, anxiety and social behavior) were performed in both adolescence (PND 30-40) and adulthood (PND 90-110) period. At the end of behavioral testing animals were sacrificed, brain was isolated for biochemical estimations (serotonin, glutathione, catalase and nitric oxide) and histopathological examination. Induction of autism significantly affected normal behavior, increased oxidative stress and serotonin level, altered histoarchitecture of cerebellum (decreased number of purkinje cells, neuronal degeneration and chromatolysis) when compared with normal control group. Treatment with B. monniera significantly (p < 0.05) improved behavioral alterations, decreased oxidative stress markers and restored histoarchitecture of cerebellum. In conclusion, the present study suggests that B. monniera ameliorates the autistic symptoms possibly due to its anti-anxiety, antioxidant and neuro-protective activity. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/36454503  Title: Partial changes in apoptotic pathways in hippocampus and hypothalamus of Cc2d1a heterozygous.  Alterations in the apoptosis pathway have been linked to changes in serotonin levels seen in autistic patients. Cc2d1a is a repressor of the HTR1A gene involved in the serotonin pathway. The hippocampus and hypothalamus of Cc2d1a ± mice were analyzed for the expression of apoptosis markers (caspase 3, 8 and 9). Gender differences were observed in the expression levels of the three caspases consistent with some altered activity in the open-field assay. The number of apoptotic cells was significantly increased. We concluded that apoptotic pathways are only partially affected in the pathogenesis of the Cc2d1a heterozygous mouse model. A) Apoptosis is suppressed because the cell does not receive a death signal, or the receptor cannot activate the caspase 8 pathway despite the death signal. B) Since Caspase 8 and Caspase 3 expression is downregulated in our mouse model, the mechanism of apoptosis is not activated. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/34132974  Title: Loss of CC2D1A in Glutamatergic Neurons Results in Autistic-Like Features in Mice.  Biallelic loss-of-function mutations in Coiled-coil and C2 domain containing 1A (CC2D1A) cause autosomal recessive intellectual disability, sometimes comorbid with other neurodevelopmental disabilities, such as autism spectrum disorder (ASD) and seizures. We recently reported that conditional deletion of Cc2d1a in glutamatergic neurons of the postnatal mouse forebrain leads to impaired hippocampal synaptic plasticity and cognitive function. However, the pathogenic origin of the autistic features of CC2D1A deficiency remains elusive. Here, we confirmed that CC2D1A is highly expressed in the cortical zones during embryonic development. Taking advantage of Cre-LoxP-mediated gene deletion strategy, we generated a novel line of Cc2d1a conditional knockout (cKO) mice by crossing floxed Cc2d1a mice with Emx1-Cre mice, in which CC2D1A is ablated specifically in glutamatergic neurons throughout all embryonic and adult stages. We found that CC2D1A deletion leads to a trend toward decreased number of cortical progenitor cells at embryonic day 12.5 and alters the cortical thickness on postnatal day 10. In addition, male Cc2d1a cKO mice display autistic-like phenotypes including self-injurious repetitive grooming and aberrant social interactions. Loss of CC2D1A also results in decreased complexity of apical dendritic arbors of medial prefrontal cortex (mPFC) layer V pyramidal neurons and increased synaptic excitation/inhibition (E/I) ratio in the mPFC. Notably, chronic treatment with minocycline rescues behavioral and morphological abnormalities, as well as E/I changes, in male Cc2d1a cKO mice. Together, these findings indicate that male Cc2d1a cKO mice recapitulate autistic-like phenotypes of human disorder and suggest that minocycline has both structural and functional benefits in treating ASD. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/34048600  Title: Digging behavior discrimination test to probe burrowing and exploratory digging in male and female mice.  Digging behavior is often used to test motor function and repetitive behaviors in mice. Different digging paradigms have been developed for behaviors related to anxiety and compulsion in mouse lines generated to recapitulate genetic mutations leading to psychiatric and neurological disorders. However, the interpretation of these tests has been confounded by the difficulty of determining the motivation behind digging in mice. Digging is a naturalistic mouse behavior that can be focused toward different goals, that is foraging for food, burrowing for shelter, burying objects, or even for recreation as has been shown for dogs, ferrets, and human children. However, the interpretation of results from current testing protocols assumes the motivation behind the behavior often concluding that increased digging is a repetitive or compulsive behavior. We asked whether providing a choice between different types of digging activities would increase sensitivity to assess digging motivation. Here, we present a test to distinguish between burrowing and exploratory digging in mice. We found that mice prefer burrowing when the option is available. When food restriction was used to promote a switch from burrowing to exploration, males readily switched from burrowing to digging outside, while females did not. In addition, when we tested a model of intellectual disability and autism spectrum disorder that had shown inconsistent results in the marble burying test, the Cc2d1a conditional knockout mouse, we found greatly reduced burrowing only in males. Our findings indicate that digging is a nuanced motivated behavior and suggest that male and female rodents may perform it differently. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/33414502  Title: Role of phosphodiesterases in the pathophysiology of neurodevelopmental disorders.  Phosphodiesterases (PDEs) are enzymes involved in the homeostasis of both cAMP and cGMP. They are members of a family of proteins that includes 11 subfamilies with different substrate specificities. Their main function is to catalyze the hydrolysis of cAMP, cGMP, or both. cAMP and cGMP are two key second messengers that modulate a wide array of intracellular processes and neurobehavioral functions, including memory and cognition. Even if these enzymes are present in all tissues, we focused on those PDEs that are expressed in the brain. We took into consideration genetic variants in patients affected by neurodevelopmental disorders, phenotypes of animal models, and pharmacological effects of PDE inhibitors, a class of drugs in rapid evolution and increasing application to brain disorders. Collectively, these data indicate the potential of PDE modulators to treat neurodevelopmental diseases characterized by learning and memory impairment, alteration of behaviors associated with depression, and deficits in social interaction. Indeed, clinical trials are in progress to treat patients with Alzheimer's disease, schizophrenia, depression, and autism spectrum disorders. Among the most recent results, the application of some PDE inhibitors (PDE2A, PDE3, PDE4/4D, and PDE10A) to treat neurodevelopmental diseases, including autism spectrum disorders and intellectual disability, is a significant advance, since no specific therapies are available for these disorders that have a large prevalence. In addition, to highlight the role of several PDEs in normal and pathological neurodevelopment, we focused here on the deregulation of cAMP and/or cGMP in Down Syndrome, Fragile X Syndrome, Rett Syndrome, and intellectual disability associated with the CC2D1A gene. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/33287601  Title: Novel alterations of CC2D1A as a candidate gene in a Turkish sample of patients with autism spectrum disorder.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder with large genetic background, but identification of pathogenic variants has proceeded slowly because hundreds of loci are involved in this complex disorder. CC2D1A gene firstly associated with the intellectual disability (ID) in a family with a large deletion. We aimed to contribute to the literature by sequencing this gene and by this way we report novel CC2D1A variations in patients with ASD. Forty families who have a child with a diagnosis of ASD were enrolled to the study. DNA samples were obtained from each family member. Bidirectional CC2D1A gene sequencing was performed with CEQ Cycle Sequencing Kit, and the products were analyzed on the Beckman CEQ 8000. All of the genetic analysis was conducted in Erciyes University Genome and Stem Cell Center (GENKOK). According to the sequencing results, we defined new alterations in this gene with two SNPs in exon 15 and 19 (rs747172992 and rs1364074600) in our patients. We found a pathogenic variant in one patient. This variant was located in the acceptor region. Six of the variants were missense mutations. Additionally, six different benign variants were detected in 30 patients; however, they were not associated with ASD. Two patients carried multiple rare variants. In vitro and in vivo functional analysis with this gene will help to understand its contribution to ASD pathogenesis. Future studies may help to elucidate the underlying biological mechanisms of these variants leading to the autism phenotype. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/32514154  Title: A heritable profile of six miRNAs in autistic patients and mouse models.  Autism spectrum disorder (ASD) is a group of developmental pathologies that impair social communication and cause repetitive behaviors. The suggested roles of noncoding RNAs in pathology led us to perform a comparative analysis of the microRNAs expressed in the serum of human ASD patients. The analysis of a cohort of 45 children with ASD revealed that six microRNAs (miR-19a-3p, miR-361-5p, miR-3613-3p, miR-150-5p, miR-126-3p, and miR-499a-5p) were expressed at low to very low levels compared to those in healthy controls. A similar but less pronounced decrease was registered in the clinically unaffected parents of the sick children and in their siblings but never in any genetically unrelated control. Results consistent with these observations were obtained in the blood, hypothalamus and sperm of two of the established mouse models of ASD: valproic acid-treated animals and Cc2d1a+/- heterozygotes. In both instances, the same characteristic miRNA profile was evidenced in the affected individuals and inherited together with disease symptoms in the progeny of crosses with healthy animals. The consistent association of these genetic regulatory changes with the disease provides a starting point for evaluating the changes in the activity of the target genes and, thus, the underlying mechanism(s). From the applied societal and medical perspectives, once properly confirmed in large cohorts, these observations provide tools for the very early identification of affected children and progenitors. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/31721010  Title: Disregulation of Autophagy in the Transgenerational Cc2d1a Mouse Model of Autism.  Autism spectrum disorder (ASD) is a heterogeneously childhood neurodevelopmental disorder, believed to be under development of various genetic and environmental factors. Autophagy and related pathways have also been implicated in the etiology of ASD. We aimed to investigate autophagic markers by generating the transgenerational inheritance of ASD-like behaviors in the Cc2d1a animal model of ASD. Cc2d1a (+/-) mouse model of ASD was built in two different groups by following three generations. After behavior test, bilateral hippocampus was sliced. Western Blot assay and quantitative real-time polymerase chain reaction (QRT-PCR) were used for measurement of LC3 and Beclin-1 as key regulators of autophagy. All of the animal and laboratory studies were conducted in the Erciyes University Genome and Stem Cell Center (GENKOK). Significant LC3 and Beclin-1 mRNA expression levels were observed in mouse hippocampus between groups and generations. Western blot confirmed the changes of the proteins in the hippocampus. LC3 expressions were increased for females and decreased for males compared to the control group. Beclin-1 expression levels were found to be significantly decreased in males and females compared to controls. This study could help explain a new pathway of autophagy in ASD mouse models. Future animal studies need to investigate sex differences in mouse modeling autism-relevant genes like CC2D1A. We anticipate our results to be a starting point for more comprehensive autophagy studies in this mouse model of ASD. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/30732858  Title: Male-Specific cAMP Signaling in the Hippocampus Controls Spatial Memory Deficits in a Mouse Model of Autism and Intellectual Disability.  The prevalence of neurodevelopmental disorders is biased toward male individuals, with male-to-female ratios of 2:1 in intellectual disability and 4:1 in autism spectrum disorder. However, the molecular mechanisms of such bias remain unknown. While characterizing a mouse model for loss of the signaling scaffold coiled-coil and C2 domain-containing protein 1A (CC2D1A), which is mutated in intellectual disability and autism spectrum disorder, we identified biochemical and behavioral differences between male and female mice, and explored whether CC2D1A controls male-specific intracellular signaling. CC2D1A is known to regulate phosphodiesterase 4D (PDE4D), which regulates cyclic adenosine monophosphate (cAMP) signaling. We tested for activation of PDE4D and downstream signaling molecules in the hippocampus of Cc2d1a-deficient mice. We then performed behavioral studies in female mice to analyze learning and memory, and then targeted PDE4D activation with a PDE4D inhibitor to define how changes in cAMP levels affect behavior in male and female mice. We found that in Cc2d1a-deficient male mice PDE4D is hyperactive, leading to a reduction in cAMP response element binding protein signaling, but this molecular deficit is not present in female mice. Cc2d1a-deficient male mice show a deficit in spatial memory, which is not present in Cc2d1a-deficient female mice. Restoring PDE4D activity using an inhibitor rescues cognitive deficits in male mice but has no effect on female mice. Our findings show that CC2D1A regulates cAMP intracellular signaling in a male-specific manner in the hippocampus, leading to male-specific cognitive deficits. We propose that male-specific signaling mechanisms are involved in establishing sex bias in neurodevelopmental disorders. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/29552027  Title: Loss of the Intellectual Disability and Autism Gene Cc2d1a and Its Homolog Cc2d1b Differentially Affect Spatial Memory, Anxiety, and Hyperactivity.  Hundreds of genes are mutated in non-syndromic intellectual disability (ID) and autism spectrum disorder (ASD), with each gene often involved in only a handful of cases. Such heterogeneity can be daunting, but rare recessive loss of function (LOF) mutations can be a good starting point to provide insight into the mechanisms of neurodevelopmental disease. Biallelic LOF mutations in the signaling scaffold CC2D1A cause a rare form of autosomal recessive ID, sometimes associated with ASD and seizures. In parallel, we recently reported that Cc2d1a-deficient mice present with cognitive and social deficits, hyperactivity and anxiety. In Drosophila, loss of the only ortholog of Cc2d1a, lgd, is embryonically lethal, while in vertebrates, Cc2d1a has a homolog Cc2d1b which appears to be compensating, indicating that Cc2d1a and Cc2d1b have a redundant function in humans and mice. Here, we generate an allelic series of Cc2d1a and Cc2d1b LOF to determine the relative role of these genes during behavioral development. We generated Cc2d1b knockout (KO), Cc2d1a/1b double heterozygous and double KO mice, then performed behavioral studies to analyze learning and memory, social interactions, anxiety, and hyperactivity. We found that Cc2d1a and Cc2d1b have partially overlapping roles. Overall, loss of Cc2d1b is less severe than loss of Cc2d1a, only leading to cognitive deficits, while Cc2d1a/1b double heterozygous animals are similar to Cc2d1a-deficient mice. These results will help us better understand the deficits in individuals with CC2D1A mutations, suggesting that recessive CC2D1B mutations and trans-heterozygous CC2D1A and CC2D1B mutations could also contribute to the genetics of ID. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/26826102  Title: Cc2d1a Loss of Function Disrupts Functional and Morphological Development in Forebrain Neurons Leading to Cognitive and Social Deficits.  Loss-of-function (LOF) mutations in CC2D1A cause a spectrum of neurodevelopmental disorders, including intellectual disability, autism spectrum disorder, and seizures, identifying a critical role for this gene in cognitive and social development. CC2D1A regulates intracellular signaling processes that are critical for neuronal function, but previous attempts to model the human LOF phenotypes have been prevented by perinatal lethality in Cc2d1a-deficient mice. To overcome this challenge, we generated a floxed Cc2d1a allele for conditional removal of Cc2d1a in the brain using Cre recombinase. While removal of Cc2d1a in neuronal progenitors using Cre expressed from the Nestin promoter still causes death at birth, conditional postnatal removal of Cc2d1a in the forebrain via calcium/calmodulin-dependent protein kinase II-alpha (CamKIIa) promoter-driven Cre generates animals that are viable and fertile with grossly normal anatomy. Analysis of neuronal morphology identified abnormal cortical dendrite organization and a reduction in dendritic spine density. These animals display deficits in neuronal plasticity and in spatial learning and memory that are accompanied by reduced sociability, hyperactivity, anxiety, and excessive grooming. Cc2d1a conditional knockout mice therefore recapitulate features of both cognitive and social impairment caused by human CC2D1A mutation, and represent a model that could provide much needed insights into the developmental mechanisms underlying nonsyndromic neurodevelopmental disorders. |
| CC2D1A |
| Url: https://pubmed.ncbi.nlm.nih.gov/26782176  Title: The roles of CC2D1A and HTR1A gene expressions in autism spectrum disorders.  Classical autism belongs to a group of heterogeneous disorders known as autism spectrum disorders (ASD). Autism is defined as a neurodevelopmental disorder, characterized by repetitive stereotypic behaviors or restricted interests, social withdrawal, and communication deficits. Numerous susceptibility genes and chromosomal abnormalities have been reported in association with autism but the etiology of this disorder is unknown in many cases. CC2D1A gene has been linked to mental retardation (MR) in a family with a large deletion before. Intellectual disability (ID) is a common feature of autistic cases. Therefore we aimed to investigate the expressions of CC2D1A and HTR1A genes with the diagnosis of autism in Turkey. Forty-four autistic patients (35 boys, 9 girls) and 27 controls were enrolled and obtained whole blood samples to isolate RNA samples from each participant. CC2D1A and HTR1A gene expressions were assessed by quantitative Real-Time PCR (qRT-PCR) in Genome and Stem Cell Center, Erciyes University. Both expressions of CC2D1A and HTR1A genes studied on ASD cases and controls were significantly different (p < 0.001). The expression of HTR1A was undetectable in the ASD samples. Comparison of ID and CC2D1A gene expression was also found statistically significant (p = 0.028). CC2D1A gene expression may be used as a candidate gene for ASD cases with ID. Further studies are needed to investigate the potential roles of these CC2D1A and HTR1A genes in their related pathways in ASD. |
| CHD1, CHD2, CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29962935  Title: Chromatin Remodeling Proteins in Epilepsy: Lessons From CHD2-Associated Epilepsy.  The chromodomain helicase DNA-binding (CHD) family of proteins are ATP-dependent chromatin remodelers that contribute to the reorganization of chromatin structure and deposition of histone variants necessary to regulate gene expression. CHD proteins play an important role in neurodevelopment, as pathogenic variants in CHD1, CHD2, CHD4, CHD7 and CHD8 have been associated with a range of neurological phenotypes, including autism spectrum disorder (ASD), intellectual disability (ID) and epilepsy. Pathogenic variants in CHD2 are associated with developmental epileptic encephalopathy (DEE) in humans, however little is known about how these variants contribute to this disorder. Of the nine CHD family members, CHD2 is the only one that leads to a brain-restricted phenotype when disrupted in humans. This suggests that despite being expressed ubiquitously, CHD2 has a unique role in human brain development and function. In this review, we will discuss the phenotypic spectrum of patients with pathogenic variants in CHD2, current animal models of CHD2 deficiency, and the role of CHD2 in proliferation, neurogenesis, neuronal differentiation, chromatin remodeling and DNA-repair. We also consider how CHD2 depletion can affect each of these biological mechanisms and how these defects may underpin neurodevelopmental disorders including epilepsy. |
| CHD2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30344048  Title: Chd2 Is Necessary for Neural Circuit Development and Long-Term Memory.  Considerable evidence suggests loss-of-function mutations in the chromatin remodeler CHD2 contribute to a broad spectrum of human neurodevelopmental disorders. However, it is unknown how CHD2 mutations lead to impaired brain function. Here we report mice with heterozygous mutations in Chd2 exhibit deficits in neuron proliferation and a shift in neuronal excitability that included divergent changes in excitatory and inhibitory synaptic function. Further in vivo experiments show that Chd2+/- mice displayed aberrant cortical rhythmogenesis and severe deficits in long-term memory, consistent with phenotypes observed in humans. We identified broad, age-dependent transcriptional changes in Chd2+/- mice, including alterations in neurogenesis, synaptic transmission, and disease-related genes. Deficits in interneuron density and memory caused by Chd2+/- were reproduced by Chd2 mutation restricted to a subset of inhibitory neurons and corrected by interneuron transplantation. Our results provide initial insight into how Chd2 haploinsufficiency leads to aberrant cortical network function and impaired memory. |
| CHD2, CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30277262  Title: Autism-linked CHD gene expression patterns during development predict multi-organ disease phenotypes.  Recent large-scale exome sequencing studies have identified mutations in several members of the CHD (Chromodomain Helicase DNA-binding protein) gene family in neurodevelopmental disorders. Mutations in the CHD2 gene have been linked to developmental delay, intellectual disability, autism and seizures, CHD8 mutations to autism and intellectual disability, whereas haploinsufficiency of CHD7 is associated with executive dysfunction and intellectual disability. In addition to these neurodevelopmental features, a wide range of other developmental defects are associated with mutants of these genes, especially with regards to CHD7 haploinsufficiency, which is the primary cause of CHARGE syndrome. Whilst the developmental expression of CHD7 has been reported previously, limited information on the expression of CHD2 and CHD8 during development is available. Here, we compare the expression patterns of all three genes during mouse development directly. We find high, widespread expression of these genes at early stages of development that gradually becomes restricted during later developmental stages. Chd2 and Chd8 are widely expressed in the developing central nervous system (CNS) at all stages of development, with moderate expression remaining in the neocortex, hippocampus, olfactory bulb and cerebellum of the postnatal brain. Similarly, Chd7 expression is seen throughout the CNS during late embryogenesis and early postnatal development, with strong enrichment in the cerebellum, but displays low expression in the cortex and neurogenic niches in early life. In addition to expression in the brain, novel sites of Chd2 and Chd8 expression are reported. These findings suggest additional roles for these genes in organogenesis and predict that mutation of these genes may predispose individuals to a range of other, non-neurological developmental defects. |
| CHD2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25786798  Title: CHD2 is Required for Embryonic Neurogenesis in the Developing Cerebral Cortex.  Chromodomain helicase DNA-binding protein 2 (CHD2) has been associated with a broad spectrum of neurodevelopmental disorders, such as autism spectrum disorders and intellectual disability. However, it is largely unknown whether and how CHD2 is involved in brain development. Here, we demonstrate that CHD2 is predominantly expressed in Pax6(+) radial glial cells (RGs) but rarely expressed in Tbr2(+) intermediate progenitors (IPs). Importantly, the suppression of CHD2 expression inhibits the self-renewal of RGs and increases the generation of IPs and the production of neurons. CHD2 mediates these functions by directly binding to the genomic region of repressor element 1-silencing transcription factor (REST), thereby regulating the expression of REST. Furthermore, the overexpression of REST rescues the defect in neurogenesis caused by CHD2 knockdown. Taken together, these findings demonstrate an essential role of CHD2 in the maintenance of the RGs self-renewal levels, the subsequent generation of IPs, and neuronal output during neurogenesis in cerebral cortical development, suggesting that inactivation of CHD2 during neurogenesis might contribute to abnormal neurodevelopment. |
| CHD3, CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31146125  Title: Regulation of neuronal connectivity in the mammalian brain by chromatin remodeling.  Precise temporal and spatial control of gene expression is essential for brain development. Besides DNA sequence-specific transcription factors, epigenetic factors play an integral role in the control of gene expression in neurons. Among epigenetic mechanisms, chromatin remodeling enzymes have emerged as essential to the control of neural circuit assembly and function in the brain. Here, we review recent studies on the roles and mechanisms of the chromodomain-helicase-DNA-binding (Chd) family of chromatin remodeling enzymes in the regulation of neuronal morphogenesis and connectivity in the mammalian brain. We explore the field through the lens of Chd3, Chd4, and Chd5 proteins, which incorporate into the nucleosome remodeling and deacetylase (NuRD) complex, and the related proteins Chd7 and Chd8, implicated in the pathogenesis of intellectual disability and autism spectrum disorders. These studies have advanced our understanding of the mechanisms that regulate neuronal connectivity in brain development and neurodevelopmental disorders of cognition. |
| CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36914558  Title: Pervasive cortical and white matter anomalies in a mouse model for CHARGE syndrome.  CHARGE (Coloboma of the eye, Heart defects, Atresia of the choanae, Retardation of growth, Genital anomalies and Ear abnormalities) syndrome is a disorder caused by mutations in the gene encoding CHD7, an ATP dependent chromatin remodelling factor, and is characterised by a diverse array of congenital anomalies. These include a range of neuroanatomical comorbidities which likely underlie the varied neurodevelopmental disorders associated with CHARGE syndrome, which include intellectual disability, motor coordination deficits, executive dysfunction, and autism spectrum disorder. Cranial imaging studies are challenging in CHARGE syndrome patients, but high-throughput magnetic resonance imaging (MRI) techniques in mouse models allow for the unbiased identification of neuroanatomical defects. Here, we present a comprehensive neuroanatomical survey of a Chd7 haploinsufficient mouse model of CHARGE syndrome. Our study uncovered widespread brain hypoplasia and reductions in white matter volume across the brain. The severity of hypoplasia appeared more pronounced in posterior areas of the neocortex compared to anterior regions. We also perform the first assessment of white matter tract integrity in this model through diffusion tensor imaging (DTI) to assess the potential functional consequences of widespread reductions in myelin, which suggested the presence of white matter integrity defects. To determine if white matter alterations correspond to cellular changes, we quantified oligodendrocyte lineage cells in the postnatal corpus callosum, uncovering reduced numbers of mature oligodendrocytes. Together, these results present a range of promising avenues of focus for future cranial imaging studies in CHARGE syndrome patients. |
| CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34088660  Title: The CHD8/CHD7/Kismet family links blood-brain barrier glia and serotonin to ASD-associated sleep defects.  Sleep disturbances in autism and neurodevelopmental disorders are common and adversely affect patient's quality of life, yet the underlying mechanisms are understudied. We found that individuals with mutations in CHD8, among the highest-confidence autism risk genes, or CHD7 suffer from disturbed sleep maintenance. These defects are recapitulated in Drosophila mutants affecting kismet, the sole CHD8/CHD7 ortholog. We show that Kismet is required in glia for early developmental and adult sleep architecture. This role localizes to subperineurial glia constituting the blood-brain barrier. We demonstrate that Kismet-related sleep disturbances are caused by high serotonin during development, paralleling a well-established but genetically unsolved autism endophenotype. Despite their developmental origin, Kismet's sleep architecture defects can be reversed in adulthood by a behavioral regime resembling human sleep restriction therapy. Our findings provide fundamental insights into glial regulation of sleep and propose a causal mechanistic link between the CHD8/CHD7/Kismet family, developmental hyperserotonemia, and autism-associated sleep disturbances. |
| CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32493418  Title: Should autism spectrum disorder be considered part of CHARGE syndrome? A cross-sectional study of 46 patients.  Behavioral problems are an important issue for people with CHARGE syndrome. The similarity of their behavioral traits with those of people with autism raises questions. In a large national cross-sectional study, we used specific standardized tools for diagnosing autism (Autism Diagnostic Interview-Revised and Diagnostic and Statistical Manual of Mental Disorders, 5th edition, DSM-5) and evaluating behavioral disorders (Developmental Behavior Checklist-Parents, DBC-P) to investigate a series of individuals with CHARGE syndrome, defined by Verloes's criteria. We evaluated their adaptive functioning level and sensory particularities and extracted several data items from medical files to assess as potential risk factors for autism and/or behavioral disorders. We investigated 64 individuals with CHARGE syndrome (35 females; mean age 10.7 years, SD 7.1 years). Among 46 participants with complete results for the Autism Diagnostic Interview-Revised (ADI-R), 13 (28%) had a diagnosis of autism according to the ADI-R, and 25 (54%) had a diagnosis of autism spectrum disorder (ASD) according to the DSM-5 criteria. The frequency of autistic traits in the entire group was a continuum. We did not identify any risk factor for ASD but found a negative correlation between the ADI-R score and adaptive functioning level. Among 48 participants with data for the DBC-P, 26 (55%) had behavioral disorders, which were more frequent in patients with radiological brain anomalies, impaired adaptive functioning, later independent walking, and more sensory particularities. ASD should be considered to be an independent risk requiring early screening and management in children born with CHARGE syndrome. |
| CHD7, NKX2-2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30108144  Title: Oligodendrocyte precursor survival and differentiation requires chromatin remodeling by Chd7 and Chd8.  Oligodendrocyte precursor cells (OPCs) constitute the main proliferative cells in the adult brain, and deregulation of OPC proliferation-differentiation balance results in either glioma formation or defective adaptive (re)myelination. OPC differentiation requires significant genetic reprogramming, implicating chromatin remodeling. Mounting evidence indicates that chromatin remodelers play important roles during normal development and their mutations are associated with neurodevelopmental defects, with CHD7 haploinsuficiency being the cause of CHARGE syndrome and CHD8 being one of the strongest autism spectrum disorder (ASD) high-risk-associated genes. Herein, we report on uncharacterized functions of the chromatin remodelers Chd7 and Chd8 in OPCs. Their OPC-chromatin binding profile, combined with transcriptome and chromatin accessibility analyses of Chd7-deleted OPCs, demonstrates that Chd7 protects nonproliferative OPCs from apoptosis by chromatin closing and transcriptional repression of p53 Furthermore, Chd7 controls OPC differentiation through chromatin opening and transcriptional activation of key regulators, including Sox10, Nkx2.2, and Gpr17 However, Chd7 is dispensable for oligodendrocyte stage progression, consistent with Chd8 compensatory function, as suggested by their common chromatin-binding profiles and genetic interaction. Finally, CHD7 and CHD8 bind in OPCs to a majority of ASD risk-associated genes, suggesting an implication of oligodendrocyte lineage cells in ASD neurological defects. Our results thus offer new avenues to understand and modulate the CHD7 and CHD8 functions in normal development and disease. |
| CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26586927  Title: Autism-Like Behavior and Epigenetic Changes Associated with Autism as Consequences of In Utero Exposure to Environmental Pollutants in a Mouse Model.  We tested the hypothesis that in utero exposure to heavy metals increases autism-like behavioral phenotypes in adult animals and induces epigenetic changes in genes that have roles in the etiology of autism. Mouse dams were treated with cadmium, lead, arsenate, manganese, and mercury via drinking water from gestational days (E) 1-10. Valproic acid (VPA) injected intraperitoneally once on (E) 8.5 served as a positive control. Young male offspring were tested for behavioral deficits using four standardized behavioral assays. In this study, in utero exposure to heavy metals resulted in multiple behavioral abnormalities that persisted into adulthood. VPA and manganese induced changes in perseverative/impulsive behavior and social dominance behavior, arsenic caused changes only in perseverative/impulsive behavior, and lead induced abnormalities in social interaction in comparison to the control animals. Brain samples from Mn, Pb, and VPA treated and control animals were evaluated for changes in CpG island methylation in promoter regions and associated changes in gene expression. The Chd7 gene, essential for neural crest cell migration and patterning, was found to be hypomethylated in each experimental animal tested compared to water-treated controls. Furthermore, distinct patterns of CpG island methylation yielded novel candidate genes for further investigation. |
| CHD7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23285124  Title: Identification and characterization of FAM124B as a novel component of a CHD7 and CHD8 containing complex.  Mutations in the chromodomain helicase DNA binding protein 7 gene (CHD7) lead to CHARGE syndrome, an autosomal dominant multiple malformation disorder. Proteins involved in chromatin remodeling typically act in multiprotein complexes. We previously demonstrated that a part of human CHD7 interacts with a part of human CHD8, another chromodomain helicase DNA binding protein presumably being involved in the pathogenesis of neurodevelopmental (NDD) and autism spectrum disorders (ASD). Because identification of novel CHD7 and CHD8 interacting partners will provide further insights into the pathogenesis of CHARGE syndrome and ASD/NDD, we searched for additional associated polypeptides using the method of stable isotope labeling by amino acids in cell culture (SILAC) in combination with mass spectrometry. The hitherto uncharacterized FAM124B (Family with sequence similarity 124B) was identified as a potential interaction partner of both CHD7 and CHD8. We confirmed the result by co-immunoprecipitation studies and showed a direct binding to the CHD8 part by direct yeast two hybrid experiments. Furthermore, we characterized FAM124B as a mainly nuclear localized protein with a widespread expression in embryonic and adult mouse tissues. Our results demonstrate that FAM124B is a potential interacting partner of a CHD7 and CHD8 containing complex. From the overlapping expression pattern between Chd7 and Fam124B at murine embryonic day E12.5 and the high expression of Fam124B in the developing mouse brain, we conclude that Fam124B is a novel protein possibly involved in the pathogenesis of CHARGE syndrome and neurodevelopmental disorders. |
| CIC |
| Url: https://pubmed.ncbi.nlm.nih.gov/35203088  Title: Increased expression of SLC25A1/CIC causes an autistic-like phenotype with altered neuron morphology.  N ε-lysine acetylation within the lumen of the endoplasmic reticulum is a recently characterized protein quality control system that positively selects properly folded glycoproteins in the early secretory pathway. Overexpression of the endoplasmic reticulum acetyl-CoA transporter AT-1 in mouse forebrain neurons results in increased dendritic branching, spine formation and an autistic-like phenotype that is attributed to altered glycoprotein flux through the secretory pathway. AT-1 overexpressing neurons maintain the cytosolic pool of acetyl-CoA by upregulation of SLC25A1, the mitochondrial citrate/malate antiporter and ATP citrate lyase, which converts cytosolic citrate into acetyl-CoA. All three genes have been associated with autism spectrum disorder, suggesting that aberrant cytosolic-to-endoplasmic reticulum flux of acetyl-CoA can be a mechanistic driver for the development of autism spectrum disorder. We therefore generated a SLC25A1 neuron transgenic mouse with overexpression specifically in the forebrain neurons. The mice displayed autistic-like behaviours with a jumping stereotypy. They exhibited increased steady-state levels of citrate and acetyl-CoA, disrupted white matter integrity with activated microglia and altered synaptic plasticity and morphology. Finally, quantitative proteomic and acetyl-proteomic analyses revealed differential adaptations in the hippocampus and cortex. Overall, our study reinforces the connection between aberrant cytosolic-to-endoplasmic reticulum acetyl-CoA flux and the development of an autistic-like phenotype. |
| CIC |
| Url: https://pubmed.ncbi.nlm.nih.gov/28288114  Title: Disruption of the ATXN1-CIC complex causes a spectrum of neurobehavioral phenotypes in mice and humans.  Gain-of-function mutations in some genes underlie neurodegenerative conditions, whereas loss-of-function mutations in the same genes have distinct phenotypes. This appears to be the case with the protein ataxin 1 (ATXN1), which forms a transcriptional repressor complex with capicua (CIC). Gain of function of the complex leads to neurodegeneration, but ATXN1-CIC is also essential for survival. We set out to understand the functions of the ATXN1-CIC complex in the developing forebrain and found that losing this complex results in hyperactivity, impaired learning and memory, and abnormal maturation and maintenance of upper-layer cortical neurons. We also found that CIC activity in the hypothalamus and medial amygdala modulates social interactions. Informed by these neurobehavioral features in mouse mutants, we identified five individuals with de novo heterozygous truncating mutations in CIC who share similar clinical features, including intellectual disability, attention deficit/hyperactivity disorder (ADHD), and autism spectrum disorder. Our study demonstrates that loss of ATXN1-CIC complexes causes a spectrum of neurobehavioral phenotypes. |
| CREBBP |
| Url: https://pubmed.ncbi.nlm.nih.gov/35422816  Title: mTOR Signaling Pathway Regulates the Release of Proinflammatory Molecule CCL5 Implicated in the Pathogenesis of Autism Spectrum Disorder.  Autism spectrum disorder (ASD) is a complex pervasive neurodevelopmental disorder and neuroinflammation may contribute to the pathogenesis of ASD. However, the exact mechanisms of abnormal release of proinflammatory mediators in ASD remain poorly understood. This study reports elevated plasma levels of the proinflammatory chemokine (C-C motif) ligand 5 (CCL5) in children with ASD, suggesting an aberrant inflammatory response appearing in the development of ASD. Mining of the expression data of brain or blood tissue from individuals with ASD reveals that mTOR signaling is aberrantly activated in ASD patients. Our in vitro study shows that suppression of mTOR reduces the gene expression and release of CCL5 from human microglia, supporting that CCL5 expression is regulated by mTOR activity. Furthermore, bacterial lipopolysaccharide (LPS)-induced CCL5 expression can be counteracted by siRNA against NF-κB, suggests a determining role of NF-κB in upregulating CCL5 expression. However, a direct regulatory relationship between the NF-κB element and the mTOR signaling pathway was not observed in rapamycin-treated cells. Our results show that the phosphorylated CREB can be induced to suppress CCL5 expression by outcompeting NF-κB in binding to CREB-binding protein (CREBBP) once the mTOR signaling pathway is inhibited. We propose that the activation of mTOR signaling in ASD may induce the suppression of phosphorylation of CREB, which in turn results in the increased binding of CREBBP to NF-κB, a competitor of phosphorylated CREB to drive expression of CCL5. Our study sheds new light on the inflammatory mechanisms of ASD and paves the way for the development of therapeutic strategy for ASD. |
| CREBBP, SATB2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25106414  Title: The clinical significance of small copy number variants in neurodevelopmental disorders.  Despite abundant evidence for pathogenicity of large copy number variants (CNVs) in neurodevelopmental disorders (NDDs), the individual significance of genome-wide rare CNVs <500 kb has not been well elucidated in a clinical context. By high-resolution chromosomal microarray analysis, we investigated the clinical significance of all rare non-polymorphic exonic CNVs sizing 1-500 kb in a cohort of 714 patients with undiagnosed NDDs. We detected 96 rare CNVs <500 kb affecting coding regions, of which 58 (60.4%) were confirmed. 6 of 14 confirmed de novo, one of two homozygous and four heterozygous inherited CNVs affected the known microdeletion regions 17q21.31, 16p11.2 and 2p21 or OMIM morbid genes (CASK, CREBBP, PAFAH1B1, SATB2; AUTS2, NRXN3, GRM8). Two further de novo CNVs affecting single genes (MED13L, CTNND2) were instrumental in delineating novel recurrent conditions. For the first time, we here report exonic deletions of CTNND2 causing low normal IQ with learning difficulties with or without autism spectrum disorder. Additionally, we discovered a homozygous out-of-frame deletion of ACOT7 associated with features comparable to the published mouse model. In total, 24.1% of the confirmed small CNVs were categorised as pathogenic or likely pathogenic (median size 130 kb), 17.2% as likely benign, 3.4% represented incidental findings and 55.2% remained unclear. These results verify the diagnostic relevance of genome-wide rare CNVs <500 kb, which were found pathogenic in ∼2% (14/714) of cases (1.1% de novo, 0.3% homozygous, 0.6% inherited) and highlight their inherent potential for discovery of new conditions. |
| CREBBP |
| Url: https://pubmed.ncbi.nlm.nih.gov/24055786  Title: Acute prenatal exposure to a moderate dose of valproic acid increases social behavior and alters gene expression in rats.  Prenatal exposure to moderate doses of valproic acid (VPA) produces brainstem abnormalities, while higher doses of this teratogen elicit social deficits in the rat. In this pilot study, we examined effects of prenatal exposure to a moderate dose of VPA on behavior and on transcriptomic expression in three brain regions that mediate social behavior. Pregnant Long Evans rats were injected with 350 mg/kg VPA or saline on gestational day 13. A modified social interaction test was used to assess social behavior and social preference/avoidance during early and late adolescence and in adulthood. VPA-exposed animals demonstrated more social investigation and play fighting than control animals. Social investigation, play fighting, and contact behavior also differed as a function of age; the frequency of these behaviors increased in late adolescence. Social preference and locomotor activity under social circumstances were unaffected by treatment or age. Thus, a moderate prenatal dose of VPA produces behavioral alterations that are substantially different from the outcomes that occur following exposure to a higher dose. At adulthood, VPA-exposed subjects exhibited transcriptomic abnormalities in three brain regions: anterior amygdala, cerebellar vermis, and orbitofrontal cortex. A common feature among the proteins encoded by the dysregulated genes was their ability to be modulated by acetylation. Analysis of the expression of individual exons also revealed that genes involved in post-translational modification and epigenetic regulation had particular isoforms that were ubiquitously dysregulated across brain regions. The vulnerability of these genes to the epigenetic effects of VPA may highlight potential mechanisms by which prenatal VPA exposure alters the development of social behavior. |
| CSDE1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31579823  Title: Disruptive variants of CSDE1 associate with autism and interfere with neuronal development and synaptic transmission.  RNA binding proteins are key players in posttranscriptional regulation and have been implicated in neurodevelopmental and neuropsychiatric disorders. Here, we report a significant burden of heterozygous, likely gene-disrupting variants in CSDE1 (encoding a highly constrained RNA binding protein) among patients with autism and related neurodevelopmental disabilities. Analysis of 17 patients identifies common phenotypes including autism, intellectual disability, language and motor delay, seizures, macrocephaly, and variable ocular abnormalities. HITS-CLIP revealed that Csde1-binding targets are enriched in autism-associated gene sets, especially FMRP targets, and in neuronal development and synaptic plasticity-related pathways. Csde1 knockdown in primary mouse cortical neurons leads to an overgrowth of the neurites and abnormal dendritic spine morphology/synapse formation and impaired synaptic transmission, whereas mutant and knockdown experiments in Drosophila result in defects in synapse growth and synaptic transmission. Our study defines a new autism-related syndrome and highlights the functional role of CSDE1 in synapse development and synaptic transmission. |
| CTCF |
| Url: https://pubmed.ncbi.nlm.nih.gov/36945527  Title: TAD Evolutionary and functional characterization reveals diversity in mammalian TAD boundary properties and function.  Topological associating domains (TADs) are self-interacting genomic units crucial for shaping gene regulation patterns. Despite their importance, the extent of their evolutionary conservation and its functional implications remain largely unknown. In this study, we generate Hi-C and ChIP-seq data and compare TAD organization across four primate and four rodent species, and characterize the genetic and epigenetic properties of TAD boundaries in correspondence to their evolutionary conservation. We find that only 14% of all human TAD boundaries are shared among all eight species (ultraconserved), while 15% are human-specific. Ultraconserved TAD boundaries have stronger insulation strength, CTCF binding, and enrichment of older retrotransposons, compared to species-specific boundaries. CRISPR-Cas9 knockouts of two ultraconserved boundaries in mouse models leads to tissue-specific gene expression changes and morphological phenotypes. Deletion of a human-specific boundary near the autism-related AUTS2 gene results in upregulation of this gene in neurons. Overall, our study provides pertinent TAD boundary evolutionary conservation annotations, and showcase the functional importance of TAD evolution. |
| CTCF |
| Url: https://pubmed.ncbi.nlm.nih.gov/35589920  Title: Differential Regulation of Mouse Hippocampal Gene Expression Sex Differences by Chromosomal Content and Gonadal Sex.  Common neurological disorders, like Alzheimer's disease (AD), multiple sclerosis (MS), and autism, display profound sex differences in prevalence and clinical presentation. However, sex differences in the brain with health and disease are often overlooked in experimental models. Sex effects originate, directly or indirectly, from hormonal or sex chromosomal mechanisms. To delineate the contributions of genetic sex (XX v. XY) versus gonadal sex (ovaries v. testes) to the epigenomic regulation of hippocampal sex differences, we used the Four Core Genotypes (FCG) mouse model which uncouples chromosomal and gonadal sex. Transcriptomic and epigenomic analyses of ~ 12-month-old FCG mouse hippocampus, revealed genomic context-specific regulatory effects of genotypic and gonadal sex on X- and autosome-encoded gene expression and DNA modification patterns. X-chromosomal epigenomic patterns, classically associated with X-inactivation, were established almost entirely by genotypic sex, independent of gonadal sex. Differences in X-chromosome methylation were primarily localized to gene regulatory regions including promoters, CpG islands, CTCF binding sites, and active/poised chromatin, with an inverse relationship between methylation and gene expression. Autosomal gene expression demonstrated regulation by both genotypic and gonadal sex, particularly in immune processes. These data demonstrate an important regulatory role of sex chromosomes, independent of gonadal sex, on sex-biased hippocampal transcriptomic and epigenomic profiles. Future studies will need to further interrogate specific CNS cell types, identify the mechanisms by which sex chromosomes regulate autosomes, and differentiate organizational from activational hormonal effects. |
| CTCF |
| Url: https://pubmed.ncbi.nlm.nih.gov/30377227  Title: CTCF Governs the Identity and Migration of MGE-Derived Cortical Interneurons.  The CCCTC-binding factor (CTCF) is a central regulator of chromatin topology recently linked to neurodevelopmental disorders such as intellectual disability, autism, and schizophrenia. The aim of this study was to identify novel roles of CTCF in the developing mouse brain. We provide evidence that CTCF is required for the expression of the LIM homeodomain factor LHX6 involved in fate determination of cortical interneurons (CINs) that originate in the medial ganglionic eminence (MGE). Conditional Ctcf ablation in the MGE of mice of either sex leads to delayed tangential migration, abnormal distribution of CIN in the neocortex, a marked reduction of CINs expressing parvalbumin and somatostatin (Sst), and an increased number of MGE-derived cells expressing Lhx8 and other markers of basal forebrain projection neurons. Likewise, Ctcf-null MGE cells transplanted into the cortex of wild-type hosts generate fewer Sst-expressing CINs and exhibit lamination defects that are efficiently rescued upon reexpression of LHX6. Collectively, these data indicate that CTCF regulates the dichotomy between Lhx6 and Lhx8 to achieve correct specification and migration of MGE-derived CINs.SIGNIFICANCE STATEMENT This work provides evidence that CCCTC-binding factor (CTCF) controls an early fate decision point in the generation of cortical interneurons mediated at least in part by Lhx6. Importantly, the abnormalities described could reflect early molecular and cellular events that contribute to human neurological disorders previously linked to CTCF, including schizophrenia, autism, and intellectual disability. |
| CTCF |
| Url: https://pubmed.ncbi.nlm.nih.gov/22395465  Title: Epigenetic and genetic variation at the IGF2/H19 imprinting control region on 11p15.5 is associated with cerebellum weight.  IGF2 is a paternally expressed imprinted gene with an important role in development and brain function. Allele-specific expression of IGF2 is regulated by DNA methylation at three differentially methylated regions (DMRs) spanning the IGF2/H19 domain on human 11p15.5. We have comprehensively assessed DNA methylation and genotype across the three DMRs and the H19 promoter using tissue from a unique collection of well-characterized and neuropathologically-dissected post-mortem human cerebellum samples (n = 106) and frontal cortex samples (n = 51). We show that DNA methylation, particularly in the vicinity of a key CTCF-binding site (CTCF3) in the imprinting control region (ICR) upstream of H19, is strongly correlated with cerebellum weight. DNA methylation at CTCF3 uniquely explains ~25% of the variance in cerebellum weight. In addition, we report that genetic variation in this ICR is strongly associated with cerebellum weight in a parental-origin specific manner, with maternally-inherited alleles associated with a 16% increase in cerebellum weight compared with paternally-inherited alleles. Given the link between structural brain abnormalities and neuropsychiatric disease, an understanding of the epigenetic and parent-of-origin specific genetic factors associated with brain morphology provides important clues about the etiology of disorders such as schizophrenia and autism. |
| CTNNB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35935366  Title: The extended clinical and genetic spectrum of CTNNB1-related neurodevelopmental disorder.  Loss-of-function mutations of CTNNB1 have been established as the cause of neurodevelopmental disorder with spastic diplegia and visual defects. Although most patients share key phenotypes such as global developmental delay and intellectual disability, patients with CTNNB1-related neurodevelopmental disorder show a broad spectrum of clinical features. We enrolled 13 Korean patients with CTNNB1-related neurodevelopmental disorder who visited Seoul National University Children's Hospital (5 female and 8 male patients with ages ranging from 4 to 22 years). They were all genetically confirmed as having pathogenic loss-of-function variants in CTNNB1 using trio or singleton whole exome sequencing. Variants called from singleton analyses were confirmed to be de novo through parental Sanger sequencing. We identified 11 de novo truncating variants in CTNNB1 in 13 patients, and two pathogenic variants, c.1867C > T (p.Gln623Ter) and c.1420C > T (p.Arg474Ter), found in two unrelated patients, respectively. Five of them were novel pathogenic variants not listed in the ClinVar database. While all patients showed varying degrees of intellectual disability, impaired motor performance, and ophthalmologic problems, none of them had structural brain abnormalities or seizure. In addition, there were three female patients who showed autistic features, such as hand stereotypy, bruxism, and abnormal breathing. A literature review revealed a female predominance of autistic features in CTNNB1-related neurodevelopmental disorder. This is one of the largest single-center cohorts of CTNNB1-related neurodevelopmental disorder. This study investigated variable clinical features of patients and has expanded the clinical and genetic spectrum of the disease. |
| CTNNB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33371872  Title: Characterization of genome-wide association study data reveals spatiotemporal heterogeneity of mental disorders.  Psychiatric disorders such as schizophrenia (SCZ), bipolar disorder (BIP), major depressive disorder (MDD), attention deficit-hyperactivity disorder (ADHD), and autism spectrum disorder (ASD) are often related to brain development. Both shared and unique biological and neurodevelopmental processes have been reported to be involved in these disorders. In this work, we developed an integrative analysis framework to seek for the sensitive spatiotemporal point during brain development underlying each disorder. Specifically, we first identified spatiotemporal gene co-expression modules for four brain regions three developmental stages (prenatal, birth to 11 years old, and older than 13 years), totaling 12 spatiotemporal sites. By integrating GWAS summary statistics and the spatiotemporal co-expression modules, we characterized the risk genes and their co-expression partners for five disorders. We found that SCZ and BIP, ASD and ADHD tend to cluster with each other and keep a distance from other psychiatric disorders. At the gene level, we identified several genes that were shared among the most significant modules, such as CTNNB1 and LNX1, and a hub gene, ATF2, in multiple modules. Moreover, we pinpointed two spatiotemporal points in the prenatal stage with active expression activities and highlighted one postnatal point for BIP. Further functional analysis of the disorder-related module highlighted the apoptotic signaling pathway for ASD and the immune-related and cell-cell adhesion function for SCZ, respectively. Our study demonstrated the dynamic changes of disorder-related genes at the network level, shedding light on the spatiotemporal regulation during brain development. |
| CTNNB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31441226  Title: Genetic Suppression of mTOR Rescues Synaptic and Social Behavioral Abnormalities in a Mouse Model of Pten Haploinsufficiency.  Heterozygous mutations in PTEN, which encodes a negative regulator of the mTOR and β-catenin signaling pathways, cause macrocephaly/autism syndrome. However, the neurobiological substrates of the core symptoms of this syndrome are poorly understood. Here, we investigate the relationship between cerebral cortical overgrowth and social behavior deficits in conditional Pten heterozygous female mice (Pten cHet) using Emx1-Cre, which is expressed in cortical pyramidal neurons and a subset of glia. We found that conditional heterozygous mutation of Ctnnb1 (encoding β-catenin) suppresses Pten cHet cortical overgrowth, but not social behavioral deficits, whereas conditional heterozygous mutation of Mtor suppresses social behavioral deficits, but not cortical overgrowth. Neuronal activity in response to social cues and excitatory synapse markers are elevated in the medial prefrontal cortex (mPFC) of Pten cHet mice, and heterozygous mutation in Mtor, but not Ctnnb1, rescues these phenotypes. These findings indicate that macroscale cerebral cortical overgrowth and social behavioral phenotypes caused by Pten haploinsufficiency can be dissociated based on responsiveness to genetic suppression of Ctnnb1 or Mtor. Furthermore, neuronal connectivity appears to be one potential substrate for mTOR-mediated suppression of social behavioral deficits in Pten haploinsufficient mice. Autism Res 2019, 12: 1463-1471. © 2019 International Society for Autism Research, Wiley Periodicals, Inc. LAY SUMMARY: A subgroup of individuals with autism display overgrowth of the head and the brain during development. Using a mouse model of an autism risk gene, Pten, that displays both brain overgrowth and social behavioral deficits, we show here that that these two symptoms can be dissociated. Reversal of social behavioral deficits in this model is associated with rescue of abnormal synaptic markers and neuronal activity. |
| CTNNB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26180201  Title: Pten Mutations Alter Brain Growth Trajectory and Allocation of Cell Types through Elevated β-Catenin Signaling.  Abnormal patterns of head and brain growth are a replicated finding in a subset of individuals with autism spectrum disorder (ASD). It is not known whether risk factors associated with ASD and abnormal brain growth (both overgrowth and undergrowth) converge on common biological pathways and cellular mechanisms in the developing brain. Heterozygous mutations in PTEN (PTEN(+/-)), which encodes a negative regulator of the PI3K-Akt-mTOR pathway, are a risk factor for ASD and macrocephaly. Here we use the developing cerebral cortex of Pten(+/-) mice to investigate the trajectory of brain overgrowth and underlying cellular mechanisms. We find that overgrowth is detectable from birth to adulthood, is driven by hyperplasia, and coincides with excess neurons at birth and excess glia in adulthood. β-Catenin signaling is elevated in the developing Pten(+/-) cortex, and a heterozygous mutation in Ctnnb1 (encoding β-catenin), itself a candidate gene for ASD and microcephaly, can suppress Pten(+/-) cortical overgrowth. Thus, a balance of Pten and β-catenin signaling regulates normal brain growth trajectory by controlling cell number, and imbalance in this relationship can result in abnormal brain growth. We report that Pten haploinsufficiency leads to a dynamic trajectory of brain overgrowth during development and altered scaling of neuronal and glial cell populations. β-catenin signaling is elevated in the developing cerebral cortex of Pten haploinsufficient mice, and a heterozygous mutation in β-catenin, itself a candidate gene for ASD and microcephaly, suppresses Pten(+/-) cortical overgrowth. This leads to the new insight that Pten and β-catenin signaling act in a common pathway to regulate normal brain growth trajectory by controlling cell number, and disruption of this pathway can result in abnormal brain growth. |
| CTNNB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24656144  Title: Prostaglandin E2 alters Wnt-dependent migration and proliferation in neuroectodermal stem cells: implications for autism spectrum disorders.  Prostaglandin E2 (PGE2) is a natural lipid-derived molecule that is involved in important physiological functions. Abnormal PGE2 signalling has been associated with pathologies of the nervous system. Previous studies provide evidence for the interaction of PGE2 and canonical Wnt signalling pathways in non-neuronal cells. Since the Wnt pathway is crucial in the development and organization of the brain, the main goal of this study is to determine whether collaboration between these pathways exists in neuronal cell types. We report that PGE2 interacts with canonical Wnt signalling through PKA and PI-3K in neuroectodermal (NE-4C) stem cells. We used time-lapse microscopy to determine that PGE2 increases the final distance from origin, path length travelled, and the average speed of migration in Wnt-activated cells. Furthermore, PGE2 alters distinct cellular phenotypes that are characteristic of Wnt-induced NE-4C cells, which corresponds to the modified splitting behaviour of the cells. We also found that in Wnt-induced cells the level of β-catenin protein was increased and the expression levels of Wnt-target genes (Ctnnb1, Ptgs2, Ccnd1, Mmp9) was significantly upregulated in response to PGE2 treatment. This confirms that PGE2 activated the canonical Wnt signalling pathway. Furthermore, the upregulated genes have been previously associated with ASD. Our findings show, for the first time, evidence for cross-talk between PGE2 and Wnt signalling in neuronal cells, where PKA and PI-3K might act as mediators between the two pathways. Given the importance of PGE2 and Wnt signalling in prenatal development of the nervous system, our study provides insight into how interaction between these two pathways may influence neurodevelopment. |
| CUL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35296809  Title: Targeting histone demethylase LSD1 for treatment of deficits in autism mouse models.  Large-scale genetic studies have revealed that the most prominent genes disrupted in autism are chromatin regulators mediating histone methylation/demethylation, suggesting the central role of epigenetic dysfunction in this disorder. Here, we show that histone lysine 4 dimethylation (H3K4me2), a histone mark linked to gene activation, is significantly decreased in the prefrontal cortex (PFC) of autistic human patients and mutant mice with the deficiency of top-ranking autism risk factor Shank3 or Cul3. A brief treatment of the autism models with highly potent and selective inhibitors of the H3K4me2 demethylase LSD1 (KDM1A) leads to the robust rescue of core symptoms of autism, including social deficits and repetitive behaviors. Concomitantly, LSD1 inhibition restores NMDA receptor function in PFC and AMPA receptor-mediated currents in striatum of Shank3-deficient mice. Genome-wide RNAseq and ChIPseq reveal that treatment of Shank3-deficient mice with the LSD1 inhibitor restores the expression and H3K4me2 occupancy of downregulated genes enriched in synaptic signaling and developmental processes. The immediate early gene tightly linked to neuronal plasticity, Egr1, is on the top list of rescued genes. The diminished transcription of Egr1 is recapitulated in PFC of autistic human patients. Overexpression of Egr1 in PFC of Shank3-deficient mice ameliorates social preference deficits. These results have for the first time revealed an important role of H3K4me2 abnormality in ASD pathophysiology, and the therapeutic potential of targeting H3K4me2 demethylase LSD1 or the downstream molecule Egr1 for ASD. |
| CUL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34031387  Title: Cul3 regulates cytoskeleton protein homeostasis and cell migration during a critical window of brain development.  De novo loss of function mutations in the ubiquitin ligase-encoding gene Cullin3 (CUL3) lead to autism spectrum disorder (ASD). In mouse, constitutive Cul3 haploinsufficiency leads to motor coordination deficits as well as ASD-relevant social and cognitive impairments. However, induction of Cul3 haploinsufficiency later in life does not lead to ASD-relevant behaviors, pointing to an important role of Cul3 during a critical developmental window. Here we show that Cul3 is essential to regulate neuronal migration and, therefore, constitutive Cul3 heterozygous mutant mice display cortical lamination abnormalities. At the molecular level, we found that Cul3 controls neuronal migration by tightly regulating the amount of Plastin3 (Pls3), a previously unrecognized player of neural migration. Furthermore, we found that Pls3 cell-autonomously regulates cell migration by regulating actin cytoskeleton organization, and its levels are inversely proportional to neural migration speed. Finally, we provide evidence that cellular phenotypes associated with autism-linked gene haploinsufficiency can be rescued by transcriptional activation of the intact allele in vitro, offering a proof of concept for a potential therapeutic approach for ASDs. |
| CUL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33727673  Title: Autism-linked Cullin3 germline haploinsufficiency impacts cytoskeletal dynamics and cortical neurogenesis through RhoA signaling.  E3-ubiquitin ligase Cullin3 (Cul3) is a high confidence risk gene for autism spectrum disorder (ASD) and developmental delay (DD). To investigate how Cul3 mutations impact brain development, we generated a haploinsufficient Cul3 mouse model using CRISPR/Cas9 genome engineering. Cul3 mutant mice exhibited social and cognitive deficits and hyperactive behavior. Brain MRI found decreased volume of cortical regions and changes in many other brain regions of Cul3 mutant mice starting from early postnatal development. Spatiotemporal transcriptomic and proteomic profiling of embryonic, early postnatal and adult brain implicated neurogenesis and cytoskeletal defects as key drivers of Cul3 functional impact. Specifically, dendritic growth, filamentous actin puncta, and spontaneous network activity were reduced in Cul3 mutant mice. Inhibition of small GTPase RhoA, a molecular substrate of Cul3 ligase, rescued dendrite length and network activity phenotypes. Our study identified defects in neuronal cytoskeleton and Rho signaling as the primary targets of Cul3 mutation during brain development. |
| CUL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32842956  Title: Genetic control of temperament traits across species: association of autism spectrum disorder risk genes with cattle temperament.  Temperament traits are of high importance across species. In humans, temperament or personality traits correlate with psychological traits and psychiatric disorders. In cattle, they impact animal welfare, product quality and human safety, and are therefore of direct commercial importance. We hypothesized that genetic factors that contribute to variation in temperament among individuals within a species will be shared between humans and cattle. Using imputed whole-genome sequence data from 9223 beef cattle from three cohorts, a series of genome-wide association studies was undertaken on cattle flight time, a temperament phenotype measured as the time taken for an animal to cover a short-fixed distance after release from an enclosure. We also investigated the association of cattle temperament with polymorphisms in bovine orthologs of risk genes for neuroticism, schizophrenia, autism spectrum disorders (ASD), and developmental delay disorders in humans. Variants with the strongest associations were located in the bovine orthologous region that is involved in several behavioural and cognitive disorders in humans. These variants were also partially validated in independent cattle cohorts. Genes in these regions (BARHL2, NDN, SNRPN, MAGEL2, ABCA12, KIFAP3, TOPAZ1, FZD3, UBE3A, and GABRA5) were enriched for the GO term neuron migration and were differentially expressed in brain and pituitary tissues in humans. Moreover, variants within 100 kb of ASD susceptibility genes were associated with cattle temperament and explained 6.5% of the total additive genetic variance in the largest cattle cohort. The ASD genes with the most significant associations were GABRB3 and CUL3. Using the same 100 kb window, a weak association was found with polymorphisms in schizophrenia risk genes and no association with polymorphisms in neuroticism and developmental delay disorders risk genes. Our analysis showed that genes identified in a meta-analysis of cattle temperament contribute to neuron development functions and are differentially expressed in human brain tissues. Furthermore, some ASD susceptibility genes are associated with cattle temperament. These findings provide evidence that genetic control of temperament might be shared between humans and cattle and highlight the potential for future analyses to leverage results between species. |
| CUL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31780330  Title: CUL3 Deficiency Causes Social Deficits and Anxiety-like Behaviors by Impairing Excitation-Inhibition Balance through the Promotion of Cap-Dependent Translation.  Autism spectrum disorders (ASD) are a group of neurodevelopmental disorders with symptoms including social deficits, anxiety, and communication difficulties. However, ASD pathogenic mechanisms are poorly understood. Mutations of CUL3, which encodes Cullin 3 (CUL3), a component of an E3 ligase complex, are thought of as risk factors for ASD and schizophrenia (SCZ). CUL3 is abundant in the brain, yet little is known of its function. Here, we show that CUL3 is critical for neurodevelopment. CUL3-deficient mice exhibited social deficits and anxiety-like behaviors with enhanced glutamatergic transmission and neuronal excitability. Proteomic analysis revealed eIF4G1, a protein for Cap-dependent translation, as a potential target of CUL3. ASD-associated cellular and behavioral deficits could be rescued by pharmacological inhibition of the eIF4G1 function and chemogenetic inhibition of neuronal activity. Thus, CUL3 is critical to neural development, neurotransmission, and excitation-inhibition (E-I) balance. Our study provides novel insight into the pathophysiological mechanisms of ASD and SCZ. |
| CUL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29088697  Title: Kctd13 deletion reduces synaptic transmission via increased RhoA.  Copy-number variants of chromosome 16 region 16p11.2 are linked to neuropsychiatric disorders and are among the most prevalent in autism spectrum disorders. Of many 16p11.2 genes, Kctd13 has been implicated as a major driver of neurodevelopmental phenotypes. The function of KCTD13 in the mammalian brain, however, remains unknown. Here we delete the Kctd13 gene in mice and demonstrate reduced synaptic transmission. Reduced synaptic transmission correlates with increased levels of Ras homolog gene family, member A (RhoA), a KCTD13/CUL3 ubiquitin ligase substrate, and is reversed by RhoA inhibition, suggesting increased RhoA as an important mechanism. In contrast to a previous knockdown study, deletion of Kctd13 or kctd13 does not increase brain size or neurogenesis in mice or zebrafish, respectively. These findings implicate Kctd13 in the regulation of neuronal function relevant to neuropsychiatric disorders and clarify the role of Kctd13 in neurogenesis and brain size. Our data also reveal a potential role for RhoA as a therapeutic target in disorders associated with KCTD13 deletion. |
| CUL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25695269  Title: Spatiotemporal 16p11.2 protein network implicates cortical late mid-fetal brain development and KCTD13-Cul3-RhoA pathway in psychiatric diseases.  The psychiatric disorders autism and schizophrenia have a strong genetic component, and copy number variants (CNVs) are firmly implicated. Recurrent deletions and duplications of chromosome 16p11.2 confer a high risk for both diseases, but the pathways disrupted by this CNV are poorly defined. Here we investigate the dynamics of the 16p11.2 network by integrating physical interactions of 16p11.2 proteins with spatiotemporal gene expression from the developing human brain. We observe profound changes in protein interaction networks throughout different stages of brain development and/or in different brain regions. We identify the late mid-fetal period of cortical development as most critical for establishing the connectivity of 16p11.2 proteins with their co-expressed partners. Furthermore, our results suggest that the regulation of the KCTD13-Cul3-RhoA pathway in layer 4 of the inner cortical plate is crucial for controlling brain size and connectivity and that its dysregulation by de novo mutations may be a potential determinant of 16p11.2 CNV deletion and duplication phenotypes. |
| CUX1, TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35944998  Title: Shared and Distinct Functional Effects of Patient-Specific Tbr1 Mutations on Cortical Development.  T-Box Brain Transcription Factor 1 (TBR1) plays essential roles in brain development, mediating neuronal migration, fate specification, and axon tract formation. While heterozygous loss-of-function and missense TBR1 mutations are associated with neurodevelopmental conditions, the effects of these heterogeneous mutations on brain development have yet to be fully explored. We characterized multiple mouse lines carrying Tbr1 mutations differing by type and exonic location, including the previously generated Tbr1 exon 2-3 knock-out (KO) line, and we analyzed male and female mice at neonatal and adult stages. The frameshift patient mutation A136PfsX80 (A136fs) caused reduced TBR1 protein in cortex similar to Tbr1 KO, while the missense patient mutation K228E caused significant TBR1 upregulation. Analysis of cortical layer formation found similar defects between KO and A136fs homozygotes in their CUX1+ and CTIP2+ layer positions, while K228E homozygosity produced layering defects distinct from these mutants. Meanwhile, the examination of cortical apoptosis found extensive cell death in KO homozygotes but limited cell death in A136fs or K228E homozygotes. Despite their discordant cortical phenotypes, these Tbr1 mutations produced several congruent phenotypes, including anterior commissure reduction in heterozygotes, which was previously observed in humans with TBR1 mutations. These results indicate that patient-specific Tbr1 mutant mice will be valuable translational models for pinpointing shared and distinct etiologies among patients with TBR1-related developmental conditions.SIGNIFICANCE STATEMENT Mutations of the TBR1 gene increase the likelihood of neurodevelopmental conditions such as intellectual disability and autism. Therefore, the study of TBR1 can offer insights into the biological mechanisms underlying these conditions, which affect millions worldwide. To improve the modeling of TBR1-related conditions over current Tbr1 knock-out mice, we created mouse lines carrying Tbr1 mutations identical to those found in human patients. Mice with one mutant Tbr1 copy show reduced amygdalar connections regardless of mutation type, suggesting a core biomarker for TBR1-related disorders. In mice with two mutant Tbr1 copies, brain phenotypes diverge by mutation type, suggesting differences in Tbr1 gene functionality in different patients. These mouse models will serve as valuable tools for understanding genotype-phenotype relationships among patients with neurodevelopmental conditions. |
| CUX1, SOX5, TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35781633  Title: Tbr1 Misexpression Alters Neuronal Development in the Cerebral Cortex.  Changes in the transcription factor (TF) expression are critical for brain development, and they may also underlie neurodevelopmental disorders. Indeed, T-box brain1 (Tbr1) is a TF crucial for the formation of neocortical layer VI, and mutations and microdeletions in that gene are associated with malformations in the human cerebral cortex, alterations that accompany autism spectrum disorder (ASD). Interestingly, Tbr1 upregulation has also been related to the occurrence of ASD-like symptoms, although limited studies have addressed the effect of increased Tbr1 levels during neocortical development. Here, we analysed the impact of Tbr1 misexpression in mouse neural progenitor cells (NPCs) at embryonic day 14.5 (E14.5), when they mainly generate neuronal layers II-IV. By E18.5, cells accumulated in the intermediate zone and in the deep cortical layers, whereas they became less abundant in the upper cortical layers. In accordance with this, the proportion of Sox5+ cells in layers V-VI increased, while that of Cux1+ cells in layers II-IV decreased. On postnatal day 7, fewer defects in migration were evident, although a higher proportion of Sox5+ cells were seen in the upper and deep layers. The abnormal neuronal migration could be partially due to the altered multipolar-bipolar neuron morphologies induced by Tbr1 misexpression, which also reduced dendrite growth and branching, and disrupted the corpus callosum. Our results indicate that Tbr1 misexpression in cortical NPCs delays or disrupts neuronal migration, neuronal specification, dendrite development and the formation of the callosal tract. Hence, genetic changes that provoke ectopic Tbr1 upregulation during development could provoke cortical brain malformations. |
| CUX1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27667684  Title: Mutations in Human Accelerated Regions Disrupt Cognition and Social Behavior.  Comparative analyses have identified genomic regions potentially involved in human evolution but do not directly assess function. Human accelerated regions (HARs) represent conserved genomic loci with elevated divergence in humans. If some HARs regulate human-specific social and behavioral traits, then mutations would likely impact cognitive and social disorders. Strikingly, rare biallelic point mutations-identified by whole-genome and targeted "HAR-ome" sequencing-showed a significant excess in individuals with ASD whose parents share common ancestry compared to familial controls, suggesting a contribution in 5% of consanguineous ASD cases. Using chromatin interaction sequencing, massively parallel reporter assays (MPRA), and transgenic mice, we identified disease-linked, biallelic HAR mutations in active enhancers for CUX1, PTBP2, GPC4, CDKL5, and other genes implicated in neural function, ASD, or both. Our data provide genetic evidence that specific HARs are essential for normal development, consistent with suggestions that their evolutionary changes may have altered social and/or cognitive behavior. PAPERCLIP. |
| CUX1, FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26939565  Title: Developmental role of the cell adhesion molecule Contactin-6 in the cerebral cortex and hippocampus.  The gene encoding the neural cell adhesion molecule Contactin-6 (Cntn6 a.k.a. NB-3) has been implicated as an autism risk gene, suggesting that its mutation is deleterious to brain development. Due to its GPI-anchor at Cntn6 may exert cell adhesion/receptor functions in complex with other membrane proteins, or serve as a ligand. We aimed to uncover novel phenotypes related to Cntn6 functions during development in the cerebral cortex of adult Cntn6(-/-) mice. We first determined Cntn6 protein and mRNA expression in the cortex, thalamic nuclei and the hippocampus at P14, which decreased specifically in the cortex at adult stages. Neuroanatomical analysis demonstrated a significant decrease of Cux1+ projection neurons in layers II-IV and an increase of FoxP2+ projection neurons in layer VI in the visual cortex of adult Cntn6(-/-) mice compared to wild-type controls. Furthermore, the number of parvalbumin+ (PV) interneurons was decreased in Cntn6(-/-) mice, while the amount of NPY+ interneurons remained unchanged. In the hippocampus the delineation and outgrowth of mossy fibers remained largely unchanged, except for the observation of a larger suprapyramidal bundle. The observed abnormalities in the cerebral cortex and hippocampus of Cntn6(-/-) mice suggests that Cntn6 serves developmental functions involving cell survival, migration and fasciculation. Furthermore, these data suggest that Cntn6 engages in both trans- and cis-interactions and may be involved in larger protein interaction networks. |
| CUX1, CUX2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25059644  Title: Cux1 and Cux2 selectively target basal and apical dendritic compartments of layer II-III cortical neurons.  A number of recent reports implicate the differential regulation of apical and basal dendrites in autism disorders and in the higher functions of the human brain. They show that apical and basal dendrites are functionally specialized and that mechanisms regulating their development have important consequences for neuron function. The molecular identity of layer II-III neurons of the cerebral cortex is determined by the overlapping expression of Cux1 and Cux2. We previously showed that both Cux1 and Cux2 are necessary and nonredundant for normal dendrite development of layer II-III neurons. Loss of function of either gene reduced dendrite arbors, while overexpression increased dendritic complexity and suggested additive functions. We herein characterize the function of Cux1 and Cux2 in the development of apical and basal dendrites. By in vivo loss and gain of function analysis, we show that while the expression level of either Cux1 or Cux2 influences both apical and basal dendrites, they have distinct effects. Changes in Cux1 result in a marked effect on the development of the basal compartment whereas modulation of Cux2 has a stronger influence on the apical compartment. These distinct effects of Cux genes might account for the functional diversification of layer II-III neurons into different subpopulations, possibly with distinct connectivity patterns and modes of neuron response. Our data suggest that by their differential effects on basal and apical dendrites, Cux1 and Cux2 can promote the integration of layer II-III neurons in the intracortical networks in highly specific ways. |
| CUX1, EN2, NFIB |
| Url: https://pubmed.ncbi.nlm.nih.gov/22180456  Title: Cut-like homeobox 1 and nuclear factor I/B mediate ENGRAILED2 autism spectrum disorder-associated haplotype function.  Both common and rare variants contribute to autism spectrum disorder (ASD) risk, but few variants have been established as functional. Previously we demonstrated that an intronic haplotype (rs1861972-rs1861973 A-C) in the homeobox transcription factor ENGRAILED2 (EN2) is significantly associated with ASD. Positive association has also been reported in six additional data sets, suggesting EN2 is an ASD susceptibility gene. Additional support for this possibility requires identification of functional variants that affect EN2 regulation or activity. In this study, we demonstrate that the A-C haplotype is a transcriptional activator. Luciferase (luc) assays in mouse neuronal cultures determined that the A-C haplotype increases expression levels (50%, P < 0.01, 24 h; 250%, P < 0.0001, 72 h). Mutational analysis indicates that the A-C haplotype activator function requires both associated A and C alleles. A minimal 202-bp element is sufficient for function and also specifically binds a protein complex. Mass spectrometry identified these proteins as the transcription factors, Cut-like homeobox 1 (Cux1) and nuclear factor I/B (Nfib). Subsequent antibody supershifts and chromatin immunoprecipitations demonstrated that human CUX1 and NFIB bind the A-C haplotype. Co-transfection and knock-down experiments determined that both CUX1 and NFIB are required for the A-C haplotype activator function. These data demonstrate that the ASD-associated A-C haplotype is a transcriptional activator, and both CUX1 and NFIB mediate this activity. These results provide biochemical evidence that the ASD-associated A-C haplotype is functional, further supporting EN2 as an ASD susceptibility gene. |
| CUX2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29795476  Title: A recurrent de novo CUX2 missense variant associated with intellectual disability, seizures, and autism spectrum disorder.  In most patients with intellectual disability (ID), the etiology is unknown, but lately several de novo variants have been associated with ID. One of the involved genes, CUX2, has twice been reported to be affected by a de novo variant c.1768G>A; p.(Glu590Lys) in patients with ID or epileptic encephalopathy. CUX2 is expressed primarily in nervous tissues where it may act as a transcription factor involved in neural specification. Here we describe a third case who was diagnosed with epilepsy including general and myoclonic seizures, moderate to severe cognitive disability, and infantile autism. The patient was heterozygous for the c.1768G>A; p.(Glu590Lys) variant in CUX2 identified by whole exome sequencing. These findings strongly suggest a causal impact of this variant and add to our understanding of a subset of patients with ID, seizures, and autism spectrum disorder as well as suggest an important role for the CUX2 gene in human brain function. |
| DDX3X, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36350923  Title: Integrated gene analyses of de novo variants from 46,612 trios with autism and developmental disorders.  Most genetic studies consider autism spectrum disorder (ASD) and developmental disorder (DD) separately despite overwhelming comorbidity and shared genetic etiology. Here, we analyzed de novo variants (DNVs) from 15,560 ASD (6,557 from SPARK) and 31,052 DD trios independently and also combined as broader neurodevelopmental disorders (NDDs) using three models. We identify 615 NDD candidate genes (false discovery rate [FDR] P n = 41) or truncating (n = 12) DNVs. We also find 10 genes with evidence of male- or female-bias enrichment, including 4 X chromosome genes with significant female burden (DDX3X, MECP2, WDR45, and HDAC8). This large-scale integrative analysis identifies candidates and functional subsets of NDD genes. |
| DDX3X |
| Url: https://pubmed.ncbi.nlm.nih.gov/35762573  Title: Aberrant cortical development is driven by impaired cell cycle and translational control in a DDX3X syndrome model.  Mutations in the RNA helicase, DDX3X, are a leading cause of Intellectual Disability and present as DDX3X syndrome, a neurodevelopmental disorder associated with cortical malformations and autism. Yet, the cellular and molecular mechanisms by which DDX3X controls cortical development are largely unknown. Here, using a mouse model of Ddx3x loss-of-function we demonstrate that DDX3X directs translational and cell cycle control of neural progenitors, which underlies precise corticogenesis. First, we show brain development is sensitive to Ddx3x dosage; complete Ddx3x loss from neural progenitors causes microcephaly in females, whereas hemizygous males and heterozygous females show reduced neurogenesis without marked microcephaly. In addition, Ddx3x loss is sexually dimorphic, as its paralog, Ddx3y, compensates for Ddx3x in the developing male neocortex. Using live imaging of progenitors, we show that DDX3X promotes neuronal generation by regulating both cell cycle duration and neurogenic divisions. Finally, we use ribosome profiling in vivo to discover the repertoire of translated transcripts in neural progenitors, including those which are DDX3X-dependent and essential for neurogenesis. Our study reveals invaluable new insights into the etiology of DDX3X syndrome, implicating dysregulated progenitor cell cycle dynamics and translation as pathogenic mechanisms. |
| DDX3X |
| Url: https://pubmed.ncbi.nlm.nih.gov/35392274  Title: Expansion of Clinical and Genetic Spectrum of DDX3X Neurodevelopmental Disorder in 23 Chinese Patients.  De novo DDX3X variants account for 1-3% of unexplained intellectual disability cases in females and very rarely in males. Yet, the clinical and genetic features of DDX3X neurodevelopmental disorder in the Chinese cohort have not been characterized. A total of 23 Chinese patients (i.e., 22 female and 1 male) with 22 de novo DDX3X deleterious variants were detected among 2,317 probands with unexplained intellectual disability (ID) undertaking whole exome sequencing (WES). The age, sex, genetic data, feeding situation, growth, developmental conditions, and auxiliary examinations of the cohort were collected. The Chinese version of the Gesell Development Diagnosis Scale (GDDS-C) was used to evaluate neurodevelopment of DDX3X patients. The Social Communication Questionnaire (SCQ)-Lifetime version was applied as a primary screener to assess risk for autism spectrum disorder (ASD). A total of 17 DDX3X variants were novel and 22 were de novo. Missense variants overall were only slightly more common than loss-of-function variants and were mainly located in two functional subdomains. The average age of this cohort was 2.67 (±1.42) years old. The overlapping phenotypic spectrum between this cohort and previously described reports includes intellectual disability (23/23, 100%) with varying degrees of severity, muscle tone abnormalities (17/23, 73.9%), feeding difficulties (13/23, 56.5%), ophthalmologic problems (11/23, 47.8%), and seizures (6/23, 26.1%). A total of 15 individuals had notable brain anatomical disruption (15/23, 65.2%), including lateral ventricle enlargement, corpus callosum abnormalities, and delayed myelination. Furthermore, 9 patients showed abnormal electroencephalogram results (9/23, 39.1%). Hypothyroidism was first noted as a novel clinical feature (6/23, 26.1%). The five primary neurodevelopmental domains of GDDS-C in 21 patients were impaired severely, and 13 individuals were above the "at-risk" threshold for ASD. Although a certain degree of phenotypic overlap with previously reported cohorts, our study described the phenotypic and variation spectrum of 23 additional individuals carrying DDX3X variants in the Chinese population, adding hypothyroidism as a novel finding. We confirmed the importance of DDX3X as a pathogenic gene in unexplained intellectual disability, supporting the necessity of the application of WES in patients with unexplained intellectual disability. |
| DDX3X |
| Url: https://pubmed.ncbi.nlm.nih.gov/35326346  Title: Autistic-like Behaviors Associated with a Novel Non-Canonical Splice-Site DDX3X Variant: A Case Report of a Rare Clinical Syndrome.  Background. Heterozygous pathogenic variants in the DDX3X gene account for 1−3% of females with intellectual and developmental disabilities (IDD). The clinical presentation is variable, including a wide range of neurological and behavioral deficits and structural defects of the brain. Approximately 52% of affected females remain nonverbal after five years of age. Case presentation: We report a 7 year old nonverbal female with a likely novel de novo pathogenic heterozygous variant in the DDX3X gene affecting the non-canonical splice-site in the intron 1 (NM\_001356:c.45+12G>A). The patient presents with features typical for the DDX3X phenotype, such as: movement disorders, behavioral problems, a diagnosis of autism spectrum disorder (ASD), and some other features uncommon for DDX3X such as: muscle hypertonia and spinal asymmetry evaluated through the scoliometer. Conclusions. Due to its rare occurrence, the clinical picture of DDX3X syndrome is yet to be fully determined. So far, behavioral disorders, including those from ASD, and neurological abnormalities seem to be the dominant features of this disorder. |
| DDX3X |
| Url: https://pubmed.ncbi.nlm.nih.gov/32135084  Title: Pathogenic DDX3X Mutations Impair RNA Metabolism and Neurogenesis during Fetal Cortical Development.  De novo germline mutations in the RNA helicase DDX3X account for 1%-3% of unexplained intellectual disability (ID) cases in females and are associated with autism, brain malformations, and epilepsy. Yet, the developmental and molecular mechanisms by which DDX3X mutations impair brain function are unknown. Here, we use human and mouse genetics and cell biological and biochemical approaches to elucidate mechanisms by which pathogenic DDX3X variants disrupt brain development. We report the largest clinical cohort to date with DDX3X mutations (n = 107), demonstrating a striking correlation between recurrent dominant missense mutations, polymicrogyria, and the most severe clinical outcomes. We show that Ddx3x controls cortical development by regulating neuron generation. Severe DDX3X missense mutations profoundly disrupt RNA helicase activity, induce ectopic RNA-protein granules in neural progenitors and neurons, and impair translation. Together, these results uncover key mechanisms underlying DDX3X syndrome and highlight aberrant RNA metabolism in the pathogenesis of neurodevelopmental disease. |
| DDX3X, KMT2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/32012899  Title: Proteomic Analysis of Brain Region and Sex-Specific Synaptic Protein Expression in the Adult Mouse Brain.  Genetic disruption of synaptic proteins results in a whole variety of human neuropsychiatric disorders including intellectual disability, schizophrenia or autism spectrum disorder (ASD). In a wide range of these so-called synaptopathies a sex bias in prevalence and clinical course has been reported. Using an unbiased proteomic approach, we analyzed the proteome at the interaction site of the pre- and postsynaptic compartment, in the prefrontal cortex, hippocampus, striatum and cerebellum of male and female adult C57BL/6J mice. We were able to reveal a specific repertoire of synaptic proteins in different brain areas as it has been implied before. Additionally, we found a region-specific set of novel synaptic proteins differentially expressed between male and female individuals including the strong ASD candidates DDX3X, KMT2C, MYH10 and SET. Being the first comprehensive analysis of brain region-specific synaptic proteomes from male and female mice, our study provides crucial information on sex-specific differences in the molecular anatomy of the synapse. Our efforts should serve as a neurobiological framework to better understand the influence of sex on synapse biology in both health and disease. |
| DDX3X |
| Url: https://pubmed.ncbi.nlm.nih.gov/31785789  Title: Sex-Based Analysis of De Novo Variants in Neurodevelopmental Disorders.  While genes with an excess of de novo mutations (DNMs) have been identified in children with neurodevelopmental disorders (NDDs), few studies focus on DNM patterns where the sex of affected children is examined separately. We considered ∼8,825 sequenced parent-child trios (n ∼26,475 individuals) and identify 54 genes with a DNM enrichment in males (n = 18), females (n = 17), or overlapping in both the male and female subsets (n = 19). A replication cohort of 18,778 sequenced parent-child trios (n = 56,334 individuals) confirms 25 genes (n = 3 in males, n = 7 in females, n = 15 in both male and female subsets). As expected, we observe significant enrichment on the X chromosome for females but also find autosomal genes with potential sex bias (females, CDK13, ITPR1; males, CHD8, MBD5, SYNGAP1); 6.5% of females harbor a DNM in a female-enriched gene, whereas 2.7% of males have a DNM in a male-enriched gene. Sex-biased genes are enriched in transcriptional processes and chromatin binding, primarily reside in the nucleus of cells, and have brain expression. By downsampling, we find that DNM gene discovery is greatest when studying affected females. Finally, directly comparing de novo allele counts in NDD-affected males and females identifies one replicated genome-wide significant gene (DDX3X) with locus-specific enrichment in females. Our sex-based DNM enrichment analysis identifies candidate NDD genes differentially affecting males and females and indicates that the study of females with NDDs leads to greater gene discovery consistent with the female-protective effect. |
| DLX2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28821666  Title: GABAergic Interneuron Differentiation in the Basal Forebrain Is Mediated through Direct Regulation of Glutamic Acid Decarboxylase Isoforms by Dlx Homeobox Transcription Factors.  GABA is the key inhibitory neurotransmitter in the cortex but regulation of its synthesis during forebrain development is poorly understood. In the telencephalon, members of the distal-less (Dlx) homeobox gene family are expressed in, and regulate the development of, the basal ganglia primodia from which many GABAergic neurons originate and migrate to other forebrain regions. The Dlx1/Dlx2 double knock-out mice die at birth with abnormal cortical development, including loss of tangential migration of GABAergic inhibitory interneurons to the neocortex (Anderson et al., 1997a). We have discovered that specific promoter regulatory elements of glutamic acid decarboxylase isoforms (Gad1 and Gad2), which regulate GABA synthesis from the excitatory neurotransmitter glutamate, are direct transcriptional targets of both DLX1 and DLX2 homeoproteins in vivo Further gain- and loss-of-function studies in vitro and in vivo demonstrated that both DLX1 and DLX2 are necessary and sufficient for Gad gene expression. DLX1 and/or DLX2 activated the transcription of both Gad genes, and defects in Dlx function disrupted the differentiation of GABAergic interneurons with global reduction in GABA levels in the forebrains of the Dlx1/Dlx2 double knock-out mouse in vivo Identification of Gad genes as direct Dlx transcriptional targets is significant; it extends our understanding of Dlx gene function in the developing forebrain beyond the regulation of tangential interneuron migration to the differentiation of GABAergic interneurons arising from the basal telencephalon, and may help to unravel the pathogenesis of several developmental brain disorders.SIGNIFICANCE STATEMENT GABA is the major inhibitory neurotransmitter in the brain. We show that Dlx1/Dlx2 homeobox genes regulate GABA synthesis during forebrain development through direct activation of glutamic acid decarboxylase enzyme isoforms that convert glutamate to GABA. This discovery helps explain how Dlx mutations result in abnormal forebrain development, due to defective differentiation, in addition to the loss of tangential migration of GABAergic inhibitory interneurons to the neocortex. Reduced numbers or function of cortical GABAergic neurons may lead to hyperactivity states such as seizures (Cobos et al., 2005) or contribute to the pathogenesis of some autism spectrum disorders. GABAergic dysfunction in the basal ganglia could disrupt the learning and development of complex motor and cognitive behaviors (Rubenstein and Merzenich, 2003). |
| DLX2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/15542242  Title: Analysis of the autism chromosome 2 linkage region: GAD1 and other candidate genes.  Autism has a strong and complex genetic component, involving several genes. Genomic screens, including our own, have shown suggestive evidence for linkage over a 20-30 cM region on chromosome 2q31-q33. Two subsequent reports showed that the linkage evidence increased in the subset of families with phrase speech delay (PSD), defined as onset of phrase speech later than 3 years of age. To further investigate the linkage in the presumptive candidate region, microsatellite markers in a 2 cM grid covering the interval from 164 to 203 cM were analyzed in 110 multiplex (2 or more sampled autism patients) families. A maximum heterogeneity LOD (HLOD) score of 1.54 was detected at D2S1776 (173 cM) in the overall dataset (dominant model), increasing to 1.71 in the PSD subset. While not conclusive, these data continue to provide suggestive evidence for linkage, particularly considering replication by multiple independent groups. Positive LOD scores extended over the entire region, continuing to define a broad candidate interval. Association studies were performed on several functional candidates mapping within the region. These included GAD1, encoding GAD67, whose levels are reduced in autopsy brain material from autistic subjects, and STK17B, ABI2, CTLA4, CD28, NEUROD1, PDE1A, HOXD1 and DLX2. We found no evidence for significant allelic association between autism and any of these candidates, suggesting that they do not play a major role in the genetics of autism or that substantial allelic heterogeneity at any one of these loci dilutes potential disease-allele association. |
| DLX3, GTF2I |
| Url: https://pubmed.ncbi.nlm.nih.gov/34671237  Title: Increased Sociability in Mice Lacking Intergenic Dlx Enhancers.  The Dlx homeodomain transcription factors play important roles in the differentiation and migration of GABAergic interneuron precursors. The mouse and human genomes each have six Dlx genes organized into three convergently transcribed bigene clusters (Dlx1/2, Dlx3/4, and Dlx5/6) with cis-regulatory elements (CREs) located in the intergenic region of each cluster. Amongst these, the I56i and I12b enhancers from the Dlx1/2 and Dlx5/6 locus, respectively, are active in the developing forebrain. I56i is also a binding site for GTF2I, a transcription factor whose function is associated with increased sociability and Williams-Beuren syndrome. In determining the regulatory roles of these CREs on forebrain development, we have generated mutant mouse-lines where Dlx forebrain intergenic enhancers have been deleted (I56i(-/-), I12b(-/-)). Loss of Dlx intergenic enhancers impairs expression of Dlx genes as well as some of their downstream targets or associated genes including Gad2 and Evf2. The loss of the I56i enhancer resulted in a transient decrease in GABA+ cells in the developing forebrain. The intergenic enhancer mutants also demonstrate increased sociability and learning deficits in a fear conditioning test. Characterizing mice with mutated Dlx intergenic enhancers will help us to further enhance our understanding of the role of these Dlx genes in forebrain development. |
| DLX3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28533516  Title: ASD restricted and repetitive behaviors associated at 17q21.33: genes prioritized by expression in fetal brains.  Autism spectrum disorder (ASD) is a behaviorally defined condition that manifests in infancy or early childhood as deficits in communication skills and social interactions. Often, restricted and repetitive behaviors (RRBs) accompany this disorder. ASD is polygenic and genetically complex, so we hypothesized that focusing analyses on intermediate core component phenotypes, such as RRBs, can reduce genetic heterogeneity and improve statistical power. Applying this approach, we mined Caucasian genome-wide association studies (GWAS) data from two of the largest ASD family cohorts, the Autism Genetics Resource Exchange and Autism Genome Project (AGP). Of the 12 RRBs measured by the Autism Diagnostic Interview-Revised, seven were found to be significantly familial and substantially variable, and hence, were tested for genome-wide association in 3104 ASD-affected children from 2045 families. Using a stringent significance threshold (P-9), GWAS in the AGP revealed an association between 'the degree of the repetitive use of objects or interest in parts of objects' and rs2898883 (P-9), which resides within the sixth intron of PHB. To identify the candidate target genes of the associated single-nucleotide polymorphisms at that locus, we applied chromosome conformation studies in developing human brains and implicated three additional genes: SLC35B1, CALCOCO2 and DLX3. Gene expression, brain imaging and fetal brain expression quantitative trait locus studies prioritize SLC35B1 and PHB. These analyses indicate that GWAS of single heritable features of genetically complex disorders followed by chromosome conformation studies in relevant tissues can be successful in revealing novel risk genes for single core features of ASD. |
| DLX6 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35681437  Title: Dlx5/6 Expression Levels in Mouse GABAergic Neurons Regulate Adult Parvalbumin Neuronal Density and Anxiety/Compulsive Behaviours.  Neuronal circuits integrating Parvalbumin-positive GABAergic inhibitory interneurons (PV) are essential for normal brain function and are often altered in psychiatric conditions. During development, Dlx5 and Dlx6 (Dlx5/6) genes are involved in the differentiation of PV-interneurons. In the adult, Dlx5/6 continue to be expressed at low levels in most telencephalic GABAergic neurons, but their importance in determining the number and distribution of adult PV-interneurons is unknown. Previously, we have shown that targeted deletion of Dlx5/6 in mouse GABAergic neurons (Dlx5/6VgatCre mice) results in altered behavioural and metabolic profiles. Here we evaluate the consequences of targeted Dlx5/6 gene dosage alterations in adult GABAergic neurons. We compare the effects on normal brain of homozygous and heterozygous (Dlx5/6VgatCre and Dlx5/6VgatCre/+ mice) Dlx5/6 deletions to those of Dlx5 targeted overexpression (GABAergicDlx5/+ mice). We find a linear correlation between Dlx5/6 allelic dosage and the density of PV-positive neurons in the adult prelimbic cortex and in the hippocampus. In parallel, we observe that Dlx5/6 expression levels in GABAergic neurons are also linearly associated with the intensity of anxiety and compulsivity-like behaviours. Our findings reinforce the notion that regulation of Dlx5/6 expression is involved in individual cognitive variability and, possibly, in the genesis of certain neuropsychiatric conditions. |
| DLX6, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17701895  Title: DLX5 and DLX6 expression is biallelic and not modulated by MeCP2 deficiency.  Mutations in MECP2 and Mecp2 (encoding methyl-CpG binding protein 2 [MeCP2]) cause distinct neurological phenotypes in humans and mice, respectively, but the molecular pathology is unclear. Recent literature claimed that the developmental homeobox gene DLX5 is imprinted and that its imprinting status is modulated by MeCP2, leading to biallelic expression in Rett syndrome and twofold overexpression of Dlx5 and Dlx6 in Mecp2-null mice. The conclusion that DLX5 is a direct target of MeCP2 has implications for research on the molecular bases of Rett syndrome, autism, and genomic imprinting. Attempting to replicate the reported data, we evaluated allele-specific expression of DLX5 and DLX6 in mouse x human somatic cell hybrids, lymphoblastoid cell lines, and frontal cortex from controls and individuals with MECP2 mutations. We identified novel single-nucleotide polymorphisms in DLX5 and DLX6, enabling the first imprinting studies of DLX6. We found that DLX5 and DLX6 are biallelically expressed in somatic cell hybrids and in human cell lines and brain, with no differences between affected and control samples. We also determined expression levels of Dlx5 and Dlx6 in forebrain from seven male Mecp2-mutant mice and eight wild-type littermates by real-time quantitative reverse-transcriptase polymerase chain reaction assays. Expression of Dlx5 and Dlx6, as well as of the imprinted gene Peg3, in mouse forebrain was highly variable, with no consistent differences between Mecp2-null mutants and controls. We conclude that DLX5 and DLX6 are not imprinted in humans and are not likely to be direct targets of MeCP2 modulation. In contrast, the imprinting status of PEG3 and PEG10 is maintained in MeCP2-deficient tissues. Our results confirm that MeCP2 plays no role in the maintenance of genomic imprinting and add PEG3 and PEG10 to the list of studied imprinted genes. |
| DNMT3A |
| Url: https://pubmed.ncbi.nlm.nih.gov/36909558  Title: Distinct disease mutations in DNMT3A result in a spectrum of behavioral, epigenetic, and transcriptional deficits.  Phenotypic heterogeneity is a common feature of monogenic neurodevelopmental disorders that can arise from differential severity of missense variants underlying disease, but how distinct alleles impact molecular mechanisms to drive variable disease presentation is not well understood. Here, we investigate missense mutations in the DNA methyltransferase DNMT3A associated with variable overgrowth, intellectual disability, and autism, to uncover molecular correlates of phenotypic heterogeneity in neurodevelopmental disease. We generate a DNMT3A P900L/+ mouse model mimicking a disease mutation with mild-to-moderate severity and compare phenotypic and epigenomic effects with a severe R878H mutation. We show that the P900L mutation leads to disease-relevant overgrowth, obesity, and social deficits shared across DNMT3A disorder models, while the R878H mutation causes more extensive epigenomic disruption leading to differential dysregulation of enhancers elements. We identify distinct gene sets disrupted in each mutant which may contribute to mild or severe disease, and detect shared transcriptomic disruption that likely drives common phenotypes across affected individuals. Finally, we demonstrate that core gene dysregulation detected in DNMT3A mutant mice overlaps effects in other developmental disorder models, highlighting the importance of DNMT3A-deposited methylation in neurodevelopment. Together, these findings define central drivers of DNMT3A disorders and illustrate how variable disruption of transcriptional mechanisms can drive the spectrum of phenotypes in neurodevelopmental disease. |
| DNMT3A, FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36758877  Title: Interaction-integrated linear mixed model reveals 3D-genetic basis underlying Autism.  Genetic interactions play critical roles in genotype-phenotype associations. We developed a novel interaction-integrated linear mixed model (ILMM) that integrates a priori knowledge into linear mixed models. ILMM enables statistical integration of genetic interactions upfront and overcomes the problems of searching for combinations. To demonstrate its utility, with 3D genomic interactions (assessed by Hi-C experiments) as a priori, we applied ILMM to whole-genome sequencing data for Autism Spectrum Disorders (ASD) and brain transcriptome data, revealing the 3D-genetic basis of ASD and 3D-expression quantitative loci (3D-eQTLs) for brain tissues. Notably, we reported a potential mechanism involving distal regulation between FOXP2 and DNMT3A, conferring the risk of ASD. |
| DNMT3A |
| Url: https://pubmed.ncbi.nlm.nih.gov/34879184  Title: Gene expression levels of DNA methyltransferase enzymes in Shank3-deficient mouse model of autism during early development.  Objectives. The balance between DNA methylation and demethylation is crucial for the brain development. Therefore, alterations in the expression of enzymes controlling DNA methylation patterns may contribute to the etiology of neurodevelopmental disorders, including autism. SH3 and multiple ankyrin repeat domains 3 (Shank3)-deficient mice are commonly used as a well-characterized transgenic model to investigate the molecular mechanisms of autistic symptoms. DNA methyltransferases (DNMTs), which modulate several cellular processes in neurodevelopment, are implicated in the pathophysiology of autism. In this study, we aimed to describe the gene expression changes of major Dnmts in the brain of Shank3-deficient mice during early development. Methods and Results. The Dnmts gene expression was analyzed by qPCR in 5-day-old homo-zygous Shank3-deficient mice. We found significantly lower Dnmt1 and Dnmt3b gene expression levels in the frontal cortex. However, no such changes were observed in the hippocampus. However, significant increase was observed in the expression of Dnmt3a and Dnmt3b genes in the hypothalamus of Shank3-deficient mice. Conclusions. The present data indicate that abnormalities in the Shank3 gene are accompanied by an altered expression of DNA methylation enzymes in the early brain development stages, therefore, specific epigenetic control mechanisms in autism-relevant models should be more extensively investigated. |
| DNMT3A, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33826889  Title: MicroRNA-29 is an essential regulator of brain maturation through regulation of CH methylation.  Although embryonic brain development and neurodegeneration have received considerable attention, the events that govern postnatal brain maturation are less understood. Here, we identify the miR-29 family to be strikingly induced during the late stages of brain maturation. Brain maturation is associated with a transient, postnatal period of de novo non-CG (CH) DNA methylation mediated by DNMT3A. We examine whether an important function of miR-29 during brain maturation is to restrict the period of CH methylation via its targeting of Dnmt3a. Deletion of miR-29 in the brain, or knockin mutations preventing miR-29 to specifically target Dnmt3a, result in increased DNMT3A expression, higher CH methylation, and repression of genes associated with neuronal activity and neuropsychiatric disorders. These mouse models also develop neurological deficits and premature lethality. Our results identify an essential role for miR-29 in restricting CH methylation in the brain and illustrate the importance of CH methylation regulation for normal brain maturation. |
| DNMT3A, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33238114  Title: DNMT3A Haploinsufficiency Results in Behavioral Deficits and Global Epigenomic Dysregulation Shared across Neurodevelopmental Disorders.  Mutations in DNA methyltransferase 3A (DNMT3A) have been detected in autism and related disorders, but how these mutations disrupt nervous system function is unknown. Here, we define the effects of DNMT3A mutations associated with neurodevelopmental disease. We show that diverse mutations affect different aspects of protein activity but lead to shared deficiencies in neuronal DNA methylation. Heterozygous DNMT3A knockout mice mimicking DNMT3A disruption in disease display growth and behavioral alterations consistent with human phenotypes. Strikingly, in these mice, we detect global disruption of neuron-enriched non-CG DNA methylation, a binding site for the Rett syndrome protein MeCP2. Loss of this methylation leads to enhancer and gene dysregulation that overlaps with models of Rett syndrome and autism. These findings define the effects of DNMT3A haploinsufficiency in the brain and uncover disruption of the non-CG methylation pathway as a convergence point across neurodevelopmental disorders. |
| DNMT3A |
| Url: https://pubmed.ncbi.nlm.nih.gov/31749842  Title: Epigenetic Regulation of the Ontogenic Expression of the Dopamine Transporter.  The dopamine transporter (DAT) is a plasma membrane transport protein responsible for regulating the duration and intensity of dopaminergic signaling. Altered expression of DAT is linked to neurodevelopmental disorders, including attention deficit hyperactivity disorder and autism spectrum disorder, and is shown to contribute to the response of psychotropic drugs and neurotoxicants. Although the postnatal levels of DAT have been characterized, there are few data regarding the mechanisms that regulate postnatal DAT expression. Here, we examine the ontogeny of DAT mRNA from postnatal days 0 to 182 in the rat brain and define a role for epigenetic mechanisms regulating DAT expression. DAT mRNA and protein significantly increased between PND 0 and 6 months in rat midbrain and striatum, respectively. The epigenetic modifiers Dnmt1, Dnmt3a, Dnmt3b, and Hdac2 demonstrated age associated decreases in mRNA expression whereas Hdac5 and Hdac8 showed increased mRNA expression with age. Chromatin immunoprecipitation studies revealed increased protein enrichment of acetylated histone 3 at lysines 9 and 14 and the dopaminergic transcription factors Nurr1 and Pitx3 within the DAT promoter in an age-related manner. Together these studies provide evidence for the role of epigenetic modifications in the regulation of DAT during development. The identification of these mechanisms may contribute to potential therapeutic interventions aimed at neurodevelopmental disorders of the dopaminergic system. |
| DNMT3A, KMT2A |
| Url: https://pubmed.ncbi.nlm.nih.gov/28386848  Title: Identification of De Novo DNMT3A Mutations That Cause West Syndrome by Using Whole-Exome Sequencing.  Epileptic encephalopathies (EEs) are a group of severe neurodevelopmental disorders with extreme genetic heterogeneity. Recent trio-based whole-exome sequencing (WES) studies have demonstrated that de novo mutations (DNMs) play prominent roles in severe EE. In this study, we searched for potential causal DNMs by using high-coverage WES of four unrelated Chinese parent-offspring trios affected by West syndrome. Through extensive bioinformatic analysis, we identified three novel DNMs in DNMT3A, CDKL5, and MAMDC2 in three trios and two compound heterozygous mutations in KMT2A in one trio. The DNMs in CDKL5 and DNMT3A were considered to be deleterious on the basis of the consensus of several genetic damage prediction tools. In addition, spatiotemporal expression patterns revealed a high level of DNMT3A expression during the early embryonic stage in nearly all brain regions. We also observed that certain high-confidence genes for epilepsy were shared among the co-expression and genetic interaction networks of DNMT3A, CDKL5, and KMT2A. Furthermore, all the candidate epilepsy genes in the co-expression network of DNMT3A were significantly enriched in the early developmental stages of the brain according to a rank-based enrichment test. In particular, we found that the DNMs of DNMT3A were shared among EE, autism spectrum disorder (ASD), and intellectual disability (ID) and mainly occurred in the functional domain of DNMT3A. Together, our findings support an association between DNMT3A mutations and EE susceptibility and suggest a shared molecular pathophysiology among EE and other neuropsychiatric disorders. |
| DNMT3A |
| Url: https://pubmed.ncbi.nlm.nih.gov/25423485  Title: Cerebellar oxidative DNA damage and altered DNA methylation in the BTBR T+tf/J mouse model of autism and similarities with human post mortem cerebellum.  The molecular pathogenesis of autism is complex and involves numerous genomic, epigenomic, proteomic, metabolic, and physiological alterations. Elucidating and understanding the molecular processes underlying the pathogenesis of autism is critical for effective clinical management and prevention of this disorder. The goal of this study is to investigate key molecular alterations postulated to play a role in autism and their role in the pathophysiology of autism. In this study we demonstrate that DNA isolated from the cerebellum of BTBR T+tf/J mice, a relevant mouse model of autism, and from human post-mortem cerebellum of individuals with autism, are both characterized by an increased levels of 8-oxo-7-hydrodeoxyguanosine (8-oxodG), 5-methylcytosine (5mC), and 5-hydroxymethylcytosine (5hmC). The increase in 8-oxodG and 5mC content was associated with a markedly reduced expression of the 8-oxoguanine DNA-glycosylase 1 (Ogg1) and increased expression of de novo DNA methyltransferases 3a and 3b (Dnmt3a and Dnmt3b). Interestingly, a rise in the level of 5hmC occurred without changes in the expression of ten-eleven translocation expression 1 (Tet1) and Tet2 genes, but significantly correlated with the presence of 8-oxodG in DNA. This finding and similar elevation in 8-oxodG in cerebellum of individuals with autism and in the BTBR T+tf/J mouse model warrant future large-scale studies to specifically address the role of OGG1 alterations in pathogenesis of autism. |
| DNMT3A, EN2, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25290267  Title: Elevated 5-hydroxymethylcytosine in the Engrailed-2 (EN-2) promoter is associated with increased gene expression and decreased MeCP2 binding in autism cerebellum.  Epigenetic mechanisms regulate programmed gene expression during prenatal neurogenesis and serve as a mediator between genetics and environment in postnatal life. The recent discovery of 5-hydroxymethylcytosine (5-hmC), with highest concentration in the brain, has added a new dimension to epigenetic regulation of neurogenesis and the development of complex behavior disorders. Here, we take a candidate gene approach to define the role 5-hmC in Engrailed-2 (EN-2) gene expression in the autism cerebellum. The EN-2 homeobox transcription factor, previously implicated in autism, is essential for normal cerebellar patterning and development. We previously reported EN-2 overexpression associated with promoter DNA hypermethylation in the autism cerebellum but because traditional DNA methylation methodology cannot distinguish 5-methylcytosine (5-mC) from 5-hmC, we now extend our investigation by quantifying global and gene-specific 5-mC and 5-hmC. Globally, 5-hmC was significantly increased in the autism cerebellum and accompanied by increases in the expression of de novo methyltransferases DNMT3A and DNMT3B, ten-eleven translocase genes TET1 and TET3, and in 8-oxo-deoxyguanosine (8-oxo-dG) content, a marker of oxidative DNA damage. Within the EN-2 promoter, there was a significant positive correlation between 5-hmC content and EN-2 gene expression. Based on reports of reduced MeCP2 affinity for 5-hmC, MeCP2 binding studies in the EN-2 promoter revealed a significant decrease in repressive MeCP2 binding that may contribute to the aberrant overexpression of EN-2. Because normal cerebellar development depends on perinatal EN-2 downregulation, the sustained postnatal overexpression suggests that a critical window of cerebellar development may have been missed in some individuals with autism with downstream developmental consequences. Epigenetic regulation of the programmed on-off switches in gene expression that occur at birth and during early brain development warrants further investigation. |
| DNMT3A, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23876774  Title: The epigenetic switches for neural development and psychiatric disorders.  The most remarkable feature of the nervous system is that the development and functions of the brain are largely reshaped by postnatal experiences, in joint with genetic landscapes. The nature vs. nurture argument reminds us that both genetic and epigenetic information is indispensable for the normal function of the brain. The epigenetic regulatory mechanisms in the central nervous system have been revealed over last a decade. Moreover, the mutations of epigenetic modulator genes have been shown to be implicated in neuropsychiatric disorders, such as autism spectrum disorders. The epigenetic study has initiated in the neuroscience field for a relative short period of time. In this review, we will summarize recent discoveries about epigenetic regulation on neural development, synaptic plasticity, learning and memory, as well as neuropsychiatric disorders. Although the comprehensive view of how epigenetic regulation contributes to the function of the brain is still not completed, the notion that brain, the most complicated organ of organisms, is profoundly shaped by epigenetic switches is widely accepted. |
| DNMT3A, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22343140  Title: Long-lived epigenetic interactions between perinatal PBDE exposure and Mecp2308 mutation.  The widespread use of persistent organic polybrominated diphenyl ethers (PBDEs) as commercial flame retardants has raised concern about potential long-lived effects on human health. Epigenetic mechanisms, such as DNA methylation, are responsive to environmental influences and have long-lasting consequences. Autism spectrum disorders (ASDs) have complex neurodevelopmental origins whereby both genetic and environmental factors are implicated. Rett syndrome is an X-linked ASD caused by mutations in the epigenetic factor methyl-CpG binding protein 2 (MECP2). In this study, an Mecp2 truncation mutant mouse (Mecp2(308)) with social behavioral defects was used to explore the long-lasting effects of PBDE exposure in a genetically and epigenetically susceptible model. Mecp2(308/+) dams were perinatally exposed daily to 2,2',4,4'-tetrabromodiphenyl ether 47 (BDE-47) and bred to wild-type C57BL/6J males, and the offspring of each sex and genotype were examined for developmental, behavioral and epigenetic outcomes. Perinatal BDE-47 exposure negatively impacted fertility of Mecp2(308/+) dams and preweaning weights of females. Global hypomethylation of adult brain DNA was observed specifically in female offspring perinatally exposed to BDE-47 and it coincided with reduced sociability in a genotype-independent manner. A reversing interaction of Mecp2 genotype on BDE-47 exposure was observed in a short-term memory test of social novelty that corresponded to increased Dnmt3a levels specifically in BDE-47-exposed Mecp2(308/+) offspring. In contrast, learning and long-term memory in the Morris water maze was impaired by BDE-47 exposure in female Mecp2(308/+) offspring. These results demonstrate that a genetic and environmental interaction relevant to social and cognitive behaviors shows sexual dimorphism, epigenetic dysregulation, compensatory molecular mechanisms and specific behavioral deficits. |
| DVL3, ZFYVE26 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34130600  Title: The spectrum of neurodevelopmental, neuromuscular and neurodegenerative disorders due to defective autophagy.  Primary dysfunction of autophagy due to Mendelian defects affecting core components of the autophagy machinery or closely related proteins have recently emerged as an important cause of genetic disease. This novel group of human disorders may present throughout life and comprises severe early-onset neurodevelopmental and more common adult-onset neurodegenerative disorders. Early-onset (or congenital) disorders of autophagy often share a recognizable "clinical signature," including variable combinations of neurological, neuromuscular and multisystem manifestations. Structural CNS abnormalities, cerebellar involvement, spasticity and peripheral nerve pathology are prominent neurological features, indicating a specific vulnerability of certain neuronal populations to autophagic disturbance. A typically biphasic disease course of late-onset neurodegeneration occurring on the background of a neurodevelopmental disorder further supports a role of autophagy in both neuronal development and maintenance. Additionally, an associated myopathy has been characterized in several conditions. The differential diagnosis comprises a wide range of other multisystem disorders, including mitochondrial, glycogen and lysosomal storage disorders, as well as ciliopathies, glycosylation and vesicular trafficking defects. The clinical overlap between the congenital disorders of autophagy and these conditions reflects the multiple roles of the proteins and/or emerging molecular connections between the pathways implicated and suggests an exciting area for future research. Therapy development for congenital disorders of autophagy is still in its infancy but may result in the identification of molecules that target autophagy more specifically than currently available compounds. The close connection with adult-onset neurodegenerative disorders highlights the relevance of research into rare early-onset neurodevelopmental conditions for much more common, age-related human diseases.Abbreviations: AC: anterior commissure; AD: Alzheimer disease; ALR: autophagic lysosomal reformation; ALS: amyotrophic lateral sclerosis; AMBRA1: autophagy and beclin 1 regulator 1; AMPK: AMP-activated protein kinase; ASD: autism spectrum disorder; ATG: autophagy related; BIN1: bridging integrator 1; BPAN: beta-propeller protein associated neurodegeneration; CC: corpus callosum; CHMP2B: charged multivesicular body protein 2B; CHS: Chediak-Higashi syndrome; CMA: chaperone-mediated autophagy; CMT: Charcot-Marie-Tooth disease; CNM: centronuclear myopathy; CNS: central nervous system; DNM2: dynamin 2; DPR: dipeptide repeat protein; DVL3: disheveled segment polarity protein 3; EPG5: ectopic P-granules autophagy protein 5 homolog; ER: endoplasmic reticulum; ESCRT: homotypic fusion and protein sorting complex; FIG4: FIG4 phosphoinositide 5-phosphatase; FTD: frontotemporal dementia; GBA: glucocerebrosidase; GD: Gaucher disease; GRN: progranulin; GSD: glycogen storage disorder; HC: hippocampal commissure; HD: Huntington disease; HOPS: homotypic fusion and protein sorting complex; HSPP: hereditary spastic paraparesis; LAMP2A: lysosomal associated membrane protein 2A; MEAX: X-linked myopathy with excessive autophagy; mHTT: mutant huntingtin; MSS: Marinesco-Sjoegren syndrome; MTM1: myotubularin 1; MTOR: mechanistic target of rapamycin kinase; NBIA: neurodegeneration with brain iron accumulation; NCL: neuronal ceroid lipofuscinosis; NPC1: Niemann-Pick disease type 1; PD: Parkinson disease; PtdIns3P: phosphatidylinositol-3-phosphate; RAB3GAP1: RAB3 GTPase activating protein catalytic subunit 1; RAB3GAP2: RAB3 GTPase activating non-catalytic protein subunit 2; RB1: RB1-inducible coiled-coil protein 1; RHEB: ras homolog, mTORC1 binding; SCAR20: SNX14-related ataxia; SENDA: static encephalopathy of childhood with neurodegeneration in adulthood; SNX14: sorting nexin 14; SPG11: SPG11 vesicle trafficking associated, spatacsin; SQSTM1: sequestosome 1; TBC1D20: TBC1 domain family member 20; TECPR2: tectonin beta-propeller repeat containing 2; TSC1: TSC complex subunit 1; TSC2: TSC complex subunit 2; UBQLN2: ubiquilin 2; VCP: valosin-containing protein; VMA21: vacuolar ATPase assembly factor VMA21; WDFY3/ALFY: WD repeat and FYVE domain containing protein 3; WDR45: WD repeat domain 45; WDR47: WD repeat domain 47; WMS: Warburg Micro syndrome; XLMTM: X-linked myotubular myopathy; ZFYVE26: zinc finger FYVE-type containing 26. |
| DVL3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26830142  Title: Prenatal β-catenin/Brn2/Tbr2 transcriptional cascade regulates adult social and stereotypic behaviors.  Social interaction is a fundamental behavior in all animal species, but the developmental timing of the social neural circuit formation and the cellular and molecular mechanisms governing its formation are poorly understood. We generated a mouse model with mutations in two Disheveled genes, Dvl1 and Dvl3, that displays adult social and repetitive behavioral abnormalities associated with transient embryonic brain enlargement during deep layer cortical neuron formation. These phenotypes were mediated by the embryonic expansion of basal neural progenitor cells (NPCs) via deregulation of a β-catenin/Brn2/Tbr2 transcriptional cascade. Transient pharmacological activation of the canonical Wnt pathway during this period of early corticogenesis rescued the β-catenin/Brn2/Tbr2 transcriptional cascade and the embryonic brain phenotypes. Remarkably, this embryonic treatment prevented adult behavioral deficits and partially rescued abnormal brain structure in Dvl mutant mice. Our findings define a mechanism that links fetal brain development and adult behavior, demonstrating a fetal origin for social and repetitive behavior deficits seen in disorders such as autism. |
| EBF3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34256850  Title: Coding and noncoding variants in EBF3 are involved in HADDS and simplex autism.  Previous research in autism and other neurodevelopmental disorders (NDDs) has indicated an important contribution of protein-coding (coding) de novo variants (DNVs) within specific genes. The role of de novo noncoding variation has been observable as a general increase in genetic burden but has yet to be resolved to individual functional elements. In this study, we assessed whole-genome sequencing data in 2671 families with autism (discovery cohort of 516 families, replication cohort of 2155 families). We focused on DNVs in enhancers with characterized in vivo activity in the brain and identified an excess of DNVs in an enhancer named hs737. We adapted the fitDNM statistical model to work in noncoding regions and tested enhancers for excess of DNVs in families with autism. We found only one enhancer (hs737) with nominal significance in the discovery (p = 0.0172), replication (p = 2.5 × 10-3), and combined dataset (p = 1.1 × 10-4). Each individual with a DNV in hs737 had shared phenotypes including being male, intact cognitive function, and hypotonia or motor delay. Our in vitro assessment of the DNVs showed they all reduce enhancer activity in a neuronal cell line. By epigenomic analyses, we found that hs737 is brain-specific and targets the transcription factor gene EBF3 in human fetal brain. EBF3 is genome-wide significant for coding DNVs in NDDs (missense p = 8.12 × 10-35, loss-of-function p = 2.26 × 10-13) and is widely expressed in the body. Through characterization of promoters bound by EBF3 in neuronal cells, we saw enrichment for binding to NDD genes (p = 7.43 × 10-6, OR = 1.87) involved in gene regulation. Individuals with coding DNVs have greater phenotypic severity (hypotonia, ataxia, and delayed development syndrome [HADDS]) in comparison to individuals with noncoding DNVs that have autism and hypotonia. In this study, we identify DNVs in the hs737 enhancer in individuals with autism. Through multiple approaches, we find hs737 targets the gene EBF3 that is genome-wide significant in NDDs. By assessment of noncoding variation and the genes they affect, we are beginning to understand their impact on gene regulatory networks in NDDs. |
| EBF3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34177436  Title: Neuroimaging Findings in Patients with EBF3 Mutations: Report of Two Cases.  Early B cell factor 3 (EBF3) is a transcription factor involved in brain development. Heterozygous, loss-of-function mutations in EBF3 have been reported in an autosomal dominant neurodevelopmental syndrome characterized by hypotonia, ataxia, and developmental delay (sometimes described as "HADD"s). We report 2 unrelated cases with novel de novo EBF3 mutations: c.455G>T (p.Arg152Leu) and c.962dup (p.Tyr321\*) to expand the genotype/phenotype correlations of this disorder; clinical, neuropsychological, and MRI studies were used to define the phenotype. IQ was in the normal range and diffusion tensor imaging revealed asymmetric alterations of the longitudinal fasciculus in both cases. Our results demonstrate that EBF3 mutations can underlie neurodevelopmental disorders without intellectual disability. Long tract abnormalities have not been previously recognized and suggest that they may be an unrecognized and characteristic feature in this syndrome. |
| EBF3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29162653  Title: De novo variants in EBF3 are associated with hypotonia, developmental delay, intellectual disability, and autism.  Using whole-exome sequencing, we identified seven unrelated individuals with global developmental delay, hypotonia, dysmorphic facial features, and an increased frequency of short stature, ataxia, and autism with de novo heterozygous frameshift, nonsense, splice, and missense variants in the Early B-cell Transcription Factor Family Member 3 (EBF3) gene. EBF3 is a member of the collier/olfactory-1/early B-cell factor (COE) family of proteins, which are required for central nervous system (CNS) development. COE proteins are highly evolutionarily conserved and regulate neuronal specification, migration, axon guidance, and dendritogenesis during development and are essential for maintaining neuronal identity in adult neurons. Haploinsufficiency of EBF3 may affect brain development and function, resulting in developmental delay, intellectual disability, and behavioral differences observed in individuals with a deleterious variant in EBF3. |
| EGR3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32698690  Title: Single administration of resveratrol improves social behavior in adult mouse models of autism spectrum disorder.  Resveratrol (RSV) is a natural polyphenol present in grapes, the skin of peanuts, and several other plants with many health benefits. Autism spectrum disorder (ASD) is a neurodevelopmental disorder that may be linked to neural and synaptic development impairments. The present study aimed to analyze the preventive effects of RSV on the development of ASD-like behavior, using oxytocin receptor gene knockout (Oxtr-KO) and valproic acid-induced ASD (VPA-ASD) model mice. Genetic deficiencies in Oxtr are suggested to be involved in ASD etiology. Twenty-four hours after a single RSV injection to the Oxtr-KO mice, the social impairments caused by OXTR deficiency were ameliorated. RSV also improved social impairments in the VPA-ASD mice. Administration of RSV up-regulated silent information regulator 1 (Sirt1) gene and early growth response factor 3 (Egr3) gene expressions in the amygdala of the Oxtr-KO mice. Our data suggest that RSV may have therapeutic effects on ASD with multiple targets. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36239013  Title: Protein kinase C beta relieves autism-like behavior in EN2 knockout mice via upregulation of the FTO/PGC-1α/UCP1 axis.  Increasing evidence suggests that disruption of neuron activity contributes to the autistic phenotype. Thus, we aimed in this study to explore the role of protein kinase C beta (PKCβ) in the regulation of neuron activity in an autism model. The expression of PKCβ in the microarray data of autism animal models was obtained from the Gene Expression Omnibus database. Then, mice with autism-like behavior were prepared in EN2 knockout (-/- ) mice. The interaction between PKCβ on fat mass and obesity-associated protein (FTO) as well as between PGC-1α and uncoupling protein 1 (UCP1) were characterized. The effect of FTO on the N6 -methyladenosine (m6A) modification level of proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α) was assayed. Following transfection of overexpressed PKCβ and/or silenced UCP1, effects of PKCβ and UCP1 in autism-like behaviors in EN2-/- mice were analyzed. Results showed that PKCβ was downregulated in EN2-/- mouse brain tissues or neurons. PKCβ promoted the expression and stability of FTO, which downregulated the m6A modification level of PGC-1α to promote its expression. Moreover, PGC-1α positively targeted the expression of UCP1. PKCβ knockdown enhanced sociability and spatial exploration ability, and reduced neuron apoptosis in EN2-/- mouse models of autism, which was reversed by UCP1 overexpression. Collectively, PKCβ overexpression leads to activation of the FTO/m6A/PGC-1α/UCP1 axis, thus inhibiting neuron apoptosis and providing neuroprotection in mice with autism-like behavior. |
| EN2, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35081728  Title: Correlation among maternal risk factors, gene methylation and disease severity in females with autism spectrum disorder.  Aim: To detect early-life environmental factors leading to DNA methylation changes of autism spectrum disorder (ASD)-related genes in young ASD females and reveal epigenetic biomarkers of disease severity. Materials & methods: We investigated blood methylation levels of MECP2, OXTR, BDNF, RELN, BCL2, EN2 and HTR1A genes in 42 ASD females. Results: Maternal gestational weight gain correlated with BDNF methylation levels (Bonferroni-corrected p = 0.034), and lack of folic acid supplementation at periconception resulted in higher disease severity in the ASD children (Bonferroni-corrected p = 0.048). RELN methylation levels were inversely correlated with disease severity (Bonferroni corrected p = 0.042). Conclusion: The present study revealed gene-environment interactions and potential epigenetic biomarkers of disease severity in ASD females. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32244845  Title: Altered Expression of GABAergic Markers in the Forebrain of Young and Adult Engrailed-2 Knockout Mice.  Impaired function of GABAergic interneurons, and the subsequent alteration of excitation/inhibition balance, is thought to contribute to autism spectrum disorders (ASD). Altered numbers of GABAergic interneurons and reduced expression of GABA receptors has been detected in the brain of ASD subjects and mouse models of ASD. We previously showed a reduced expression of GABAergic interneuron markers parvalbumin (PV) and somatostatin (SST) in the forebrain of adult mice lacking the Engrailed2 gene (En2-/- mice). Here, we extended this analysis to postnatal day (P) 30 by using in situ hybridization, immunohistochemistry, and quantitative RT-PCR to study the expression of GABAergic interneuron markers in the hippocampus and somatosensory cortex of En2-/- and wild type (WT) mice. In addition, GABA receptor subunit mRNA expression was investigated by quantitative RT-PCR in the same brain regions of P30 and adult En2-/- and WT mice. As observed in adult animals, PV and SST expression was decreased in En2-/- forebrain of P30 mice. The expression of GABA receptor subunits (including the ASD-relevant Gabrb3) was also altered in young and adult En2-/- forebrain. Our results suggest that GABAergic neurotransmission deficits are already evident at P30, confirming that neurodevelopmental defects of GABAergic interneurons occur in the En2 mouse model of ASD. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31972897  Title: Morphological alterations of the reticular thalamic nucleus in Engrailed-2 knockout mice.  The reticular thalamic nucleus (Rt) is a sheet of neurons that surrounds the dorsal thalamus laterally, along its dorso-ventral and rostro-caudal axes. It consists of inhibitory neurons releasing gamma-aminobutyric acid (GABA). This nucleus participates in the circuitry between the thalamus and the cerebral cortex, and its impairment is associated with neuro-psychiatric disorders. In this study, we investigated the Rt anatomy of Engrailed-2 knockout mice (En2-/- ), a mouse model of autism spectrum disorder (ASD), using parvalbumin as an immunohistochemical marker. We compared 4- and 6-week-old wild type (WT) and En2-/- mice using various morphometric parameters: cell area, shape factor, circularity and cell density. Significant differences were present in 6-week-old male mice with different genetic background (WT vs. En2-/- ): the Rt neurons of En2-/- mice showed a bigger cell area, shape factor and circularity when compared with WT. Age (4 weeks vs. 6 weeks) influenced the shape factor of WT females, the circularity and cell density of En2-/- males, and the shape factor and circularity of En2-/- females. Gender affected cell density in 4-week-old WT mice, shape factor and cellularity of 6-week-old WT mice, and cell area, shape factor and cell density of En2-/- at 6 weeks. Intrasubject (left-right) asymmetry of Rt was never observed. These results show for the first time that sex- and age-related changes occur in the Rt GABAergic neurons of the En2-/- ASD mouse model. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30980901  Title: Retinal defects in mice lacking the autism-associated gene Engrailed-2.  Defective cortical processing of visual stimuli and altered retinal function have been described in autism spectrum disorder (ASD) patients. In keeping with these findings, anatomical and functional defects have been found in the visual cortex and retina of mice bearing mutations for ASD-associated genes. Here we sought to investigate the anatomy and function of the adult retina of Engrailed 2 knockout (En2-/-) mice, a model for ASD. Our results showed that En2 is expressed in all three nuclear layers of the adult retina. When compared to age-matched En2+/+ controls, En2-/- adult retinas showed a significant decrease in the number of calbindin+ horizontal cells, and a significant increase in calbindin+ amacrine/ganglion cells. The total number of ganglion cells was not altered in the adult En2-/- retina, as shown by Brn3a+ cell counts. In addition, En2-/- adult mice showed a significant reduction of photoreceptor (rhodopsin) and bipolar cell (Pcp2, PKCα) markers. Functional defects were also present in the retina of En2 mutants, as indicated by electroretinogram recordings showing a significant reduction in both a-wave and b-wave amplitude in En2-/- mice as compared to controls. These data show for the first time that anatomical and functional defects are present in the retina of the En2 ASD mouse model. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30593497  Title: Aberrant Somatosensory Processing and Connectivity in Mice Lacking Engrailed-2.  Overreactivity and defensive behaviors in response to tactile stimuli are common symptoms in autism spectrum disorder (ASD) patients. Similarly, somatosensory hypersensitivity has also been described in mice lacking ASD-associated genes such as Fmr1 (fragile X mental retardation protein 1). Fmr1 knock-out mice also show reduced functional connectivity between sensory cortical areas, which may represent an endogenous biomarker for their hypersensitivity. Here, we measured whole-brain functional connectivity in Engrailed-2 knock-out (En2-/-) adult mice, which show a lower expression of Fmr1 and anatomical defects common to Fmr1 knock-outs. MRI-based resting-state functional connectivity in adult En2-/- mice revealed significantly reduced synchronization in somatosensory-auditory/associative cortices and dorsal thalamus, suggesting the presence of aberrant somatosensory processing in these mutants. Accordingly, when tested in the whisker nuisance test, En2-/- but not WT mice of both sexes showed fear behavior in response to repeated whisker stimulation. En2-/- mice undergoing this test exhibited decreased c-Fos-positive neurons (a marker of neuronal activity) in layer IV of the primary somatosensory cortex and increased immunoreactive cells in the basolateral amygdala compared with WT littermates. Conversely, when tested in a sensory maze, En2-/- and WT mice spent a comparable time in whisker-guided exploration, indicating that whisker-mediated behaviors are otherwise preserved in En2 mutants. Therefore, fearful responses to somatosensory stimuli in En2-/- mice are accompanied by reduced basal connectivity of sensory regions, reduced activation of somatosensory cortex, and increased activation of the basolateral amygdala, suggesting that impaired somatosensory processing is a common feature in mice lacking ASD-related genes.SIGNIFICANCE STATEMENT Overreactivity to tactile stimuli is a common symptom in autism spectrum disorder (ASD) patients. Recent studies performed in mice bearing ASD-related mutations confirmed these findings. Here, we evaluated the behavioral response to whisker stimulation in mice lacking the ASD-related gene Engrailed-2 (En2-/- mice). Compared with WT controls, En2-/- mice showed reduced functional connectivity in the somatosensory cortex, which was paralleled by fear behavior, reduced activation of somatosensory cortex, and increased activation of the basolateral amygdala in response to repeated whisker stimulation. These results suggest that impaired somatosensory signal processing is a common feature in mice harboring ASD-related mutations. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30147646  Title: Clinical Phenotypes Associated to Engrailed 2 Gene Alterations in a Series of Neuropediatric Patients.  The engrailed homeobox protein (EN) plays an important role in the regionalization of the neural tube. EN distribution regulates the cerebellum and midbrain morphogenesis, as well as retinotectal synaptogenesis. In humans, the EN1 and EN2 genes code for the EN family of transcription factors. Genetic alterations in the expression of EN2 have been related to different neurologic conditions and more particularly to autism spectrum disorders (ASD). We aimed to study and compare the phenotypes of three series of patients: (1) patients with encephalic structural anomalies (ESA) and abnormalities in the genomic (DNA) and/or transcriptomic (RNAm) of EN2 (EN2-g), (2) ESA patients having other gene mutations (OG-g), and (3) ESA patients free of these mutations (NM-g). Subjects and Methods: We have performed a descriptive study on 109 patients who suffer from mental retardation (MR), cerebral palsy (CP), epilepsy (EP), and behavioral disorders (BD), showing also ESA in their encephalic MRI. We studied genomic DNA and transcriptional analysis (cDNA) on EN2 gene (EN2), and in other genes (OG): LIS1, PTAFR, PAFAH1B2, PAFAH1B3, FGF8, PAX2, D17S379, D17S1866, and SMG6 (D17S5), as a routine genetic diagnosis in ESA patients. Results: From 109 patients, fifteen meet the exclusion criteria. From the remaining 94 patients, 12 (12.8%) showed mutations in EN2 (EN2-g), 20 showed mutations in other studied genes (OG-g), and 62 did not showed any mutation (NM-g). All EN2-g patients, suffered from MR, nine EP, seven BD and four CP. The proportions of these phenotypes in EN2-g did not differ from those in the OG-g, but it was significantly higher when comparing EN2-g with NM-g (MR: p = 0.013; EP: p = 0.001; BD: p = 0.0001; CP: p = 0.07, ns). Groups EN2-g and OG-g showed a 100 and a 70% of comorbidity, respectively, being significantly (p = 0.04) greater than NM-group (62.9%). Conclusion: Our series reflects a significant effect of EN2 gene alterations in neurodevelopmental abnormalities associated to ESA. Conversely, although these EN2 related anomalies might represent a predisposition to develop brain diseases, our results did not support direct relationship between EN2 mutations and specific clinical phenotypes. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30115672  Title: Fluoxetine Affects Differentiation of Midbrain Dopaminergic Neurons In Vitro.  Recent meta-analyses found an association between prenatal exposure to the antidepressant fluoxetine (FLX) and an increased risk of autism in children. This developmental disorder has been related to dysfunctions in the brains' rewards circuitry, which, in turn, has been linked to dysfunctions in dopaminergic (DA) signaling. The present study investigated if FLX affects processes involved in dopaminergic neuronal differentiation. Mouse neuronal precursors were differentiated into midbrain dopaminergic precursor cells (mDPCs) and concomitantly exposed to clinically relevant doses of FLX. Subsequently, dopaminergic precursors were evaluated for expression of differentiation and stemness markers using quantitative polymerase chain reaction. FLX treatment led to increases in early regional specification markers orthodenticle homeobox 2 (Otx2) and homeobox engrailed-1 and -2 (En1 and En2). On the other hand, two transcription factors essential for midbrain dopaminergic (mDA) neurogenesis, LIM homeobox transcription factor 1 α (Lmx1a) and paired-like homeodomain transcription factor 3 (Pitx3) were downregulated by FLX treatment. The stemness marker nestin (Nes) was increased, whereas the neuronal differentiation marker β3-tubulin (Tubb3) decreased. Additionally, we observed that FLX modulates the expression of several genes associated with autism spectrum disorder and downregulates the estrogen receptors (ERs) α and β Further investigations using ERβ knockout (BERKO) mDPCs showed that FLX had no or even opposite effects on several of the genes analyzed. These findings suggest that FLX affects differentiation of the dopaminergic system by increasing production of dopaminergic precursors, yet decreasing their maturation, partly via interference with the estrogen system. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29964155  Title: Impaired Neuronal Differentiation of Neural Stem Cells Lacking the Engrailed-2 Gene.  The Engrailed-2 (En2) gene codes for a homeobox-containing transcription factor, involved in midbrain-hindbrain embryonic development. In postnatal brain, En2 is expressed in the ventral mesencephalon, cerebellum, hippocampus and neocortex. Two single-nucleotide polymorphisms (SNPs) that are associated to autism spectrum disorders (ASD) have been identified in the human EN2 gene. Accordingly, mice lacking the En2 homeodomain (En2hd/hd, referred to as En2-/-) show molecular, anatomical and behavioral "ASD-like" features. Among these, we previously showed a partial loss of GABAergic interneurons in the En2-/- postnatal hippocampus and neocortex, accompanied by a marked decrease of brain-derived neurotrophic factor (BDNF) signaling, a crucial determinant of GABAergic differentiation. In order to better investigate the role of En2 in GABAergic interneuron differentiation, we generated and subsequently differentiated neural stem cells (NSCs) from basal ganglia and neocortex of En2+/+ and En2-/- mouse embryos. Wild-type NSCs from both basal ganglia and neocortex express En2, while mutant ones do not, as expected. As compared to En2+/+ NSCs, En2-/- NSCs derived from basal ganglia show impaired GABAergic differentiation accompanied by a reduced expression of the BDNF receptor trkB. Conversely, En2-/- NSCs derived from the neocortex expressed high levels of trkB and readily differentiated into neurons, as En2+/+ NSCs. Our results suggest that En2 contributes to GABAergic neuron differentiation from basal ganglia NSCs through a trkB-dependent BDNF signaling, thus providing a possible explanation for the reduced number of GABAergic interneurons detected in the En2-/- postnatal forebrain. |
| EN2, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28534217  Title: Autism spectrum disorder-associated genes and the development of dentate granule cells.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder characterized by severe clinical symptoms such as the deficiency of the social communication, repetitive and stereotyped behaviors, and restricted interests. Although complex genetic and environmental factors are thought to contribute to the development of ASD, the precise etiologies are largely unknown. Neuroanatomical observations have been made of developmental abnormalities in different brain regions, including dentate gyrus of hippocampus, which is widely accepted as the center for learning and memory. However, little is known about what roles ASD-associated genes play in the development of hippocampal dentate granule cells. In this article, we summarized functions and pathophysiological significance of 6 representative ASD-associated genes, SEMA5A, PTEN, NLGN, EN-2, FMR1, and MECP2, by focusing on the development of dentate gyrus. We then introduced a recently developed gene transfer method directed to neonatal dentate granule cells. This new method will be useful for elucidating physiological as well as pathophysiological significance of ASD-associated genes in the development of hippocampal formation. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27610074  Title: Comparative Gene Expression Analysis of Two Mouse Models of Autism: Transcriptome Profiling of the BTBR and En2 (-/-) Hippocampus.  Autism spectrum disorders (ASD) are characterized by a high degree of genetic heterogeneity. Genomic studies identified common pathological processes underlying the heterogeneous clinical manifestations of ASD, and transcriptome analyses revealed that gene networks involved in synapse development, neuronal activity, and immune function are deregulated in ASD. Mouse models provide unique tools to investigate the neurobiological basis of ASD; however, a comprehensive approach to identify transcriptional abnormalities in different ASD models has never been performed. Here we used two well-recognized ASD mouse models, BTBR T(+) Itpr3 (tf) /J (BTBR) and Engrailed-2 knockout (En2 (-/-)), to identify conserved ASD-related molecular signatures. En2 (-/-) mice bear a mutation within the EN2 transcription factor homeobox, while BTBR is an inbred strain with unknown genetic defects. Hippocampal RNA samples from BTBR, En2 (-/-) and respective control (C57Bl/6J and En2 (+/+)) adult mice were assessed for differential gene expression using microarrays. A total of 153 genes were similarly deregulated in the BTBR and En2 (-/-) hippocampus. Mouse phenotype and gene ontology enrichment analyses were performed on BTBR and En2 (-/-) hippocampal differentially expressed genes (DEGs). Pathways represented in both BTBR and En2 (-/-) hippocampal DEGs included abnormal behavioral response and chemokine/MAP kinase signaling. Genes involved in abnormal function of the immune system and abnormal synaptic transmission/seizures were significantly represented among BTBR and En2 (-/-) DEGs, respectively. Interestingly, both BTBR and En2 (-/-) hippocampal DEGs showed a significant enrichment of ASD and schizophrenia (SCZ)-associated genes. Specific gene sets were enriched in the two models: microglial genes were significantly enriched among BTBR DEGs, whereas GABAergic/glutamatergic postsynaptic genes, FMRP-interacting genes and epilepsy-related genes were significantly enriched among En2 (-/-) DEGs. Weighted correlation network analysis (WGCNA) performed on BTBR and En2 (-/-) hippocampal transcriptomes together identified six modules significantly enriched in ASD-related genes. Each of these modules showed a specific enrichment profile in neuronal and glial genes, as well as in genes associated to ASD comorbidities such as epilepsy and SCZ. Our data reveal significant transcriptional similarities and differences between the BTBR and En2 (-/-) hippocampus, indicating that transcriptome analysis of ASD mouse models may contribute to identify novel molecular targets for pharmacological studies. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27080080  Title: Ketogenic diet exposure during the juvenile period increases social behaviors and forebrain neural activation in adult Engrailed 2 null mice.  Prolonged consumption of ketogenic diets (KD) has reported neuroprotective benefits. Several studies suggest KD interventions could be useful in the management of neurological and developmental disorders. Alterations in the Engrailed (En) genes, specifically Engrailed 2 (En2), have neurodevelopmental consequences and produce autism-related behaviors. The following studies used En2 knockout (KO; En2(-/-)), and wild-type (WT; En2(+/+)), male mice fed either KD (80% fat, 0.1% carbohydrates) or control diet (CD; 10% fat, 70% carbohydrates). The objective was to determine whether a KD fed from weaning at postnatal day (PND) 21 to adulthood (PND 60) would alter brain monoamines concentrations, previously found dysregulated, and improve social outcomes. In WT animals, there was an increase in hypothalamic norepinephrine content in the KD-fed group. However, regional monoamines were not altered in KO mice in KD-fed compared with CD-fed group. In order to determine the effects of juvenile exposure to KD in mice with normal blood ketone levels, separate experiments were conducted in mice removed from the KD or CD and fed standard chow for 2days (PND 62). In a three-chamber social test with a novel mouse, KO mice previously exposed to the KD displayed similar social and self-grooming behaviors compared with the WT group. Groups previously exposed to a KD, regardless of genotype, had more c-Fos-positive cells in the cingulate cortex, lateral septal nuclei, and anterior bed nucleus of the stria terminalis. In the novel object condition, KO mice previously exposed to KD had similar behavioral responses and pattern of c-Fos immunoreactivity compared with the WT group. Thus, juvenile exposure to KD resulted in short-term consequences of improving social interactions and appropriate exploratory behaviors in a mouse model that displays autism-related behaviors. Such findings further our understanding of metabolic-based therapies for neurological and developmental disorders. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26987954  Title: Brain-derived neurotrophic factor signaling is altered in the forebrain of Engrailed-2 knockout mice.  Engrailed-2 (En2), a homeodomain transcription factor involved in regionalization and patterning of the midbrain and hindbrain regions has been associated to autism spectrum disorders (ASDs). En2 knockout (En2(-/-)) mice show ASD-like features accompanied by a significant loss of GABAergic subpopulations in the hippocampus and neocortex. Brain-derived neurotrophic factor (BDNF) is a crucial factor for the postnatal development of forebrain GABAergic neurons, and altered GABA signaling has been hypothesized to underlie the symptoms of ASD. Here we sought to determine whether interneuron loss in the En2(-/-) forebrain might be related to altered expression of BDNF and its signaling receptors. We first evaluated the expression of different BDNF mRNA isoforms in the neocortex and hippocampus of wild-type (WT) and En2(-/-) mice. Quantitative RT-PCR showed a marked down-regulation of several splicing variants of BDNF mRNA in the neocortex but not hippocampus of adult En2(-/-) mice, as compared to WT controls. Accordingly, levels of mature BDNF protein were lower in the neocortex but not hippocampus of En2(-/-) mice, as compared to WT. Increased levels of phosphorylated TrkB and decreased levels of p75 receptor were also detected in the neocortex of mutant mice. Accordingly, the expression of low density lipoprotein receptor (LDLR) and RhoA, two genes regulated via p75 was significantly altered in forebrain areas of mutant mice. These data indicate that BDNF signaling alterations might be involved in the anatomical changes observed in the En2(-/-) forebrain and suggest a pathogenic role of altered BDNF signaling in this mouse model of ASD. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26220976  Title: Engrailed-2 (En2) deletion produces multiple neurodevelopmental defects in monoamine systems, forebrain structures and neurogenesis and behavior.  Many genes involved in brain development have been associated with human neurodevelopmental disorders, but underlying pathophysiological mechanisms remain undefined. Human genetic and mouse behavioral analyses suggest that ENGRAILED-2 (EN2) contributes to neurodevelopmental disorders, especially autism spectrum disorder. In mouse, En2 exhibits dynamic spatiotemporal expression in embryonic mid-hindbrain regions where monoamine neurons emerge. Considering their importance in neuropsychiatric disorders, we characterized monoamine systems in relation to forebrain neurogenesis in En2-knockout (En2-KO) mice. Transmitter levels of serotonin, dopamine and norepinephrine (NE) were dysregulated from Postnatal day 7 (P7) to P21 in En2-KO, though NE exhibited the greatest abnormalities. While NE levels were reduced ∼35% in forebrain, they were increased 40 -: 75% in hindbrain and cerebellum, and these patterns paralleled changes in locus coeruleus (LC) fiber innervation, respectively. Although En2 promoter was active in Embryonic day 14.5 -: 15.5 LC neurons, expression diminished thereafter and gene deletion did not alter brainstem NE neuron numbers. Significantly, in parallel with reduced NE levels, En2-KO forebrain regions exhibited reduced growth, particularly hippocampus, where P21 dentate gyrus granule neurons were decreased 16%, suggesting abnormal neurogenesis. Indeed, hippocampal neurogenic regions showed increased cell death (+77%) and unexpectedly, increased proliferation. Excess proliferation was restricted to early Sox2/Tbr2 progenitors whereas increased apoptosis occurred in differentiating (Dcx) neuroblasts, accompanied by reduced newborn neuron survival. Abnormal neurogenesis may reflect NE deficits because intra-hippocampal injections of β-adrenergic agonists reversed cell death. These studies suggest that disruption of hindbrain patterning genes can alter monoamine system development and thereby produce forebrain defects that are relevant to human neurodevelopmental disorders. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26002543  Title: Serotonin abnormalities in Engrailed-2 knockout mice: New insight relevant for a model of Autism Spectrum Disorder.  Autism spectrum disorder (ASD) is a congenital neurodevelopmental behavioral disorder that appears in early childhood. Recent human genetic studies identified the homeobox transcription factor, Engrailed 2 (EN2), as a possible ASD susceptibility gene. En2 knockout mice (En2-/-) display subtle cerebellar neuropathological changes and reduced levels of tyrosine hydroxylase, noradrenaline and serotonin in the hippocampus and cerebral cortex similar to those ones which have been observed in the ASD brain. Furthermore other similarities link En2 knockout mice to ASD patients. Several lines of evidence suggest that serotonin may play an important role in the pathophysiology of the disease. In the present study we measured, by using an HPLC, the 5-HT levels in different brain areas and at different ages in En2-/- mice. In the frontal and occipital cortex, the content of 5HT was reduced in En2-/- 1 and 3 months old mice; in 6 month old mice, the difference was still present, but it was not statistically significant. The 5-HT content of cerebellar cortex was significantly reduced at 1 month old but significantly high when the KO mice reached 3 months of age. The increase was present even at 6 months of age. A similar trend was highlighted by SERT immunolabeling in En2-/- mice compared to control in the same areas and age analyzed. Our findings, in agreement with the current knowledge on the 5-HT system alterations in ASD, confirm the early neurotransmitter deficit with a late compensatory recovery in En2 KO-mice further suggesting that this experimental animal may be considered a good predictive model for the human disease. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25992917  Title: Immunohistochemical visualization of hippocampal neuron activity after spatial learning in a mouse model of neurodevelopmental disorders.  Induction of phosphorylated extracellular-regulated kinase (pERK) is a reliable molecular readout of learning-dependent neuronal activation. Here, we describe a pERK immunohistochemistry protocol to study the profile of hippocampal neuron activation following exposure to a spatial learning task in a mouse model characterized by cognitive deficits of neurodevelopmental origin. Specifically, we used pERK immunostaining to study neuronal activation following Morris water maze (MWM, a classical hippocampal-dependent learning task) in Engrailed-2 knockout (En2(-/-)) mice, a model of autism spectrum disorders (ASD). As compared to wild-type (WT) controls, En2(-/-) mice showed significant spatial learning deficits in the MWM. After MWM, significant differences in the number of pERK-positive neurons were detected in specific hippocampal subfields of En2(-/-) mice, as compared to WT animals. Thus, our protocol can robustly detect differences in pERK-positive neurons associated to hippocampal-dependent learning impairment in a mouse model of ASD. More generally, our protocol can be applied to investigate the profile of hippocampal neuron activation in both genetic or pharmacological mouse models characterized by cognitive deficits. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25463523  Title: Reduced phosphorylation of synapsin I in the hippocampus of Engrailed-2 knockout mice, a model for autism spectrum disorders.  Mice lacking the homeodomain transcription factor Engrailed-2 (En2(-/-) mice) are a well-characterized model for autism spectrum disorders (ASD). En2(-/-) mice present molecular, neuropathological and behavioral deficits related to ASD, including down-regulation of ASD-associated genes, cerebellar hypoplasia, interneuron loss, enhanced seizure susceptibility, decreased sociability and impaired cognition. Specifically, impaired spatial learning in the Morris water maze (MWM) is associated with reduced expression of neurofibromin and increased phosphorylation of extracellular-regulated kinase (ERK) in the hippocampus of En2(-/-) adult mice. In the attempt to better understand the molecular cascades underlying neurofibromin-dependent cognitive deficits in En2 mutant mice, we investigated the expression and phosphorylation of synapsin I (SynI; a major target of neurofibromin-dependent signaling) in the hippocampus of wild-type (WT) and En2(-/-) mice before and after MWM. Here we show that SynI mRNA and protein levels are down-regulated in the hippocampus of naïve and MWM-treated En2(-/-) mice, as compared to WT controls. This down-regulation is paralleled by reduced levels of SynI phosphorylation at Ser549 and Ser553 residues in the hilus of mutant mice, before and after MWM. These data indicate that in En2(-/-) hippocampus, neurofibromin-dependent pathways converging on SynI phosphorylation might underlie hippocampal-dependent learning deficits observed in En2(-/-) mice. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25274808  Title: Hippocampal dysregulation of neurofibromin-dependent pathways is associated with impaired spatial learning in engrailed 2 knock-out mice.  Genome-wide association studies indicated the homeobox-containing transcription factor Engrailed-2 (En2) as a candidate gene for autism spectrum disorders (ASD). Accordingly, En2 knock-out (En2(-/-)) mice show anatomical and behavioral "ASD-like" features, including decreased sociability and learning deficits. The molecular pathways underlying these deficits in En2(-/-) mice are not known. Deficits in signaling pathways involving neurofibromin and extracellular-regulated kinase (ERK) have been associated with impaired learning. Here we investigated the neurofibromin-ERK cascade in the hippocampus of wild-type (WT) and En2(-/-) mice before and after spatial learning testing. When compared with WT littermates, En2(-/-) mice showed impaired performance in the Morris water maze (MWM), which was accompanied by lower expression of the activity-dependent gene Arc. Quantitative RT-PCR, immunoblotting, and immunohistochemistry experiments showed a marked downregulation of neurofibromin expression in the dentate gyrus of both naive and MWM-treated En2(-/-) mice. ERK phosphorylation, known to be induced in the presence of neurofibromin deficiency, was increased in the dentate gyrus of En2(-/-) mice after MWM. Treatment of En2(-/-) mice with lovastatin, an indirect inhibitor of ERK phosphorylation, markedly reduced ERK phosphorylation in the dentate gyrus, but was unable to rescue learning deficits in MWM-trained mutant mice. Further investigation is needed to unravel the complex molecular mechanisms linking dysregulation of neurofibromin-dependent pathways to spatial learning deficits in the En2 mouse model of ASD. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25225635  Title: GH Dysfunction in Engrailed-2 Knockout Mice, a Model for Autism Spectrum Disorders.  Insulin-like growth factor 1 (IGF-1) signaling promotes brain development and plasticity. Altered IGF-1 expression has been associated to autism spectrum disorders (ASD). IGF-1 levels were found increased in the blood and decreased in the cerebrospinal fluid of ASD children. Accordingly, IGF-1 treatment can rescue behavioral deficits in mouse models of ASD, and IGF-1 trials have been proposed for ASD children. IGF-1 is mainly synthesized in the liver, and its synthesis is dependent on growth hormone (GH) produced in the pituitary gland. GH also modulates cognitive functions, and altered levels of GH have been detected in ASD patients. Here, we analyzed the expression of GH, IGF-1, their receptors, and regulatory hormones in the neuroendocrine system of adult male mice lacking the homeobox transcription factor Engrailed-2 (En2 (-/-) mice). En2 (-/-) mice display ASD-like behaviors (social interactions, defective spatial learning, increased seizure susceptibility) accompanied by relevant neuropathological changes (loss of cerebellar and forebrain inhibitory neurons). Recent studies showed that En2 modulates IGF-1 activity during postnatal cerebellar development. We found that GH mRNA expression was markedly deregulated throughout the neuroendocrine axis in En2 (-/-) mice, as compared to wild-type controls. In mutant mice, GH mRNA levels were significantly increased in the pituitary gland, blood, and liver, whereas decreased levels were detected in the hippocampus. These changes were paralleled by decreased levels of GH protein in the hippocampus but not other tissues of En2 (-/-) mice. IGF-1 mRNA was significantly up-regulated in the liver and down-regulated in the En2 (-/-) hippocampus, but no differences were detected in the levels of IGF-1 protein between the two genotypes. Our data strengthen the notion that altered GH levels in the hippocampus may be involved in learning disabilities associated to ASD. |
| EN2, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25199916  Title: Clustering autism: using neuroanatomical differences in 26 mouse models to gain insight into the heterogeneity.  Autism is a heritable disorder, with over 250 associated genes identified to date, yet no single gene accounts for >1-2% of cases. The clinical presentation, behavioural symptoms, imaging and histopathology findings are strikingly heterogeneous. A more complete understanding of autism can be obtained by examining multiple genetic or behavioural mouse models of autism using magnetic resonance imaging (MRI)-based neuroanatomical phenotyping. Twenty-six different mouse models were examined and the consistently found abnormal brain regions across models were parieto-temporal lobe, cerebellar cortex, frontal lobe, hypothalamus and striatum. These models separated into three distinct clusters, two of which can be linked to the under and over-connectivity found in autism. These clusters also identified previously unknown connections between Nrxn1α, En2 and Fmr1; Nlgn3, BTBR and Slc6A4; and also between X monosomy and Mecp2. With no single treatment for autism found, clustering autism using neuroanatomy and identifying these strong connections may prove to be a crucial step in predicting treatment response. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24987331  Title: Altered GABAergic markers, increased binocularity and reduced plasticity in the visual cortex of Engrailed-2 knockout mice.  The maturation of the GABAergic system is a crucial determinant of cortical development during early postnatal life, when sensory circuits undergo a process of activity-dependent refinement. An altered excitatory/inhibitory balance has been proposed as a possible pathogenic mechanism of autism spectrum disorders (ASD). The homeobox-containing transcription factor Engrailed-2 (En2) has been associated to ASD, and En2 knockout (En2 (-/-)) mice show ASD-like features accompanied by a partial loss of cortical GABAergic interneurons. Here we studied GABAergic markers and cortical function in En2 (-/-) mice, by exploiting the well-known anatomical and functional features of the mouse visual system. En2 is expressed in the visual cortex at postnatal day 30 and during adulthood. When compared to age-matched En2 (+/+) controls, En2 (-/-) mice showed an increased number of parvalbumin (PV(+)), somatostatin (SOM(+)), and neuropeptide Y (NPY(+)) positive interneurons in the visual cortex at P30, and a decreased number of SOM(+) and NPY(+) interneurons in the adult. At both ages, the differences in distinct interneuron populations observed between En2 (+/+) and En2 (-/-) mice were layer-specific. Adult En2 (-/-) mice displayed a normal eye-specific segregation in the retino-geniculate pathway, and in vivo electrophysiological recordings showed a normal development of basic functional properties (acuity, response latency, receptive field size) of the En2 (-/-) primary visual cortex. However, a significant increase of binocularity was found in P30 and adult En2 (-/-) mice, as compared to age-matched controls. Differently from what observed in En2 (+/+) mice, the En2 (-/-) primary visual cortex did not respond to a brief monocular deprivation performed between P26 and P29, during the so-called "critical period." These data suggest that altered GABAergic circuits impact baseline binocularity and plasticity in En2 (-/-) mice, while leaving other visual functional properties unaffected. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24730055  Title: Chronic desipramine treatment rescues depression-related, social and cognitive deficits in Engrailed-2 knockout mice.  Engrailed-2 (En2) is a homeobox transcription factor that regulates neurodevelopmental processes including neuronal connectivity and elaboration of monoaminergic neurons in the ventral hindbrain. We previously reported abnormalities in brain noradrenergic concentrations in En2 null mutant mice that were accompanied by increased immobility in the forced swim test, relevant to depression. An EN2 genetic polymorphism has been associated with autism spectrum disorders, and mice with a deletion in En2 display social abnormalities and cognitive deficits that may be relevant to multiple neuropsychiatric conditions. This study evaluated the ability of chronic treatment with desipramine (DMI), a selective norepinephrine (NE) reuptake inhibitor and classical antidepressant, to reverse behavioral abnormalities in En2−/− mice. Desipramine treatment significantly reduced immobility in the tail suspension and forced swim tests, restored sociability in the three-chambered social approach task and reversed impairments in contextual fear conditioning in En2−/− mice. Our findings indicate that modulation of brain noradrenergic systems rescues the depression-related phenotype in En2−/− mice and suggest new roles for NE in the pathophysiology of the social and cognitive deficits seen in neuropsychiatric disorders such as autism or schizophrenia. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24520327  Title: Autism associated gene, engrailed2, and flanking gene levels are altered in post-mortem cerebellum.  Previous genetic studies demonstrated association between the transcription factor engrailed2 (EN2) and Autism Spectrum Disorder (ASD). Subsequent molecular analysis determined that the EN2 ASD-associated haplotype (rs1861972-rs1861973 A-C) functions as a transcriptional activator to increase gene expression. EN2 is flanked by 5 genes, serotonin receptor5a (HTR5A), insulin induced gene1 (INSIG1), canopy1 homolog (CNPY1), RNA binding motif protein33 (RBM33), and sonic hedgehog (SHH). These flanking genes are co-expressed with EN2 during development and coordinate similar developmental processes. To investigate if mRNA levels for these genes are altered in individuals with autism, post-mortem analysis was performed. qRT-PCR quantified mRNA levels for EN2 and the 5 flanking genes in 78 post-mortem cerebellar samples. mRNA levels were correlated with both affection status and rs1861972-rs1861973 genotype. Molecular analysis investigated whether EN2 regulates flanking gene expression. EN2 levels are increased in affected A-C/G-T individuals (p = .0077). Affected individuals also display a significant increase in SHH and a decrease in INSIG1 levels. Rs1861972-rs1861973 genotype is correlated with significant increases for SHH (A-C/G-T) and CNPY1 (G-T/G-T) levels. Human cell line over-expression and knock-down as well as mouse knock-out analysis are consistent with EN2 and SHH being co-regulated, which provides a possible mechanism for increased SHH post-mortem levels. EN2 levels are increased in affected individuals with an A-C/G-T genotype, supporting EN2 as an ASD susceptibility gene. SHH, CNPY1, and INSIG1 levels are also significantly altered depending upon affection status or rs1861972-rs1861973 genotype. Increased EN2 levels likely contribute to elevated SHH expression observed in the post-mortem samples. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24507165  Title: Engrailed2 modulates cerebellar granule neuron precursor proliferation, differentiation and insulin-like growth factor 1 signaling during postnatal development.  The homeobox transcription factor Engrailed2 (En2) has been studied extensively in neurodevelopment, particularly in the midbrain/hindbrain region and cerebellum, where it exhibits dynamic patterns of expression and regulates cell patterning and morphogenesis. Because of its roles in regulating cerebellar development and evidence of cerebellar pathology in autism spectrum disorder (ASD), we previously examined an ENGRAILED2 association and found evidence to support EN2 as a susceptibility gene, a finding replicated by several other investigators. However, its functions at the cell biological level remain undefined. In the mouse, En2 gene is expressed in granule neuron precursors (GNPs) just as they exit the cell cycle and begin to differentiate, raising the possibility that En2 may modulate these developmental processes. To define En2 functions, we examined proliferation, differentiation and signaling pathway activation in En2 knockout (KO) and wild-type (WT) GNPs in response to a variety of extracellular growth factors and following En2 cDNA overexpression in cell culture. In vivo analyses of cerebellar GNP proliferation as well as responses to insulin-like growth factor-1 (IGF1) treatment were also conducted. Proliferation markers were increased in KO GNPs in vivo and in 24-h cultures, suggesting En2 normally serves to promote cell cycle exit. Significantly, IGF1 stimulated greater DNA synthesis in KO than WT cells in culture, a finding associated with markedly increased phospho-S6 kinase activation. Similarly, there was three-fold greater DNA synthesis in the KO cerebellum in response to IGF1 in vivo. On the other hand, KO GNPs exhibited reduced neurite outgrowth and differentiation. Conversely, En2 overexpression increased cell cycle exit and promoted neuronal differentiation. In aggregate, our observations suggest that the ASD-associated gene En2 promotes GNP cell cycle exit and differentiation, and modulates IGF1 activity during postnatal cerebellar development. Thus, genetic/epigenetic alterations of EN2 expression may impact proliferation, differentiation and IGF1 signaling as possible mechanisms that may contribute to ASD pathogenesis. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24355397  Title: Transcriptome profiling in engrailed-2 mutant mice reveals common molecular pathways associated with autism spectrum disorders.  Transcriptome analysis has been used in autism spectrum disorder (ASD) to unravel common pathogenic pathways based on the assumption that distinct rare genetic variants or epigenetic modifications affect common biological pathways. To unravel recurrent ASD-related neuropathological mechanisms, we took advantage of the En2-/- mouse model and performed transcriptome profiling on cerebellar and hippocampal adult tissues. Cerebellar and hippocampal tissue samples from three En2-/- and wild type (WT) littermate mice were assessed for differential gene expression using microarray hybridization followed by RankProd analysis. To identify functional categories overrepresented in the differentially expressed genes, we used integrated gene-network analysis, gene ontology enrichment and mouse phenotype ontology analysis. Furthermore, we performed direct enrichment analysis of ASD-associated genes from the SFARI repository in our differentially expressed genes. Given the limited number of animals used in the study, we used permissive criteria and identified 842 differentially expressed genes in En2-/- cerebellum and 862 in the En2-/- hippocampus. Our functional analysis revealed that the molecular signature of En2-/- cerebellum and hippocampus shares convergent pathological pathways with ASD, including abnormal synaptic transmission, altered developmental processes and increased immune response. Furthermore, when directly compared to the repository of the SFARI database, our differentially expressed genes in the hippocampus showed enrichment of ASD-associated genes significantly higher than previously reported. qPCR was performed for representative genes to confirm relative transcript levels compared to those detected in microarrays. Despite the limited number of animals used in the study, our bioinformatic analysis indicates the En2-/- mouse is a valuable tool for investigating molecular alterations related to ASD. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23423141  Title: Complex epigenetic regulation of engrailed-2 (EN-2) homeobox gene in the autism cerebellum.  The elucidation of epigenetic alterations in the autism brain has potential to provide new insights into the molecular mechanisms underlying abnormal gene expression in this disorder. Given strong evidence that engrailed-2 (EN-2) is a developmentally expressed gene relevant to cerebellar abnormalities and autism, the epigenetic evaluation of this candidate gene was undertaken in 26 case and control post-mortem cerebellar samples. Assessments included global DNA methylation, EN-2 promoter methylation, EN-2 gene expression and EN-2 protein levels. Chromatin immunoprecipitation was used to evaluate trimethylation status of histone H3 lysine 27 (H3K27) associated with gene downregulation and histone H3 lysine 4 (H3K4) associated with gene activation. The results revealed an unusual pattern of global and EN-2 promoter region DNA hypermethylation accompanied by significant increases in EN-2 gene expression and protein levels. Consistent with EN-2 overexpression, histone H3K27 trimethylation mark in the EN-2 promoter was significantly decreased in the autism samples relative to matched controls. Supporting a link between reduced histone H3K27 trimethylation and increased EN-2 gene expression, the mean level of histone H3K4 trimethylation was elevated in the autism cerebellar samples. Together, these results suggest that the normal EN-2 downregulation that signals Purkinje cell maturation during late prenatal and early-postnatal development may not have occurred in some individuals with autism and that the postnatal persistence of EN-2 overexpression may contribute to autism cerebellar abnormalities. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23360806  Title: Loss of GABAergic neurons in the hippocampus and cerebral cortex of Engrailed-2 null mutant mice: implications for autism spectrum disorders.  The homeobox-containing transcription factor Engrailed-2 (En2) is involved in patterning and neuronal differentiation of the midbrain/hindbrain region, where it is prominently expressed. En2 mRNA is also expressed in the adult mouse hippocampus and cerebral cortex, indicating that it might also function in these brain areas. Genome-wide association studies revealed that En2 is a candidate gene for autism spectrum disorders (ASD), and mice devoid of its expression (En2(-/-) mice) display anatomical, behavioral and clinical "autistic-like" features. Since reduced GABAergic inhibition has been proposed as a possible pathogenic mechanism of ASD, we hypothesized that the phenotype of En2(-/-) mice might include defective GABAergic innervation in the forebrain. Here we show that the Engrailed proteins are present in postnatal GABAergic neurons of the mouse hippocampus and cerebral cortex, and adult En2(-/-) mice show reduced expression of GABAergic marker mRNAs in these areas. In addition, reduction in parvalbumin (PV), somatostatin (SOM) and neuropeptide Y (NPY) expressing interneurons is detected in the hippocampus and cerebral cortex of adult En2(-/-) mice. Our results raise the possibility of a link between altered function of En2, anatomical deficits of GABAergic forebrain neurons and the pathogenesis of ASD. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22829897  Title: Autism-relevant social abnormalities and cognitive deficits in engrailed-2 knockout mice.  ENGRAILED 2 (En2), a homeobox transcription factor, functions as a patterning gene in the early development and connectivity of rodent hindbrain and cerebellum, and regulates neurogenesis and development of monoaminergic pathways. To further understand the neurobiological functions of En2, we conducted neuroanatomical expression profiling of En2 wildtype mice. RTQPCR assays demonstrated that En2 is expressed in adult brain structures including the somatosensory cortex, hippocampus, striatum, thalamus, hypothalamus and brainstem. Human genetic studies indicate that EN2 is associated with autism. To determine the consequences of En2 mutations on mouse behaviors, including outcomes potentially relevant to autism, we conducted comprehensive phenotyping of social, communication, repetitive, and cognitive behaviors. En2 null mutants exhibited robust deficits in reciprocal social interactions as juveniles and adults, and absence of sociability in adults, replicated in two independent cohorts. Fear conditioning and water maze learning were impaired in En2 null mutants. High immobility in the forced swim test, reduced prepulse inhibition, mild motor coordination impairments and reduced grip strength were detected in En2 null mutants. No genotype differences were found on measures of ultrasonic vocalizations in social contexts, and no stereotyped or repetitive behaviors were observed. Developmental milestones, general health, olfactory abilities, exploratory locomotor activity, anxiety-like behaviors and pain responses did not differ across genotypes, indicating that the behavioral abnormalities detected in En2 null mutants were not attributable to physical or procedural confounds. Our findings provide new insight into the role of En2 in complex behaviors and suggest that disturbances in En2 signaling may contribute to neuropsychiatric disorders marked by social and cognitive deficits, including autism spectrum disorders. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20877431  Title: Neural stem cell regulation, fibroblast growth factors, and the developmental origins of neuropsychiatric disorders.  There is increasing appreciation for the neurodevelopmental underpinnings of many psychiatric disorders. Disorders that begin in childhood such as autism, language disorders or mental retardation as well as adult-onset mental disorders may have origins early in neurodevelopment. Neural stem cells (NSCs) can be defined as self-renewing, multipotent cells that are present in both the embryonic and adult brain. Several recent research findings demonstrate that psychiatric illness may begin with abnormal specification, growth, expansion and differentiation of embryonic NSCs. For example, candidate susceptibility genes for schizophrenia, autism and major depression include the signaling molecule Disrupted In Schizophrenia-1 (DISC-1), the homeodomain gene engrailed-2 (EN-2), and several receptor tyrosine kinases, including brain-derived growth factor and fibroblast growth factors, all of which have been shown to play important roles in NSCs or neuronal precursors. We will discuss here stem cell biology, signaling factors that affect these cells, and the potential contribution of these processes to the etiology of neuropsychiatric disorders. Hypotheses about how some of these factors relate to psychiatric disorders will be reviewed. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20050924  Title: Family-based studies indicate association of Engrailed 2 gene with autism in an Indian population.  Engrailed 2 (EN2) is a homeobox transcription factor involved in the patterning of cerebellum during brain development. Linkage analysis and studies on knockout mice support EN2, located on chromosome 7q36.3, as a potential risk locus for autism. Candidate gene approach also suggested association of EN2 with autism spectrum disorder (ASD) in various populations. Here, we have investigated the association of five markers [rs3735653 (C/T) in exon 1; rs34808376 (GC/-) and rs6150410 (CGCATCCCC/-) in promoter region; rs1861972 (A/G) and rs1861973 (C/T) in the intron] of the gene with autism and ASD in Indian population using family-based approach. Probands have been recruited using Diagnostic and Statistical Manual of Mental Disorders Fourth Edition (DSM-IV) diagnostic criteria. Genotypic distributions conform to Hardy-Weinberg equilibrium. Genotyping analysis showed that the intronic single nucleotide polymorphisms (SNPs) are in complete linkage disequilibrium showing A-C and corresponding G-T allelic association. We observed significant preferential transmission of C allele of rs1861973 from the parents to affected offspring [transmission disequilibrium test (TDT): narrow diagnosis likelihood ratio statistics (LRS) = 6.63, P = 0.006; broad diagnosis LRS = 4.47, P = 0.05]. Interestingly, gender-based investigations showed significant transmission of C allele to the affected females [TDT: LRS = 7.36, P = 0.0025; haplotype-based haplotype relative risk (HHRR): LRS = 7.16, P = 0.02]. A maternal overtransmission for these alleles was also noted (TDT: LRS = 3.65, P = 0.036; HHRR: LRS = 2.81, P = 0.036). Bioinformatic analysis using TFSearch showed generation of Sp1 binding site in the presence of C allele. While Del-T haplotype formed from rs34808376-rs1861973 markers showed increased non-transmission, the Ins-C showed significant transmission suggesting protective effect and risk, respectively, conferred by these haplotypes in autism etiology. These results suggest positive genetic correlation of EN2 with autism in the Indian population. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19615670  Title: Autism-associated haplotype affects the regulation of the homeobox gene, ENGRAILED 2.  Association analysis identified the homeobox transcription factor, ENGRAILED 2 (EN2), as a possible autism spectrum disorder (ASD) susceptibility gene (ASD [MIM 608636]; EN2 [MIM 131310]). The common alleles (underlined) of two intronic single nucleotide polymorphisms (SNPs), rs1861972 (A/G) and rs1861973 (C/T), are over-transmitted to affected individuals both singly and as a haplotype in three separate datasets (518 families total, haplotype p = .00000035). Further support that EN2 is a possible ASD susceptibility gene requires the identification of a risk allele, a DNA variant that is consistently associated with ASD but is also functional. To identify possible risk alleles, additional association analysis and linkage disequilibrium (LD) mapping were performed. Candidate polymorphisms were then tested for functional differences by luciferase (Luc) reporter transfections and electrophoretic mobility shift assays (EMSAs). Association analysis of additional EN2 polymorphisms and LD mapping with Hapmap SNPs identified the rs1861972-rs1861973 haplotype as the most appropriate candidate to test for functional differences. Luciferase reporters for the two common rs1861972-rs1861973 haplotypes (A-C and G-T) were then transfected into human and rat cell lines as well as primary mouse neuronal cultures. In all cases the A-C haplotype resulted in a significant increase in Luc levels (p < .005). The EMSAs were then performed, and nuclear factors were bound specifically to the A and C alleles of both SNPs. These data indicate that the A-C haplotype is functional and, together with the association and LD mapping results, supports EN2 as a likely ASD susceptibility gene and the A-C haplotype as a possible risk allele. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19186208  Title: Increased susceptibility to kainic acid-induced seizures in Engrailed-2 knockout mice.  The En2 gene, coding for the homeobox-containing transcription factor Engrailed-2 (EN2), has been associated to autism spectrum disorder (ASD). Due to neuroanatomical and behavioral abnormalities, which partly resemble those observed in ASD patients, En2 knockout (En2(-/-)) mice have been proposed as a model for ASD. In the mouse embryo, En2 is involved in the specification of midbrain/hindbrain regions, being predominantly expressed in the developing cerebellum and ventral midbrain, and its expression is maintained in these structures until adulthood. Here we show that in the adult mouse brain, En2 mRNA is expressed also in the hippocampus and cerebral cortex. Hippocampal En2 mRNA content decreased after seizures induced by kainic acid (KA). This suggests that En2 might also influence the functioning of forebrain areas during adulthood and in response to seizures. Indeed, a reduced expression of parvalbumin and somatostatin was detected in the hippocampus of En2(-/-) mice as compared to wild-type (WT) mice, indicating an altered GABAergic innervation of limbic circuits in En2(-/-) mice. In keeping with these results, En2(-/-) mice displayed an increased susceptibility to KA-induced seizures. KA (20 mg/kg) determined more severe and prolonged generalized seizures in En2(-/-) mice, when compared to WT animals. Seizures were accompanied by a widespread c-fos and c-jun mRNA induction in the brain of En2(-/-) but not WT mice. Long-term histopathological changes (CA1 cell loss, upregulation of neuropeptide Y) also occurred in the hippocampus of KA-treated En2(-/-) but not WT mice. These findings suggest that En2(-/-) mice might be used as a novel tool to study the link between epilepsy and ASD. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19016890  Title: Social approach in genetically engineered mouse lines relevant to autism.  Profound impairment in social interaction is a core symptom of autism, a severe neurodevelopmental disorder. Deficits can include a lack of interest in social contact and low levels of approach and proximity to other children. In this study, a three-chambered choice task was used to evaluate sociability and social novelty preference in five lines of mice with mutations in genes implicated in autism spectrum disorders. Fmr1(tm1Cgr/Y)(Fmr1(-/y)) mice represent a model for fragile X, a mental retardation syndrome that is partially comorbid with autism. We tested Fmr1(-/y)mice on two genetic backgrounds, C57BL/6J and FVB/N-129/OlaHsd (FVB/129). Targeted disruption of Fmr1 resulted in low sociability on one measure, but only when the mutation was expressed on FVB/129. Autism has been associated with altered serotonin levels and polymorphisms in SLC6A4 (SERT), the serotonin transporter gene. Male mice with targeted disruption of Slc6a4 displayed significantly less sociability than wild-type controls. Mice with conditional overexpression of Igf-1 (insulin-like growth factor-1) offered a model for brain overgrowth associated with autism. Igf-1 transgenic mice engaged in levels of social approach similar to wild-type controls. Targeted disruption in other genes of interest, En2 (engrailed-2) and Dhcr7, was carried on genetic backgrounds that showed low levels of exploration in the choice task, precluding meaningful interpretations of social behavior scores. Overall, results show that loss of Fmr1 or Slc6a4 gene function can lead to deficits in sociability. Findings from the fragile X model suggest that the FVB/129 background confers enhanced susceptibility to consequences of Fmr1 mutation on social approach. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17055592  Title: The mouse Engrailed genes: a window into autism.  The complex behavioral symptoms and neuroanatomical abnormalities observed in autistic individuals strongly suggest a multi-factorial basis for this perplexing disease. Although not the perfect model, we believe the Engrailed genes provide an invaluable "window" into the elusive etiology of autism spectrum disorder. The Engrailed-2 gene has been associated with autism in genetic linkage studies. The En2 knock-out mouse harbors cerebellar abnormalities that are similar to those found in autistic individuals and, as we report here, has a distinct anterior shift in the position of the amygdala in the cerebral cortex. Our initial analysis of background effects in the En1 mouse knock-out provides insight as to possible molecular mechanisms and gender differences associated with autism. These findings further the connection between Engrailed and autism and provide new avenues to explore in the ongoing study of the biological basis of this multifaceted disease. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/16935268  Title: En2 knockout mice display neurobehavioral and neurochemical alterations relevant to autism spectrum disorder.  Autism spectrum disorder (ASD) is a prevalent and inheritable neurodevelopmental disorder. Recent human genetic studies are consistent with the homeobox transcription factor, ENGRAILED 2 (EN2), being an ASD susceptibility gene. En2 knockout mice (En2(-/-)) display subtle cerebellar neuropathological changes similar to what has been observed in the ASD brain. To investigate whether En2(-/-) mice displayed abnormal behavior relevant to ASD, they were monitored in tasks designed to assess social maturation as well as learning and memory. Deficits in social behavior were detected in En2(-/-) mice across maturation that included decreased play, reduced social sniffing and allogrooming, and less aggressive behavior. Deficits in two spatial learning and memory tasks were also observed. Because locomotor activity was a component of many of the behavioral tasks, this was measured at various stages of development. Locomotor activity was not compromised in the knockout. However, a more thorough analysis of motor behavior in En2(-/-) mice revealed deficits in specific motor tasks. To determine whether neurochemical changes were associated with these behavioral phenotypes, monoamine levels in specific brain regions were assessed. A cerebellar-specific increase in serotonin and its metabolite was observed. Interestingly, several reports have suggested that the serotonin pathway is affected in ASD. We conclude that En2(-/-) mice display behavioral and neurochemical changes, in addition to genetic and neuropathological changes, relevant to ASD. Therefore, these mice may be useful as an animal model of autism. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/16252243  Title: Support for the homeobox transcription factor gene ENGRAILED 2 as an autism spectrum disorder susceptibility locus.  Our previous research involving 167 nuclear families from the Autism Genetic Resource Exchange (AGRE) demonstrated that two intronic SNPs, rs1861972 and rs1861973, in the homeodomain transcription factor gene ENGRAILED 2 (EN2) are significantly associated with autism spectrum disorder (ASD). In this study, significant replication of association for rs1861972 and rs1861973 is reported for two additional data sets: an independent set of 222 AGRE families (rs1861972-rs1861973 haplotype, P=.0016) and a separate sample of 129 National Institutes of Mental Health families (rs1861972-rs1861973 haplotype, P=.0431). Association analysis of the haplotype in the combined sample of both AGRE data sets (389 families) produced a P value of .0000033, whereas combining all three data sets (518 families) produced a P value of .00000035. Population-attributable risk calculations for the associated haplotype, performed using the entire sample of 518 families, determined that the risk allele contributes to as many as 40% of ASD cases in the general population. Linkage disequilibrium (LD) mapping with the use of polymorphisms distributed throughout the gene has shown that only intronic SNPs are in strong LD with rs1861972 and rs1861973. Resequencing and association analysis of all intronic SNPs have identified alleles associated with ASD, which makes them candidates for future functional analysis. Finally, to begin defining the function of EN2 during development, mouse En2 was ectopically expressed in cortical precursors. Fewer En2-transfected cells than controls displayed a differentiated phenotype. Together, these data provide further genetic evidence that EN2 might act as an ASD susceptibility locus, and they suggest that a risk allele that perturbs the spatial/temporal expression of EN2 could significantly alter normal brain development. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/15024396  Title: Association of the homeobox transcription factor, ENGRAILED 2, 3, with autism spectrum disorder.  Mouse mutants of the homeobox transcription factor Engrailed2 (En2) and autistic individuals display similar cerebellar morphological abnormalities, which include hypoplasia and a decrease in the number of Purkinje cells. Human EN2 maps to 7q36, a chromosomal region that has demonstrated suggestive linkage to autism spectrum disorder (ASD). To investigate EN2 for evidence of association with ASD, four single-nucleotide polymorphisms (SNPs) (rs3735653, rs1861972, rs1861973, rs2361689) that span the majority of the 8.0 kb gene were assessed by the transmission/disequilibrium test. Initially, 138 triads of autistic individuals and their parents were tested. Two intronic SNPs (rs1861972 and rs1861973) demonstrated significant association with autism (rs1861972, P=0.0018; rs1861973, P=0.0003; haplotype, P=0.000005). Flanking exonic SNPs (rs3735653 and rs2361689) did not display association. This analysis was then extended to include 167 small nuclear ASD pedigrees and significant association was again only observed for rs1861972 and rs1861973 under both the narrow and broad diagnostic criteria (narrow: rs1861972 P=0.0290, rs1861973 P=0.0073, haplotype P=0.0009; broad: rs1861972 P=0.0175, rs1861973 P=0.0107, haplotype P=0.0024). These data demonstrate association between a cerebellar patterning gene and ASD, suggesting a role for EN2 as a susceptibility locus and supporting a neurodevelopmental defect hypothesis in the etiology of autism. |
| EN2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/10212552  Title: Etiology of infantile autism: a review of recent advances in genetic and neurobiological research.  The etiology of autism is complex, and in most cases the underlying pathologic mechanisms are unknown. Autism is a hetereogeneous disorder, diagnosed subjectively on the basis of a large number of criteria. Recent research has investigated genetics, in utero insults and brain function as well as neurochemical and immunological factors. On the basis of family and twin studies, there appears to be a genetic basis for a wide "autistic syndrome." About a quarter of cases of autism are associated with genetic disorders such as fragile X syndrome or with infectious diseases such as congenital rubella. Genetic studies have shown an association between autism markers of brain development such as 3 markers of the c-Harvey-ros oncogene and the homeobox gene EN2. In some cases, autism is associated with insults early in gestation, including thalidomide embryopathy. Autism may arise from abnormal central nervous system functioning, since most autistic patients have indications of brain dysfunction, and about half of them have abnormal electroencephalograms. Similarly, the pattern of evoked response potentials and conduction time is altered in autistic children. There is substantial evidence from neuroimaging studies that dysfunctions in the cerebellum and possibly the temporal lobe and association cortex occur in autistic symptoms. Neurochemical studies have investigated the role of serotonin, epinephrine and norepinephrine, since levels of these neurotransmitters are altered in autism, although other hypotheses implicate overactive brain opioid systems and changes in oxytocin neurotransmission. Autoimmunity may also play a role; antibodies against myelin basic protein are often found in children with autism, who also have increased eosinophil and basophil response to IgE-mediated reactions. In summary, the prevailing view is that autism is caused by a pathophysiologic process arising from the interaction of an early environmental insult and a genetic predisposition. |
| ERBIN, EZH2, YY1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33080834  Title: Comprehensive Analysis of RNA-Seq Gene Expression Profiling of Brain Transcriptomes Reveals Novel Genes, Regulators, and Pathways in Autism Spectrum Disorder.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder with deficits in social communication ability and repetitive behavior. The pathophysiological events involved in the brain of this complex disease are still unclear. In this study, we aimed to profile the gene expression signatures of brain cortex of ASD patients, by using two publicly available RNA-seq studies, in order to discover new ASD-related genes. We detected 1567 differentially expressed genes (DEGs) by meta-analysis, where 1194 were upregulated and 373 were downregulated genes. Several ASD-related genes previously reported were also identified. Our meta-analysis identified 235 new DEGs that were not detected using the individual RNA-seq studies used. Some of those genes, including seven DEGs (PAK1, DNAH17, DOCK8, DAPP1, PCDHAC2, and ERBIN, SLC7A7), have been confirmed in previous reports to be associated with ASD. Gene Ontology (GO) and pathways analysis showed several molecular pathways enriched by the DEGs, namely, osteoclast differentiation, TNF signaling pathway, complement and coagulation cascade. Topological analysis of protein-protein interaction of the ASD brain cortex revealed proteomics hub gene signatures: MYC, TP53, HDAC1, CDK2, BAG3, CDKN1A, GABARAPL1, EZH2, VIM, and TRAF1. We also identified the transcriptional factors (TFs) regulating DEGs, namely, FOXC1, GATA2, YY1, FOXL1, USF2, NFIC, NFKB1, E2F1, TFAP2A, HINFP. Novel core genes and molecular signatures involved with ASD were identified by our meta-analysis. |
| ERG |
| Url: https://pubmed.ncbi.nlm.nih.gov/36834288  Title: The Use of Optical Coherence Tomography and Electrophysiological Tests in the Early Diagnosis of Inflammatory Changes in the CNS in children with ASD-A Review of Contemporary Literature.  This article is a review of the contemporary literature on the possibility of using modern ophthalmological diagnostics, such as optical coherence tomography and electrophysiological tests, in the assessment of changes in eyesight correlating with inflammatory changes in the central nervous system (CNS) as one of the risk factors for neurodevelopmental disorders in children with ASD. A significant role is attributed to the activation of nerve and glial cells, as well as inflammatory changes in the brain, both of which can be of great importance in regard to an autism development predisposition. This fact indicates the possibility of using certain ophthalmic markers to depict an early correlation between the CNS and its outermost layer, i.e., the retina. A comprehensive ophthalmological assessment, and above all, characteristic changes in the functional function of photoreceptors and disorders of the structures of the retina or optic nerve fibers found in the latest OCT or ERG tests may in the future become diagnostic tools, further confirming the early characteristics of autism in children and adolescents. The above information, therefore, emphasizes the importance of cooperation between specialists in improving the diagnosis and treatment of children with autism. |
| ERG, FAN1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32712126  Title: Molecular, physiological and behavioral characterization of the heterozygous Df[h15q13]/+ mouse model associated with the human 15q13.3 microdeletion syndrome.  The human 15q13.3 microdeletion syndrome (DS) is caused by a heterozygous microdeletion (MD) affecting six genes: FAN1; MTMR10; TRPM1; KLF13; OTUD7A; and CHRNA7. Carriers are at risk for intellectual disability, epilepsy, autism spectrum disorder, and schizophrenia. Here we used the Df[h15q13]/+ mouse model with an orthologous deletion to further characterize molecular, neurophysiological, and behavioral parameters that are relevant to the 15q13.3 DS. First, we verified the expression and distribution of the α7 nicotinic acetylcholine receptor (nAChR), a gene product of the CHRNA7, in cortical and subcortical areas. Results revealed similar mRNA distribution pattern in wildtype (WT) and heterozygous (Het) mice, with about half the number of α7 nAChR binding sites in mutants. Hippocampal recordings showed similar input/output responses of field excitatory post-synaptic potentials and theta-burst induced long-term potentiation in WT and Het mice. Het males exhibited impaired spatial learning acquisition in the Barnes Maze. Indicative of increased seizure susceptibility, Het mice developed secondary seizures after 6-Hz corneal stimulation, and had significantly increased sensitivity to the chemoconvulsant pentylenetetrazol resulting in increased spiking in hippocampal EEG recordings. Basal mRNA expression of brain derived neurotrophic factor and activity regulated immediate early genes (c-fos, Arc, Erg-1 and Npas4) during adolescence, a critical period of brain maturation, was unaffected by genotype. Thus, the MD did not show gross neuroanatomical, molecular, and neurophysiological abnormalities despite deficits in spatial learning and increased susceptibility to seizures. Altogether, our results verify the phenotypic profile of the heterozygous Df[h15q13]/+ mouse model and underscore its translational relevance for human 15q13.3 DS. |
| ERG |
| Url: https://pubmed.ncbi.nlm.nih.gov/24121062  Title: The brain through the retina: the flash electroretinogram as a tool to investigate psychiatric disorders.  Investigating the living brain remains one of the major obstacles in psychiatry research in order to better understand the biological underpinning of brain disorders. Novel approaches are needed to study brain functions indirectly. Since it is part of the central nervous system, retinal functions as measured with the flash electroretinogram (ERG) may reflect the central dysfunctions reported in psychiatric disorders. This review describes the flash ERG anomalies reported in patients with psychiatric disorders such as seasonal affective disorder, schizophrenia, autism spectrum disorder and drug addiction and discusses how changes in retinal functions might be used as biomarkers for psychiatric disorder as well as a potential aid to diagnosis in psychiatry. |
| ESR2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28299627  Title: Analysis of estrogen receptor β gene methylation in autistic males in a Chinese Han population.  Autism spectrum disorder (ASD) is a neurodevelopment disorder with abnormalities of social interaction, communication and repetitive behaviors. The higher prevalence of ASD in men implies a potential relationship between sex hormones and ASD etiology. The ESR2 gene encodes estrogen receptor beta (ESR2) and plays an important role during brain development. A relationship between ESR2 and ASD has been suggested by studies on single nucleotide polymorphisms and mRNA and protein expression levels in ASD patients. Here, we explored the possible epigenetic regulation of the ESR2 gene in autism. We collected genomic DNA from the peripheral blood of Chinese Han males with autism and age-matched normal males and measured DNA methylation of CpG islands in the ESR2 gene, which consisted of 41 CpG sites among the proximal promoter region and an untranslated exon, by bisulfite sequencing. We also investigated a relationship between DNA methylation and phenotypic features of autism, as assessed by the Children Autism Rating Scale. We found little overall difference in the DNA methylation of the ESR2 5'-flanking region in individuals with autism compared with normal individuals. However, detailed analyses revealed that eight specific CpG sites were hypermethylated in autistic individuals and that four specific CpG sites were positively associated with the severity of autistic symptoms. Our study indicates that the epigenetic dysregulation of ESR2 may govern the development of autism. |
| EZH2, MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/33016573  Title: Critical regulation of a NDIME/MEF2C axis in embryonic stem cell neural differentiation and autism.  A microdeletion within human chromosome 5q14.3 has been associated with the occurrence of neurodevelopmental disorders, such as autism and intellectual disability, and MEF2C haploinsufficiency was identified as main cause. Here, we report that a brain-enriched long non-coding RNA, NDIME, is located near the MEF2C locus and is required for normal neural differentiation of mouse embryonic stem cells (mESCs). NDIME interacts with EZH2, the major component of polycomb repressive complex 2 (PRC2), and blocks EZH2-mediated trimethylation of histone H3 lysine 27 (H3K27me3) at the Mef2c promoter, promoting MEF2C transcription. Moreover, the expression levels of both NDIME and MEF2C were strongly downregulated in the hippocampus of a mouse model of autism, and the adeno-associated virus (AAV)-mediated expression of NDIME in the hippocampus of these mice significantly increased MEF2C expression and ameliorated autism-like behaviors. The results of this study reveal an epigenetic mechanism by which NDIME regulates MEF2C transcription and neural differentiation and suggest potential effects and therapeutic approaches of the NDIME/MEF2C axis in autism. |
| EZH2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28720872  Title: Associating transcription factors and conserved RNA structures with gene regulation in the human brain.  Anatomical subdivisions of the human brain can be associated with different neuronal functions. This functional diversification is reflected by differences in gene expression. By analyzing post-mortem gene expression data from the Allen Brain Atlas, we investigated the impact of transcription factors (TF) and RNA secondary structures on the regulation of gene expression in the human brain. First, we modeled the expression of a gene as a linear combination of the expression of TFs. We devised an approach to select robust TF-gene interactions and to determine localized contributions to gene expression of TFs. Among the TFs with the most localized contributions, we identified EZH2 in the cerebellum, NR3C1 in the cerebral cortex and SRF in the basal forebrain. Our results suggest that EZH2 is involved in regulating ZIC2 and SHANK1 which have been linked to neurological diseases such as autism spectrum disorder. Second, we associated enriched regulatory elements inside differentially expressed mRNAs with RNA secondary structure motifs. We found a group of purine-uracil repeat RNA secondary structure motifs plus other motifs in neuron related genes such as ACSL4 and ERLIN2. |
| FAN1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27459725  Title: 15q13.3 homozygous knockout mouse model display epilepsy-, autism- and schizophrenia-related phenotypes.  The 15q13.3 microdeletion syndrome is caused by a 1.5-MB hemizygous microdeletion located on 15q13.3 affecting seven genes: FAN1; MTMR10; TRPM1; miR-211; KLF13; OTUD7A; and CHRNA7. The 15q13.3 microdeletion increases the risk of intellectual disability, epilepsy, autism spectrum disorder and schizophrenia, though the clinical profile varies considerably. Two mouse models of this syndrome, with hemizygous deletion of the orthologous region in the murine genome, have recently been shown to recapitulate a number of the behavioral and physiological deficits that characterize the human condition. Still, little is known of the underlying biological mechanisms. Eleven human cases with homozygous deletion of the 15q13.3 region have been reported, all with severe functional and physiological impairments. We therefore hypothesized that a 15q13.3 homozygous knockout would confer more pronounced behavioral and physiological deficits in mice than the 15q13.3 hemizygous deletion. Here we report the characterization of a 15q13.3 knockout mouse. We observed marked deficits including altered seizure susceptibility, autistic behavior-related phenotypes, and auditory sensory processing. Several of these deficits, albeit less pronounced, were also found in the 15q13.3 hemizygous littermates indicating a gene-dosage dependency. Our findings strongly indicate that studies of the hemi- and homozygous 15q13.3 mouse strains will facilitate understanding of the biological mechanisms of severe mental disorders. |
| FEZF2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35273083  Title: Formation of the Mouse Internal Capsule and Cerebral Peduncle: A Pioneering Role for Striatonigral Axons as Revealed in Isl1 Conditional Mutants.  The projection neurons of the striatum, the principal nucleus of the basal ganglia, belong to one of the following two major pathways: the striatopallidal (indirect) pathway or the striatonigral (direct) pathway. Striatonigral axons project long distances and encounter ascending tracts (thalamocortical) while coursing alongside descending tracts (corticofugal) as they extend through the internal capsule and cerebral peduncle. These observations suggest that striatal circuitry may help to guide their trajectories. To investigate the developmental contributions of striatonigral axons to internal capsule formation, we have made use of Sox8-EGFP (striatal direct pathway) and Fezf2-TdTomato (corticofugal pathway) BAC transgenic reporter mice in combination with immunohistochemical markers to trace these axonal pathways throughout development. We show that striatonigral axons pioneer the internal capsule and cerebral peduncle and are temporally and spatially well positioned to provide guidance for corticofugal and thalamocortical axons. Using Isl1 conditional knock-out (cKO) mice, which exhibit disrupted striatonigral axon outgrowth, we observe both corticofugal and thalamocortical axon defects with either ventral forebrain- or telencephalon-specific Isl1 inactivation, despite Isl1 not being expressed in either cortical or thalamic projection neurons. Striatonigral axon defects can thus disrupt internal capsule formation. Our genome-wide transcriptomic analysis in Isl1 cKOs reveals changes in gene expression relevant to cell adhesion, growth cone dynamics, and extracellular matrix composition, suggesting potential mechanisms by which the striatonigral pathway exerts this guidance role. Together, our data support a novel pioneering role for the striatal direct pathway in the correct assembly of the ascending and descending axon tracts within the internal capsule and cerebral peduncle.SIGNIFICANCE STATEMENT The basal ganglia are a group of subcortical nuclei with established roles in the coordination of voluntary motor programs, aspects of cognition, and the selection of appropriate social behaviors. Hence, disruptions in basal ganglia connectivity have been implicated in the motor, cognitive, and social dysfunction characterizing common neurodevelopmental disorders such as attention-deficit/hyperactivity disorder, autism spectrum disorder, obsessive-compulsive disorder, and tic disorder. Here, we identified a novel role for the striatonigral (direct) pathway in pioneering the internal capsule and cerebral peduncle, and in guiding axons extending to and from the cortex. Our findings suggest that the abnormal development of basal ganglia circuits can drive secondary internal capsule defects and thereby may contribute to the pathology of these disorders. |
| FEZF2, SATB2, TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24155294  Title: Stereotypical alterations in cortical patterning are associated with maternal illness-induced placental dysfunction.  We have previously shown in mice that cytokine-mediated damage to the placenta can temporarily limit the flow of nutrients and oxygen to the fetus. The placental vulnerability is pronounced before embryonic day 11, when even mild immune challenge results in fetal loss. As gestation progresses, the placenta becomes increasingly resilient to maternal inflammation, but there is a narrow window in gestation when the placenta is still vulnerable to immune challenge yet resistant enough to allow for fetal survival. This gestational window correlates with early cortical neurogenesis in the fetal brain. Here, we show that maternal illness during this period selectively alters the abundance and laminar positioning of neuronal subtypes influenced by the Tbr1, Satb2, and Ctip2/Fezf2 patterning axis. The disturbances also lead to a laminar imbalance in the proportions of projection neurons and interneurons in the adult and are sufficient to cause changes in social behavior and cognition. These data illustrate how the timing of an illness-related placental vulnerability causes developmental alterations in neuroanatomical systems and behaviors that are relevant to autism spectrum disorders. |
| FOXG1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36419462  Title: Thyroid hormone elicits intergenerational epigenetic effects on adult social behavior and fetal brain expression of autism susceptibility genes.  Genetic mutations identified in genome-wide association studies can only explain a small percentage of the cases of complex, highly heritable human conditions, including neurological and neurodevelopmental disorders. This suggests that intergenerational epigenetic effects, possibly triggered by environmental circumstances, may contribute to their etiology. We previously described altered DNA methylation signatures in the sperm of mice that experienced developmental overexposure to thyroid hormones as a result of a genetic defect in hormone clearance (DIO3 deficiency). Here we studied fetal brain gene expression and adult social behavior in genetically normal F2 generation descendants of overexposed mice. The brain of F2 generation E13.5 fetuses exhibited abnormal expression of genes associated with autism in humans, including Auts2, Disc1, Ldlr, Per2, Shank3, Oxtr, Igf1, Foxg1, Cd38, Grid2, Nrxn3, and Reln. These abnormal gene expression profiles differed depending on the sex of the exposed ancestor. In the three-chamber social box test, adult F2 generation males manifested significantly decreased interest in social interaction and social novelty, as revealed by decrease total time, distance traveled and time immobile in the area of interaction with novel strangers. F1 generation mice, compared to appropriate controls also exhibited altered profiles in fetal brain gene expression, although these profiles were substantially different to those in the F2 generation. Likewise adult F1 generation mice showed some abnormalities in social behavior that were sexually dimorphic and milder than those in F2 generation mice. Our results indicate that developmental overexposure to thyroid hormone causes intergenerational epigenetic effects impacting social behavior and the expression of autism-related genes during early brain development. Our results open the possibility that altered thyroid hormone states, by eliciting changes in the epigenetic information of the germ line, contribute to the susceptibility and the missing-but heriTables-etiology of complex neurodevelopmental conditions characterized by social deficits, including autism and schizophrenia. |
| FOXG1, SATB2, TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34184026  Title: scRNA sequencing uncovers a TCF4-dependent transcription factor network regulating commissure development in mouse.  Transcription factor 4 (TCF4) is a crucial regulator of neurodevelopment and has been linked to the pathogenesis of autism, intellectual disability and schizophrenia. As a class I bHLH transcription factor (TF), it is assumed that TCF4 exerts its neurodevelopmental functions through dimerization with proneural class II bHLH TFs. Here, we aim to identify TF partners of TCF4 in the control of interhemispheric connectivity formation. Using a new bioinformatic strategy integrating TF expression levels and regulon activities from single cell RNA-sequencing data, we find evidence that TCF4 interacts with non-bHLH TFs and modulates their transcriptional activity in Satb2+ intercortical projection neurons. Notably, this network comprises regulators linked to the pathogenesis of neurodevelopmental disorders, e.g. FOXG1, SOX11 and BRG1. In support of the functional interaction of TCF4 with non-bHLH TFs, we find that TCF4 and SOX11 biochemically interact and cooperatively control commissure formation in vivo, and regulate the transcription of genes implicated in this process. In addition to identifying new candidate interactors of TCF4 in neurodevelopment, this study illustrates how scRNA-Seq data can be leveraged to predict TF networks in neurodevelopmental processes. |
| FOXG1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34145239  Title: FoxG1 regulates the formation of cortical GABAergic circuit during an early postnatal critical period resulting in autism spectrum disorder-like phenotypes.  Abnormalities in GABAergic inhibitory circuits have been implicated in the aetiology of autism spectrum disorder (ASD). ASD is caused by genetic and environmental factors. Several genes have been associated with syndromic forms of ASD, including FOXG1. However, when and how dysregulation of FOXG1 can result in defects in inhibitory circuit development and ASD-like social impairments is unclear. Here, we show that increased or decreased FoxG1 expression in both excitatory and inhibitory neurons results in ASD-related circuit and social behavior deficits in our mouse models. We observe that the second postnatal week is the critical period when regulation of FoxG1 expression is required to prevent subsequent ASD-like social impairments. Transplantation of GABAergic precursor cells prior to this critical period and reduction in GABAergic tone via Gad2 mutation ameliorates and exacerbates circuit functionality and social behavioral defects, respectively. Our results provide mechanistic insight into the developmental timing of inhibitory circuit formation underlying ASD-like phenotypes in mouse models. |
| FOXG1, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33429932  Title: Role of DNA Methyl-CpG-Binding Protein MeCP2 in Rett Syndrome Pathobiology and Mechanism of Disease.  Rett Syndrome (RTT) is a severe, rare, and progressive developmental disorder with patients displaying neurological regression and autism spectrum features. The affected individuals are primarily young females, and more than 95% of patients carry de novo mutation(s) in the Methyl-CpG-Binding Protein 2 (MECP2) gene. While the majority of RTT patients have MECP2 mutations (classical RTT), a small fraction of the patients (atypical RTT) may carry genetic mutations in other genes such as the cyclin-dependent kinase-like 5 (CDKL5) and FOXG1. Due to the neurological basis of RTT symptoms, MeCP2 function was originally studied in nerve cells (neurons). However, later research highlighted its importance in other cell types of the brain including glia. In this regard, scientists benefitted from modeling the disease using many different cellular systems and transgenic mice with loss- or gain-of-function mutations. Additionally, limited research in human postmortem brain tissues provided invaluable findings in RTT pathobiology and disease mechanism. MeCP2 expression in the brain is tightly regulated, and its altered expression leads to abnormal brain function, implicating MeCP2 in some cases of autism spectrum disorders. In certain disease conditions, MeCP2 homeostasis control is impaired, the regulation of which in rodents involves a regulatory microRNA (miR132) and brain-derived neurotrophic factor (BDNF). Here, we will provide an overview of recent advances in understanding the underlying mechanism of disease in RTT and the associated genetic mutations in the MECP2 gene along with the pathobiology of the disease, the role of the two most studied protein variants (MeCP2E1 and MeCP2E2 isoforms), and the regulatory mechanisms that control MeCP2 homeostasis network in the brain, including BDNF and miR132. |
| FOXG1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32158381  Title: Transcription and Beyond: Delineating FOXG1 Function in Cortical Development and Disorders.  Forkhead Box G1 (FOXG1) is a member of the Forkhead family of genes with non-redundant roles in brain development, where alteration of this gene's expression significantly affects the formation and function of the mammalian cerebral cortex. FOXG1 haploinsufficiency in humans is associated with prominent differences in brain size and impaired intellectual development noticeable in early childhood, while homozygous mutations are typically fatal. As such, FOXG1 has been implicated in a wide spectrum of congenital brain disorders, including the congenital variant of Rett syndrome, infantile spasms, microcephaly, autism spectrum disorder (ASD) and schizophrenia. Recent technological advances have yielded greater insight into phenotypic variations observed in FOXG1 syndrome, molecular mechanisms underlying pathogenesis of the disease, and multifaceted roles of FOXG1 expression. In this review, we explore the emerging mechanisms of FOXG1 in a range of transcriptional to posttranscriptional events in order to evolve our current view of how a single transcription factor governs the assembly of an elaborate cortical circuit responsible for higher cognitive functions and neurological disorders. |
| FOXG1, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32105570  Title: Rett Syndrome, a Neurodevelopmental Disorder, Whole-Transcriptome, and Mitochondrial Genome Multiomics Analyses Identify Novel Variations and Disease Pathways.  Rett syndrome (RTT) is a severe neurodevelopmental disorder reported worldwide in diverse populations. RTT is diagnosed primarily in females, with clinical findings manifesting early in life. Despite the variable rates across populations, RTT has an estimated prevalence of ∼1 in 10,000 live female births. Among 215 Saudi Arabian patients with neurodevelopmental and autism spectrum disorders, we identified 33 patients with RTT who were subsequently examined by genome-wide transcriptome and mitochondrial genome variations. To the best of our knowledge, this is the first in-depth molecular and multiomics analyses of a large cohort of Saudi RTT cases with a view to informing the underlying mechanisms of this disease that impact many patients and families worldwide. The patients were unrelated, except for 2 affected sisters, and comprised of 25 classic and eight atypical RTT cases. The cases were screened for methyl-CpG binding protein 2 (MECP2), CDKL5, FOXG1, NTNG1, and mitochondrial DNA (mtDNA) variants, as well as copy number variations (CNVs) using a genome-wide experimental strategy. We found that 15 patients (13 classic and two atypical RTT) have MECP2 mutations, 2 of which were novel variants. Two patients had novel FOXG1 and CDKL5 variants (both atypical RTT). Whole mtDNA sequencing of the patients who were MECP2 negative revealed two novel mtDNA variants in two classic RTT patients. Importantly, the whole-transcriptome analysis of our RTT patients' blood and further comparison with previous expression profiling of brain tissue from patients with RTT revealed 77 significantly dysregulated genes. The gene ontology and interaction network analysis indicated potentially critical roles of MAPK9, NDUFA5, ATR, SMARCA5, RPL23, SRSF3, and mitochondrial dysfunction, oxidative stress response and MAPK signaling pathways in the pathogenesis of RTT genes. This study expands our knowledge on RTT disease networks and pathways as well as presents novel mutations and mtDNA alterations in RTT in a population sample that was not previously studied. |
| FOXG1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27995975  Title: RNA activation of haploinsufficient Foxg1 gene in murine neocortex.  More than one hundred distinct gene hemizygosities are specifically linked to epilepsy, mental retardation, autism, schizophrenia and neuro-degeneration. Radical repair of these gene deficits via genome engineering is hardly feasible. The same applies to therapeutic stimulation of the spared allele by artificial transactivators. Small activating RNAs (saRNAs) offer an alternative, appealing approach. As a proof-of-principle, here we tested this approach on the Rett syndrome-linked, haploinsufficient, Foxg1 brain patterning gene. We selected a set of artificial small activating RNAs (saRNAs) upregulating it in neocortical precursors and their derivatives. Expression of these effectors achieved a robust biological outcome. saRNA-driven activation (RNAa) was limited to neural cells which normally express Foxg1 and did not hide endogenous gene tuning. saRNAs recognized target chromatin through a ncRNA stemming from it. Gene upregulation required Ago1 and was associated to RNApolII enrichment throughout the Foxg1 locus. Finally, saRNA delivery to murine neonatal brain replicated Foxg1-RNAa in vivo. |
| FOXG1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27760116  Title: Chromosome conformation elucidates regulatory relationships in developing human brain.  Three-dimensional physical interactions within chromosomes dynamically regulate gene expression in a tissue-specific manner. However, the 3D organization of chromosomes during human brain development and its role in regulating gene networks dysregulated in neurodevelopmental disorders, such as autism or schizophrenia, are unknown. Here we generate high-resolution 3D maps of chromatin contacts during human corticogenesis, permitting large-scale annotation of previously uncharacterized regulatory relationships relevant to the evolution of human cognition and disease. Our analyses identify hundreds of genes that physically interact with enhancers gained on the human lineage, many of which are under purifying selection and associated with human cognitive function. We integrate chromatin contacts with non-coding variants identified in schizophrenia genome-wide association studies (GWAS), highlighting multiple candidate schizophrenia risk genes and pathways, including transcription factors involved in neurogenesis, and cholinergic signalling molecules, several of which are supported by independent expression quantitative trait loci and gene expression analyses. Genome editing in human neural progenitors suggests that one of these distal schizophrenia GWAS loci regulates FOXG1 expression, supporting its potential role as a schizophrenia risk gene. This work provides a framework for understanding the effect of non-coding regulatory elements on human brain development and the evolution of cognition, and highlights novel mechanisms underlying neuropsychiatric disorders. |
| FOXG1, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27001178  Title: Visual impairment in FOXG1-mutated individuals and mice.  The Forkead Box G1 (FOXG1 in humans, Foxg1 in mice) gene encodes for a DNA-binding transcription factor, essential for the development of the telencephalon in mammalian forebrain. Mutations in FOXG1 have been reported to be involved in the onset of Rett Syndrome, for which sequence alterations of MECP2 and CDKL5 are known. While visual alterations are not classical hallmarks of Rett syndrome, an increasing body of evidence shows visual impairment in patients and in MeCP2 and CDKL5 animal models. Herein we focused on the functional role of FOXG1 in the visual system of animal models (Foxg1(+/Cre) mice) and of a cohort of subjects carrying FOXG1 mutations or deletions. Visual physiology of Foxg1(+/Cre) mice was assessed by visually evoked potentials, which revealed a significant reduction in response amplitude and visual acuity with respect to wild-type littermates. Morphological investigation showed abnormalities in the organization of excitatory/inhibitory circuits in the visual cortex. No alterations were observed in retinal structure. By examining a cohort of FOXG1-mutated individuals with a panel of neuro-ophthalmological assessments, we found that all of them exhibited visual alterations compatible with high-level visual dysfunctions. In conclusion our data show that Foxg1 haploinsufficiency results in an impairment of mouse and human visual cortical function. |
| FOXG1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26186191  Title: FOXG1-Dependent Dysregulation of GABA/Glutamate Neuron Differentiation in Autism Spectrum Disorders.  Autism spectrum disorder (ASD) is a disorder of brain development. Most cases lack a clear etiology or genetic basis, and the difficulty of re-enacting human brain development has precluded understanding of ASD pathophysiology. Here we use three-dimensional neural cultures (organoids) derived from induced pluripotent stem cells (iPSCs) to investigate neurodevelopmental alterations in individuals with severe idiopathic ASD. While no known underlying genomic mutation could be identified, transcriptome and gene network analyses revealed upregulation of genes involved in cell proliferation, neuronal differentiation, and synaptic assembly. ASD-derived organoids exhibit an accelerated cell cycle and overproduction of GABAergic inhibitory neurons. Using RNA interference, we show that overexpression of the transcription factor FOXG1 is responsible for the overproduction of GABAergic neurons. Altered expression of gene network modules and FOXG1 are positively correlated with symptom severity. Our data suggest that a shift toward GABAergic neuron fate caused by FOXG1 is a developmental precursor of ASD. |
| FOXG1, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21441262  Title: The core FOXG1 syndrome phenotype consists of postnatal microcephaly, severe mental retardation, absent language, dyskinesia, and corpus callosum hypogenesis.  Submicroscopic deletions in 14q12 spanning FOXG1 or intragenic mutations have been reported in patients with a developmental disorder described as a congenital variant of Rett syndrome. This study aimed to further characterise and delineate the phenotype of FOXG1 mutation positive patients. The study mapped the breakpoints of a 2;14 translocation by fluorescence in situ hybridisation and analysed three chromosome rearrangements in 14q12 by cytogenetic analysis and/or array comparative genomic hybridisation. The FOXG1 gene was sequenced in 210 patients, including 129 patients with unexplained developmental disorders and 81 MECP2 mutation negative individuals. One known mutation, seen in two patients, and nine novel mutations of FOXG1 including two deletions, two chromosome rearrangements disrupting or displacing putative cis-regulatory elements from FOXG1, and seven sequence changes, are reported. Analysis of 11 patients in this study, and a further 15 patients reported in the literature, demonstrates a complex constellation of features including mild postnatal growth deficiency, severe postnatal microcephaly, severe mental retardation with absent language development, deficient social reciprocity resembling autism, combined stereotypies and frank dyskinesias, epilepsy, poor sleep patterns, irritability in infancy, unexplained episodes of crying, recurrent aspiration, and gastro-oesophageal reflux. Brain imaging studies reveal simplified gyral pattern and reduced white matter volume in the frontal lobes, corpus callosum hypogenesis, and variable mild frontal pachgyria. These findings have significantly expanded the number of FOXG1 mutations and identified two affecting possible cis-regulatory elements. While the phenotype of the patients overlaps both classic and congenital Rett syndrome, extensive clinical evaluation demonstrates a distinctive and clinically recognisable phenotype which the authors suggest designating as the FOXG1 syndrome. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36672612  Title: L-Serine Influences Epigenetic Modifications to Improve Cognition and Behaviors in Growth Hormone-Releasing Hormone Knockout Mice.  Neurodegenerative diseases feature changes in cognition, and anxiety-like and autism-like behaviors, which are associated with epigenetic alterations such as DNA methylation and histone modifications. The amino acid L-serine has been shown to have beneficial effects on neurological symptoms. Here, we found that growth hormone-releasing hormone knockout (GHRH-KO) mice, a GH-deficiency mouse model characterized by extended lifespan and enhanced insulin sensitivity, showed a lower anxiety symptom and impairment of short-term object recognition memory and autism-like behaviors. Interestingly, L-serine administration exerted anxiolytic effects in mice and ameliorated the behavioral deficits in GHRH-KO. L-serine treatment upregulated histone epigenetic markers of H3K4me, H3K9ac, H3K14ac and H3K18ac in the hippocampus and H3K4me in the cerebral cortex in both GHRH-KO mice and wild type controls. L-serine-modulated epigenetic marker changes, in turn, were found to regulate mRNA expression of BDNF, grm3, foxp1, shank3, auts2 and marcksl1, which are involved in anxiety-, cognitive- and autism-like behaviors. Our study provides a novel insight into the beneficial effects of L-serine intervention on neuropsychological impairments. |
| FOXP1, GTF2I |
| Url: https://pubmed.ncbi.nlm.nih.gov/36292679  Title: Profiling Genome-Wide DNA Methylation in Children with Autism Spectrum Disorder and in Children with Fragile X Syndrome.  Autism spectrum disorder (ASD) is an early onset, developmental disorder whose genetic cause is heterogeneous and complex. In total, 70% of ASD cases are due to an unknown etiology. Among the monogenic causes of ASD, fragile X syndrome (FXS) accounts for 2-4% of ASD cases, and 60% of individuals with FXS present with ASD. Epigenetic changes, specifically DNA methylation, which modulates gene expression levels, play a significant role in the pathogenesis of both disorders. Thus, in this study, using the Human Methylation EPIC Bead Chip, we examined the global DNA methylation profiles of biological samples derived from 57 age-matched male participants (2-6 years old), including 23 subjects with ASD, 23 subjects with FXS with ASD (FXSA) and 11 typical developing (TD) children. After controlling for technical variation and white blood cell composition, using the conservatory threshold of the false discovery rate (FDR ≤ 0.05), in the three comparison groups, TD vs. AD, TD vs. FXSA and ASD vs. FXSA, we identified 156, 79 and 3100 differentially methylated sites (DMS), and 14, 13 and 263 differential methylation regions (DMRs). Interestingly, several genes differentially methylated among the three groups were among those listed in the SFARI Gene database, including the PAK2, GTF2I and FOXP1 genes important for brain development. Further, enrichment analyses identified pathways involved in several functions, including synaptic plasticity. Our preliminary study identified a significant role of altered DNA methylation in the pathology of ASD and FXS, suggesting that the characterization of a DNA methylation signature may help to unravel the pathogenicity of FXS and ASD and may help the development of an improved diagnostic classification of children with ASD and FXSA. In addition, it may pave the way for developing therapeutic interventions that could reverse the altered methylome profile in children with neurodevelopmental disorders. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35165191  Title: Mitochondrial dysfunction and oxidative stress contribute to cognitive and motor impairment in FOXP1 syndrome.  FOXP1 syndrome caused by haploinsufficiency of the forkhead box protein P1 (FOXP1) gene is a neurodevelopmental disorder that manifests motor dysfunction, intellectual disability, autism, and language impairment. In this study, we used a Foxp1+/- mouse model to address whether cognitive and motor deficits in FOXP1 syndrome are associated with mitochondrial dysfunction and oxidative stress. Here, we show that genes with a role in mitochondrial biogenesis and dynamics (e.g., Foxo1, Pgc-1α, Tfam, Opa1, and Drp1) were dysregulated in the striatum of Foxp1+/- mice at different postnatal stages. Furthermore, these animals exhibit a reduced mitochondrial membrane potential and complex I activity, as well as decreased expression of the antioxidants superoxide dismutase 2 (Sod2) and glutathione (GSH), resulting in increased oxidative stress and lipid peroxidation. These features can explain the reduced neurite branching, learning and memory, endurance, and motor coordination that we observed in these animals. Taken together, we provide strong evidence of mitochondrial dysfunction in Foxp1+/- mice, suggesting that insufficient energy supply and excessive oxidative stress underlie the cognitive and motor impairment in FOXP1 deficiency. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35052467  Title: Disrupted Mitochondrial Network Drives Deficits of Learning and Memory in a Mouse Model of FOXP1 Haploinsufficiency.  Reduced cognitive flexibility, characterized by restricted interests and repetitive behavior, is associated with atypical memory performance in autism spectrum disorder (ASD), suggesting hippocampal dysfunction. FOXP1 syndrome is a neurodevelopmental disorder characterized by ASD, language deficits, global developmental delay, and mild to moderate intellectual disability. Strongly reduced Foxp1 expression has been detected in the hippocampus of Foxp1+/- mice, a brain region required for learning and memory. To investigate learning and memory performance in these animals, fear conditioning tests were carried out, which showed impaired associative learning compared with wild type (WT) animals. To shed light on the underlying mechanism, we analyzed various components of the mitochondrial network in the hippocampus. Several proteins regulating mitochondrial biogenesis (e.g., Foxo1, Pgc-1α, Tfam) and dynamics (Mfn1, Opa1, Drp1 and Fis1) were significantly dysregulated, which may explain the increased mitophagy observed in the Foxp1+/- hippocampus. The reduced activity of complex I and decreased expression of Sod2 most likely increase the production of reactive oxygen species and the expression of the pre-apoptotic proteins Bcl-2 and Bax in this tissue. In conclusion, we provide evidence that a disrupted mitochondrial network and the resulting oxidative stress in the hippocampus contribute to the altered learning and cognitive impairment in Foxp1+/- mice, suggesting that similar alterations also play a major role in patients with FOXP1 syndrome. |
| FOXP1, HIVEP2, WAC |
| Url: https://pubmed.ncbi.nlm.nih.gov/35052433  Title: Exploring the Contribution to ADHD of Genes Involved in Mendelian Disorders Presenting with Hyperactivity and/or Inattention.  Attention-deficit hyperactivity disorder (ADHD) is a complex neurodevelopmental disorder characterized by hyperactivity, impulsivity, and/or inattention, which are symptoms also observed in many rare genetic disorders. We searched for genes involved in Mendelian disorders presenting with ADHD symptoms in the Online Mendelian Inheritance in Man (OMIM) database, to curate a list of new candidate risk genes for ADHD. We explored the enrichment of functions and pathways in this gene list, and tested whether rare or common variants in these genes are associated with ADHD or with its comorbidities. We identified 139 genes, causal for 137 rare disorders, mainly related to neurodevelopmental and brain function. Most of these Mendelian disorders also present with other psychiatric traits that are often comorbid with ADHD. Using whole exome sequencing (WES) data from 668 ADHD cases, we found rare variants associated with the dimension of the severity of inattention symptoms in three genes: KIF11, WAC, and CRBN. Then, we focused on common variants and identified six genes associated with ADHD (in 19,099 cases and 34,194 controls): MANBA, UQCC2, HIVEP2, FOPX1, KANSL1, and AUH. Furthermore, HIVEP2, FOXP1, and KANSL1 were nominally associated with autism spectrum disorder (ASD) (18,382 cases and 27,969 controls), as well as HIVEP2 with anxiety (7016 cases and 14,475 controls), and FOXP1 with aggression (18,988 individuals), which is in line with the symptomatology of the rare disorders they are responsible for. In conclusion, inspecting Mendelian disorders and the genes responsible for them constitutes a valuable approach for identifying new risk genes and the mechanisms of complex disorders. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34657631  Title: Combinatorial patterns of gene expression changes contribute to variable expressivity of the developmental delay-associated 16p12.1 deletion.  Recent studies have suggested that individual variants do not sufficiently explain the variable expressivity of phenotypes observed in complex disorders. For example, the 16p12.1 deletion is associated with developmental delay and neuropsychiatric features in affected individuals, but is inherited in > 90% of cases from a mildly-affected parent. While children with the deletion are more likely to carry additional "second-hit" variants than their parents, the mechanisms for how these variants contribute to phenotypic variability are unknown. We performed detailed clinical assessments, whole-genome sequencing, and RNA sequencing of lymphoblastoid cell lines for 32 individuals in five large families with multiple members carrying the 16p12.1 deletion. We identified contributions of the 16p12.1 deletion and "second-hit" variants towards a range of expression changes in deletion carriers and their family members, including differential expression, outlier expression, alternative splicing, allele-specific expression, and expression quantitative trait loci analyses. We found that the deletion dysregulates multiple autism and brain development genes such as FOXP1, ANK3, and MEF2. Carrier children also showed an average of 5323 gene expression changes compared with one or both parents, which matched with 33/39 observed developmental phenotypes. We identified significant enrichments for 13/25 classes of "second-hit" variants in genes with expression changes, where 4/25 variant classes were only enriched when inherited from the noncarrier parent, including loss-of-function SNVs and large duplications. In 11 instances, including for ZEB2 and SYNJ1, gene expression was synergistically altered by both the deletion and inherited "second-hits" in carrier children. Finally, brain-specific interaction network analysis showed strong connectivity between genes carrying "second-hits" and genes with transcriptome alterations in deletion carriers. Our results suggest a potential mechanism for how "second-hit" variants modulate expressivity of complex disorders such as the 16p12.1 deletion through transcriptomic perturbation of gene networks important for early development. Our work further shows that family-based assessments of transcriptome data are highly relevant towards understanding the genetic mechanisms associated with complex disorders. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33892622  Title: FOXP1 syndrome: a review of the literature and practice parameters for medical assessment and monitoring.  FOXP1 syndrome is a neurodevelopmental disorder caused by mutations or deletions that disrupt the forkhead box protein 1 (FOXP1) gene, which encodes a transcription factor important for the early development of many organ systems, including the brain. Numerous clinical studies have elucidated the role of FOXP1 in neurodevelopment and have characterized a phenotype. FOXP1 syndrome is associated with intellectual disability, language deficits, autism spectrum disorder, hypotonia, and congenital anomalies, including mild dysmorphic features, and brain, cardiac, and urogenital abnormalities. Here, we present a review of human studies summarizing the clinical features of individuals with FOXP1 syndrome and enlist a multidisciplinary group of clinicians (pediatrics, genetics, psychiatry, neurology, cardiology, endocrinology, nephrology, and psychology) to provide recommendations for the assessment of FOXP1 syndrome. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33065217  Title: A genome-wide association study identifies a gene network associated with paranoid schizophrenia and antipsychotics-induced tardive dyskinesia.  In the present study we conducted a genome-wide association study (GWAS) in a cohort of 505 patients with paranoid schizophrenia (SCZ), of which 95 had tardive dyskinesia (TD), and 503 healthy controls. Using data generated by the PsychENCODE Consortium (PEC) and other bioinformatic databases, we revealed a gene network, implicated in neurodevelopment and brain function, associated with both these disorders. Almost all these genes are in gene or isoform co-expression PEC network modules important for the functioning of the brain; the activity of these networks is also altered in SCZ, bipolar disorder and autism spectrum disorders. The associated PEC network modules are enriched for gene ontology terms relevant to the brain development and function (CNS development, neuron development, axon ensheathment, synapse, synaptic vesicle cycle, and signaling receptor activity) and to the immune system (inflammatory response). Results of the present study suggest that orofacial and limbtruncal types of TD seem to share the molecular network with SCZ. Paranoid SCZ and abnormal involuntary movements that indicate the orofacial type of TD are associated with the same genomic loci on chromosomes 3p22.2, 8q21.13, and 13q14.2. The limbtruncal type of TD is associated with a locus on chromosome 3p13 where the best functional candidate is FOXP1, a high-confidence SCZ gene. The results of this study shed light on common pathogenic mechanisms for SCZ and TD, and indicate that the pathogenesis of the orofacial and limbtruncal types of TD might be driven by interacting genes implicated in neurodevelopment. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32130906  Title: Single-Cell Analysis of Foxp1-Driven Mechanisms Essential for Striatal Development.  The striatum is a critical forebrain structure integrating cognitive, sensory, and motor information from diverse brain regions into meaningful behavioral output. However, the transcriptional mechanisms underlying striatal development at single-cell resolution remain unknown. Using single-cell RNA sequencing (RNA-seq), we examine the cellular diversity of the early postnatal striatum and show that Foxp1, a transcription factor strongly linked to autism and intellectual disability, regulates the cellular composition, neurochemical architecture, and connectivity of the striatum in a cell-type-dependent fashion. We also identify Foxp1-regulated target genes within distinct cell types and connect these molecular changes to functional and behavioral deficits relevant to phenotypes described in patients with FOXP1 loss-of-function mutations. Using this approach, we could also examine the non-cell-autonomous effects produced by disrupting one cell type and the molecular compensation that occurs in other populations. These data reveal the cell-type-specific transcriptional mechanisms regulated by Foxp1 that underlie distinct features of striatal circuitry. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32049024  Title: Foxp1 Regulates Neural Stem Cell Self-Renewal and Bias Toward Deep Layer Cortical Fates.  The laminar architecture of the mammalian neocortex depends on the orderly generation of distinct neuronal subtypes by apical radial glia (aRG) during embryogenesis. Here, we identify critical roles for the autism risk gene Foxp1 in maintaining aRG identity and gating the temporal competency for deep-layer neurogenesis. Early in development, aRG express high levels of Foxp1 mRNA and protein, which promote self-renewing cell divisions and deep-layer neuron production. Foxp1 levels subsequently decline during the transition to superficial-layer neurogenesis. Sustained Foxp1 expression impedes this transition, preserving a population of cells with aRG identity throughout development and extending the early neurogenic period into postnatal life. FOXP1 expression is further associated with the initial formation and expansion of basal RG (bRG) during human corticogenesis and can promote the formation of cells exhibiting characteristics of bRG when misexpressed in the mouse cortex. Together, these findings reveal broad functions for Foxp1 in cortical neurogenesis. |
| FOXP1, FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31999079  Title: FOXP transcription factors in vertebrate brain development, function, and disorders.  FOXP transcription factors are an evolutionarily ancient protein subfamily coordinating the development of several organ systems in the vertebrate body. Association of their genes with neurodevelopmental disorders has sparked particular interest in their expression patterns and functions in the brain. Here, FOXP1, FOXP2, and FOXP4 are expressed in distinct cell type-specific spatiotemporal patterns in multiple regions, including the cortex, hippocampus, amygdala, basal ganglia, thalamus, and cerebellum. These varied sites and timepoints of expression have complicated efforts to link FOXP1 and FOXP2 mutations to their respective developmental disorders, the former affecting global neural functions and the latter specifically affecting speech and language. However, the use of animal models, particularly those with brain region- and cell type-specific manipulations, has greatly advanced our understanding of how FOXP expression patterns could underlie disorder-related phenotypes. While many questions remain regarding FOXP expression and function in the brain, studies to date have illuminated the roles of these transcription factors in vertebrate brain development and have greatly informed our understanding of human development and disorders. This article is categorized under: Nervous System Development > Vertebrates: General Principles Gene Expression and Transcriptional Hierarchies > Gene Networks and Genomics Nervous System Development > Vertebrates: Regional Development. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31611379  Title: Gastrointestinal dysfunction in autism displayed by altered motility and achalasia in Foxp1+/- mice.  Gastrointestinal dysfunctions in individuals with autism spectrum disorder are poorly understood, although they are common among this group of patients. FOXP1 haploinsufficiency is characterized by autistic behavior, language impairment, and intellectual disability, but feeding difficulties and gastrointestinal problems have also been reported. Whether these are primary impairments, the result of altered eating behavior, or side effects of psychotropic medication remains unclear. To address this question, we investigated Foxp1+/- mice reflecting FOXP1 haploinsufficiency. These animals show decreased body weight and altered feeding behavior with reduced food and water intake. A pronounced muscular atrophy was detected in the esophagus and colon, caused by reduced muscle cell proliferation. Nitric oxide-induced relaxation of the lower esophageal sphincter was impaired and achalasia was confirmed in vivo by manometry. Foxp1 targets (Nexn, Rbms3, and Wls) identified in the brain were dysregulated in the adult Foxp1+/- esophagus. Total gastrointestinal transit was significantly prolonged due to impaired colonic contractility. Our results have uncovered a previously unknown dysfunction (achalasia and impaired gut motility) that explains the gastrointestinal disturbances in patients with FOXP1 syndrome, with potential wider relevance for autism. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30124790  Title: An Autism-Related, Nonsense Foxp1 Mutant Induces Autophagy and Delays Radial Migration of the Cortical Neurons.  Autism spectrum disorder (ASD) is a complex neurodevelopmental disorder that has a strong genetic component. Disruptions of FOXP1, a transcription factor expressed in the developing cerebral cortex, were associated with ASD. FOXP1(R525X) is a de novo heterozygous mutation found in patients with autism and severe mental retardation. To explore the neuronal basis of FOXP1(R525X) in ASD, we created Foxp1(R521X), a mouse homolog of the human variant. Ectopic expression of Foxp1(R521X) led to cytoplasmic aggregates and activated macroautophagy in neuroblastoma N2a cells and the developing neuronal cells. Cortical neurons expressing Foxp1(R521X) exhibited delayed migration and altered dendritic morphology. As a control, mutant Y435X that was expressed diffusively in the cytoplasm did not induce autophagy and migration delay in the cortex. The embryonic cortical cells had a minimal activity of nonsense-mediated mRNA decay (NMD) as assayed by a splicing-dependent NMD reporter. We hypothesize that the developing neuronal cells use autophagy but not NMD as a safeguard mechanism against nonsense mutant aggregates, resulting in impairment of the cortical development. This study suggests a novel mechanism other than heterozygous loss of FOXP1 for the development of ASD and may advance our understanding of the complex relationships between gene mutation and the related psychiatric disorders. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29141232  Title: FOXP1 Promotes Embryonic Neural Stem Cell Differentiation by Repressing Jagged1 Expression.  Mutations in FOXP1 have been linked to neurodevelopmental disorders including intellectual disability and autism; however, the underlying molecular mechanisms remain ill-defined. Here, we demonstrate with RNA and chromatin immunoprecipitation sequencing that FOXP1 directly regulates genes controlling neurogenesis. We show that FOXP1 is expressed in embryonic neural stem cells (NSCs), and modulation of FOXP1 expression affects both neuron and astrocyte differentiation. Using a murine model of cortical development, FOXP1-knockdown in utero was found to reduce NSC differentiation and migration during corticogenesis. Furthermore, transplantation of FOXP1-knockdown NSCs in neonatal mice after hypoxia-ischemia challenge demonstrated that FOXP1 is also required for neuronal differentiation and functionality in vivo. FOXP1 was found to repress the expression of Notch pathway genes including the Notch-ligand Jagged1, resulting in inhibition of Notch signaling. Finally, blockade of Jagged1 in FOXP1-knockdown NSCs rescued neuronal differentiation in vitro. Together, these data support a role for FOXP1 in regulating embryonic NSC differentiation by modulating Notch signaling. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29138280  Title: Foxp1 regulation of neonatal vocalizations via cortical development.  The molecular mechanisms driving brain development at risk in autism spectrum disorders (ASDs) remain mostly unknown. Previous studies have implicated the transcription factor FOXP1 in both brain development and ASD pathophysiology. However, the specific molecular pathways both upstream of and downstream from FOXP1 are not fully understood. To elucidate the contribution of FOXP1-mediated signaling to brain development and, in particular, neocortical development, we generated forebrain-specific Foxp1 conditional knockout mice. We show that deletion of Foxp1 in the developing forebrain leads to impairments in neonatal vocalizations as well as neocortical cytoarchitectonic alterations via neuronal positioning and migration. Using a genomics approach, we identified the transcriptional networks regulated by Foxp1 in the developing neocortex and found that such networks are enriched for downstream targets involved in neurogenesis and neuronal migration. We also uncovered mechanistic insight into Foxp1 function by demonstrating that sumoylation of Foxp1 during embryonic brain development is necessary for mediating proper interactions between Foxp1 and the NuRD complex. Furthermore, we demonstrated that sumoylation of Foxp1 affects neuronal differentiation and migration in the developing neocortex. Together, these data provide critical mechanistic insights into the function of FOXP1 in the developing neocortex and may reveal molecular pathways at risk in ASD. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29090079  Title: Prospective investigation of FOXP1 syndrome.  Haploinsufficiency of the forkhead-box protein P1 (FOXP1) gene leads to a neurodevelopmental disorder termed FOXP1 syndrome. Previous studies in individuals carrying FOXP1 mutations and deletions have described the presence of autism spectrum disorder (ASD) traits, intellectual disability, language impairment, and psychiatric features. The goal of the present study was to comprehensively characterize the genetic and clinical spectrum of FOXP1 syndrome. This is the first study to prospectively examine the genotype-phenotype relationship in multiple individuals with FOXP1 syndrome, using a battery of standardized clinical assessments. Genetic and clinical data was obtained and analyzed from nine children and adolescents between the ages of 5-17 with mutations in FOXP1. Phenotypic characterization included gold standard ASD testing and norm-referenced measures of cognition, adaptive behavior, language, motor, and visual-motor integration skills. In addition, psychiatric, medical, neurological, and dysmorphology examinations were completed by a multidisciplinary team of clinicians. A comprehensive review of reported cases was also performed. All missense and in-frame mutations were mapped onto the three-dimensional structure of DNA-bound FOXP1. We have identified nine de novo mutations, including three frameshift, one nonsense, one mutation in an essential splice site resulting in frameshift and insertion of a premature stop codon, three missense, and one in-frame deletion. Reviewing prior literature, we found seven instances of recurrent mutations and another 34 private mutations. The majority of pathogenic missense and in-frame mutations, including all four missense mutations in our cohort, lie in the DNA-binding domain. Through structural analyses, we show that the mutations perturb amino acids necessary for binding to the DNA or interfere with the domain swapping that mediates FOXP1 dimerization. Individuals with FOXP1 syndrome presented with delays in early motor and language milestones, language impairment (expressive language > receptive language), ASD symptoms, visual-motor integration deficits, and complex psychiatric presentations characterized by anxiety, obsessive-compulsive traits, attention deficits, and externalizing symptoms. Medical features included non-specific structural brain abnormalities and dysmorphic features, endocrine and gastrointestinal problems, sleep disturbances, and sinopulmonary infections. This study identifies novel FOXP1 mutations associated with FOXP1 syndrome, identifies recurrent mutations, and demonstrates significant clustering of missense mutations in the DNA-binding domain. Clinical findings confirm the role FOXP1 plays in development across multiple domains of functioning. The genetic findings can be incorporated into clinical genetics practice to improve accurate genetic diagnosis of FOXP1 syndrome and the clinical findings can inform monitoring and treatment of individuals with FOXP1 syndrome. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28978667  Title: Foxp1 in Forebrain Pyramidal Neurons Controls Gene Expression Required for Spatial Learning and Synaptic Plasticity.  Genetic perturbations of the transcription factor Forkhead Box P1 (FOXP1) are causative for severe forms of autism spectrum disorder that are often comorbid with intellectual disability. Recent work has begun to reveal an important role for FoxP1 in brain development, but the brain-region-specific contributions of Foxp1 to autism and intellectual disability phenotypes have yet to be determined fully. Here, we describe Foxp1 conditional knock-out (Foxp1cKO) male and female mice with loss of Foxp1 in the pyramidal neurons of the neocortex and the CA1/CA2 subfields of the hippocampus. Foxp1cKO mice exhibit behavioral phenotypes that are of potential relevance to autism spectrum disorder, including hyperactivity, increased anxiety, communication impairments, and decreased sociability. In addition, Foxp1cKO mice have gross deficits in learning and memory tasks of relevance to intellectual disability. Using a genome-wide approach, we identified differentially expressed genes in the hippocampus of Foxp1cKO mice associated with synaptic function and development. Furthermore, using magnetic resonance imaging, we uncovered a significant reduction in the volumes of both the entire hippocampus as well as individual hippocampal subfields of Foxp1cKO mice. Finally, we observed reduced maintenance of LTP in area CA1 of the hippocampus in these mutant mice. Together, these data suggest that proper expression of Foxp1 in the pyramidal neurons of the forebrain is important for regulating gene expression pathways that contribute to specific behaviors reminiscent of those seen in autism and intellectual disability. In particular, Foxp1 regulation of gene expression appears to be crucial for normal hippocampal development, CA1 plasticity, and spatial learning.SIGNIFICANCE STATEMENT Loss-of-function mutations in the transcription factor Forkhead Box P1 (FOXP1) lead to autism spectrum disorder and intellectual disability. Understanding the potential brain-region-specific contributions of FOXP1 to disease-relevant phenotypes could be a critical first step in the management of patients with these mutations. Here, we report that Foxp1 conditional knock-out (Foxp1cKO) mice with loss of Foxp1 in the neocortex and hippocampus display autism and intellectual-disability-relevant behaviors. We also show that these phenotypes correlate with changes in both the genomic and physiological profiles of the hippocampus in Foxp1cKO mice. Our work demonstrates that brain-region-specific FOXP1 expression may relate to distinct, clinically relevant phenotypes. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26494785  Title: FoxP1 orchestration of ASD-relevant signaling pathways in the striatum.  Mutations in the transcription factor Forkhead box p1 (FOXP1) are causative for neurodevelopmental disorders such as autism. However, the function of FOXP1 within the brain remains largely uncharacterized. Here, we identify the gene expression program regulated by FoxP1 in both human neural cells and patient-relevant heterozygous Foxp1 mouse brains. We demonstrate a role for FoxP1 in the transcriptional regulation of autism-related pathways as well as genes involved in neuronal activity. We show that Foxp1 regulates the excitability of striatal medium spiny neurons and that reduction of Foxp1 correlates with defects in ultrasonic vocalizations. Finally, we demonstrate that FoxP1 has an evolutionarily conserved role in regulating pathways involved in striatal neuron identity through gene expression studies in human neural progenitors with altered FOXP1 levels. These data support an integral role for FoxP1 in regulating signaling pathways vulnerable in autism and the specific regulation of striatal pathways important for vocal communication. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25853299  Title: A de novo FOXP1 variant in a patient with autism, intellectual disability and severe speech and language impairment.  FOXP1 (forkhead box protein P1) is a transcription factor involved in the development of several tissues, including the brain. An emerging phenotype of patients with protein-disrupting FOXP1 variants includes global developmental delay, intellectual disability and mild to severe speech/language deficits. We report on a female child with a history of severe hypotonia, autism spectrum disorder and mild intellectual disability with severe speech/language impairment. Clinical exome sequencing identified a heterozygous de novo FOXP1 variant c.1267\_1268delGT (p.V423Hfs\*37). Functional analyses using cellular models show that the variant disrupts multiple aspects of FOXP1 activity, including subcellular localization and transcriptional repression properties. Our findings highlight the importance of performing functional characterization to help uncover the biological significance of variants identified by genomics approaches, thereby providing insight into pathways underlying complex neurodevelopmental disorders. Moreover, our data support the hypothesis that de novo variants represent significant causal factors in severe sporadic disorders and extend the phenotype seen in individuals with FOXP1 haploinsufficiency. |
| FOXP1, FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25407828  Title: Differential FoxP2 and FoxP1 expression in a vocal learning nucleus of the developing budgerigar.  The forkhead domain FOXP2 and FOXP1 transcription factors are implicated in several cognitive disorders with language deficits, notably autism, and thus play a central role in learned vocal motor behavior in humans. Although a similar role for FoxP2 and FoxP1 is proposed for other vertebrate species, including songbirds, the neurodevelopmental expression of these genes are unknown in a species with lifelong vocal learning abilities. Like humans, budgerigars (Melopsittacus undulatus) learn new vocalizations throughout their entire lifetime. Like songbirds, budgerigars have distinct brain nuclei for vocal learning, which include the magnocellular nucleus of the medial striatum (MMSt), a basal ganglia region that is considered developmentally and functionally analogous to Area X in songbirds. Here, we used in situ hybridization and immunohistochemistry to investigate FoxP2 and FoxP1 expression in the MMSt of juvenile and adult budgerigars. We found FoxP2 mRNA and protein expression levels in the MMSt that were lower than the surrounding striatum throughout development and adulthood. In contrast, FoxP1 mRNA and protein had an elevated MMSt/striatum expression ratio as birds matured, regardless of their sex. These results show that life-long vocal plasticity in budgerigars is associated with persistent low-level FoxP2 expression in the budgerigar MMSt, and suggests the possibility that FoxP1 plays an organizational role in the neurodevelopment of vocal motor circuitry. Thus, developmental regulation of the FoxP2 and FoxP1 genes in the basal ganglia appears essential for vocal mimicry in a range of species that possess this relatively rare trait. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25266127  Title: Brain-specific Foxp1 deletion impairs neuronal development and causes autistic-like behaviour.  Neurodevelopmental disorders are multi-faceted and can lead to intellectual disability, autism spectrum disorder and language impairment. Mutations in the Forkhead box FOXP1 gene have been linked to all these disorders, suggesting that it may play a central role in various cognitive and social processes. To understand the role of Foxp1 in the context of neurodevelopment leading to alterations in cognition and behaviour, we generated mice with a brain-specific Foxp1 deletion (Nestin-Cre(Foxp1-/-)mice). The mutant mice were viable and allowed for the first time the analysis of pre- and postnatal neurodevelopmental phenotypes, which included a pronounced disruption of the developing striatum and more subtle alterations in the hippocampus. More detailed analysis in the CA1 region revealed abnormal neuronal morphogenesis that was associated with reduced excitability and an imbalance of excitatory to inhibitory input in CA1 hippocampal neurons in Nestin-Cre(Foxp1-/-) mice. Foxp1 ablation was also associated with various cognitive and social deficits, providing new insights into its behavioural importance. |
| FOXP1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22960337  Title: The role of the FOXP family of transcription factors in ASD.  Autism spectrum disorders (ASD) is a neurodevelopmental disease with complex genetics; however, the genes that are responsible for this disease still remain mostly unknown. Here, we focus on the FOXP family of transcription factors as there is emerging evidence strongly linking these genes to ASD and other genes implicated in ASD. The FOXP family of genes includes three genes expressed in the central nervous system: FOXP1, FOPX2, and FOXP4. This unique group of transcription factors has known functions in brain development as well as the evolution of language. We will also discuss the other genes including transcriptional targets of FOXP genes that have been found to be associated with language and may be important in the pathophysiology of ASD. Finally, we will review the emerging animal models currently being used to study the function of the FOXP genes within the context of ASD symptomology. The combination of gene expression and animal behavior is critical for elucidating how genes such as the FOXP family members are key players within the framework of the developing brain. |
| FOXP1, FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20950788  Title: De novo mutations in FOXP1 in cases with intellectual disability, autism, and language impairment.  Heterozygous mutations in FOXP2, which encodes a forkhead transcription factor, have been shown to cause developmental verbal dyspraxia and language impairment. FOXP2 and its closest homolog, FOXP1, are coexpressed in brain regions that are important for language and cooperatively regulate developmental processes, raising the possibility that FOXP1 may also be involved in developmental conditions that are associated with language impairment. In order to explore this possibility, we searched for mutations in FOXP1 in patients with intellectual disability (ID; mental retardation) and/or autism spectrum disorders (ASD). We first performed array-based genomic hybridization on sporadic nonsyndromic ID (NSID) (n = 30) or ASD (n = 80) cases. We identified a de novo intragenic deletion encompassing exons 4-14 of FOXP1 in a patient with NSID and autistic features. In addition, sequencing of all coding exons of FOXP1 in sporadic NSID (n = 110) or ASD (n = 135) cases, as well as in 570 controls, revealed the presence of a de novo nonsense mutation (c.1573C>T [p.R525X]) in the conserved forkhead DNA-binding domain in a patient with NSID and autism. Luciferase reporter assays showed that the p.R525X alteration disrupts the activity of the protein. Formal assessments revealed that both patients with de novo mutations in FOXP1 also show severe language impairment, mood lability with physical aggressiveness, and specific obsessions and compulsions. In conclusion, both FOXP1 and FOXP2 are associated with language impairment, but decrease of the former has a more global impact on brain development than that of the latter. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36382065  Title: FOXP2 down expression is associated with executive dysfunctions and electrophysiological abnormalities of brain in Autism spectrum disorder; a neuroimaging genetic study.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder characterized by language impairment, and challenges with social interaction, communication, and repetitive behaviors. Although genetics are a primary cause of ASD, the exact genes and molecular mechanisms involved in its pathogenesis are not completely clear. The FOXP2 gene encodes a transcription factor that is known for its major role in language development and severe speech problems. The present study aimed to evaluate the role of FOXP2 in ASD etiology, executive functions, and brain activities. In the present study, we recruited 450 children with ASD and 490 neurotypical control children. Three domains of executive functions (working memory, response inhibition, and vigilance) were assessed. In addition, five-minute eyes closed electroencephalography was obtained from some of the children with ASD and neurotypical children. DNA sequence and expression level of FOXP2 in blood samples of children with ASD and the control group were evaluated by using sequencing and Real-time PCR, respectively. The results showed no mutations but a significant down expression of FOXP2 genes in children with ASD vs. neurotypical children. Several cognitive and executive function deficiencies were detected in children with ASD. Low alpha and gamma bands in the frontal lobe and high theta bands in the occipital lobe were revealed in children with ASD. We also found several correlations between FOXP2 expression levels and clinical assessments. Our finding revealed the down expression of FOXP2, which could be considered as a biomarker for ASD as well as cognitive and executive dysfunction. Based on brain mapping data, FOXP2 may be related to the theta wave abnormality of children with ASD. FOXP2 may be considered a target of novel treatment to improve memory and executive functions. Our findings highlight the role of FOXP2 mRNA level in ASD etiology, executive functions, and brain wave frequencies. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35783275  Title: Retinoic Acid Supplementation Rescues the Social Deficits in Fmr1 Knockout Mice.  Autism spectrum disorder (ASD) is a heritable neurodevelopmental disorder with the underlying etiology yet incompletely understood and no cure treatment. Patients of fragile X syndrome (FXS) also manifest symptoms, e.g. deficits in social behaviors, that are core traits with ASD. Several studies demonstrated that a mutual defect in retinoic acid (RA) signaling was observed in FXS and ASD. However, it is still unknown whether RA replenishment could pose a positive effect on autistic-like behaviors in FXS. Herein, we found that RA signaling was indeed down-regulated when the expression of FMR1 was impaired in SH-SY5Y cells. Furthermore, RA supplementation rescued the atypical social novelty behavior, but failed to alleviate the defects in sociability behavior or hyperactivity, in Fmr1 knock-out (KO) mouse model. The repetitive behavior and motor coordination appeared to be normal. The RNA sequencing results of the prefrontal cortex in Fmr1 KO mice indicated that deregulated expression of Foxp2, Tnfsf10, Lepr and other neuronal genes was restored to normal after RA treatment. Gene ontology terms of metabolic processes, extracellular matrix organization and behavioral pathways were enriched. Our findings provided a potential therapeutic intervention for social novelty defects in FXS. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35279744  Title: Dissecting the cross-trait effects of the FOXP2 GWAS hit on clinical and brain phenotypes in adults with ADHD.  The Forkhead box P2 (FOXP2) encodes for a transcription factor with a broad role in embryonic development. It is especially represented among GWAS hits for neurodevelopmental disorders and related traits, including attention-deficit/hyperactivity disorder (ADHD), autism spectrum disorder, neuroticism, and risk-taking behaviors. While several functional studies are underway to understand the consequences of FOXP2 variation, this study aims to expand previous findings to clinically and genetically related phenotypes and neuroanatomical features among subjects with ADHD. The sample included 407 adults with ADHD and 463 controls. Genotyping was performed on the Infinium PsychArray-24 BeadChip, and the FOXP2 gene region was extracted. A gene-wide approach was adopted to evaluate the combined effects of FOXP2 variants (n = 311) on ADHD status, severity, comorbidities, and personality traits. Independent risk variants presenting potential functional effects were further tested for association with cortical surface areas in a subsample of cases (n = 87). The gene-wide analyses within the ADHD sample showed a significant association of the FOXP2 gene with harm avoidance (P = 0.001; PFDR = 0.015) and nominal associations with hyperactivity symptoms (P = 0.026; PFDR = 0.130) and antisocial personality disorder (P = 0.026; PFDR = 0.130). An insertion/deletion variant (rs79622555) located downstream of FOXP2 was associated with the three outcomes and nominally with the surface area of superior parietal and anterior cingulate cortices. Our results extend and refine previous GWAS findings pointing to a role of FOXP2 in several neurodevelopment-related phenotypes, mainly those involving underlying symptomatic domains of self-regulation and inhibitory control. Taken together, the available evidence may constitute promising insights into the puzzle of the FOXP2-related pathophysiology. |
| FOXP2, MEF2C, NR2F1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34667117  Title: Positive selection in noncoding genomic regions of vocal learning birds is associated with genes implicated in vocal learning and speech functions in humans.  Vocal learning, the ability to imitate sounds from conspecifics and the environment, is a key component of human spoken language and learned song in three independently evolved avian groups-oscine songbirds, parrots, and hummingbirds. Humans and each of these three bird clades exhibit specialized behavioral, neuroanatomical, and brain gene expression convergence related to vocal learning, speech, and song. To understand the evolutionary basis of vocal learning gene specializations and convergence, we searched for and identified accelerated genomic regions (ARs), a marker of positive selection, specific to vocal learning birds. We found avian vocal learner-specific ARs, and they were enriched in noncoding regions near genes with known speech functions or brain gene expression specializations in humans and vocal learning birds, including FOXP2, NEUROD6, ZEB2, and MEF2C, and near genes with major neurodevelopmental functions, including NR2F1, NRP2, and BCL11B We also found enrichment near the SFARI class S genes associated with syndromic vocal communication forms of autism spectrum disorders. These findings reveal strong candidate noncoding regions near genes for the evolutionary adaptations that distinguish vocal learning species from their close vocal nonlearning relatives and provide further evidence of molecular convergence between birdsong and human spoken language. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34650032  Title: Increased locomotor activity via regulation of GABAergic signalling in foxp2 mutant zebrafish-implications for neurodevelopmental disorders.  Recent advances in the genetics of neurodevelopmental disorders (NDDs) have identified the transcription factor FOXP2 as one of numerous risk genes, e.g. in autism spectrum disorders (ASD) and attention-deficit/hyperactivity disorder (ADHD). FOXP2 function is suggested to be involved in GABAergic signalling and numerous studies demonstrate that GABAergic function is altered in NDDs, thus disrupting the excitation/inhibition balance. Interestingly, GABAergic signalling components, including glutamate-decarboxylase 1 (Gad1) and GABA receptors, are putative transcriptional targets of FOXP2. However, the specific role of FOXP2 in the pathomechanism of NDDs remains elusive. Here we test the hypothesis that Foxp2 affects behavioural dimensions via GABAergic signalling using zebrafish as model organism. We demonstrate that foxp2 is expressed by a subset of GABAergic neurons located in brain regions involved in motor functions, including the subpallium, posterior tuberculum, thalamus and medulla oblongata. Using CRISPR/Cas9 gene-editing we generated a novel foxp2 zebrafish loss-of-function mutant that exhibits increased locomotor activity. Further, genetic and/or pharmacological disruption of Gad1 or GABA-A receptors causes increased locomotor activity, resembling the phenotype of foxp2 mutants. Application of muscimol, a GABA-A receptor agonist, rescues the hyperactive phenotype induced by the foxp2 loss-of-function. By reverse translation of the therapeutic effect on hyperactive behaviour exerted by methylphenidate, we note that application of methylphenidate evokes different responses in wildtype compared to foxp2 or gad1b loss-of-function animals. Together, our findings support the hypothesis that foxp2 regulates locomotor activity via GABAergic signalling. This provides one targetable mechanism, which may contribute to behavioural phenotypes commonly observed in NDDs. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34421555  Title: Sex-Specific Social Behavior and Amygdala Proteomic Deficits in Foxp2 +/- Mutant Mice.  In humans, mutations in the transcription factor encoding gene, FOXP2, are associated with language and Autism Spectrum Disorders (ASD), the latter characterized by deficits in social interactions. However, little is known regarding the function of Foxp2 in male or female social behavior. Our previous studies in mice revealed high expression of Foxp2 within the medial subnucleus of the amygdala (MeA), a limbic brain region highly implicated in innate social behaviors such as mating, aggression, and parental care. Here, using a comprehensive panel of behavioral tests in male and female Foxp2 +/- heterozygous mice, we investigated the role Foxp2 plays in MeA-linked innate social behaviors. We reveal significant deficits in olfactory processing, social interaction, mating, aggressive, and parental behaviors. Interestingly, some of these deficits are displayed in a sex-specific manner. To examine the consequences of Foxp2 loss of function specifically in the MeA, we conducted a proteomic analysis of microdissected MeA tissue. This analyses revealed putative sex differences expression of a host of proteins implicated in neuronal communication, connectivity, and dopamine signaling. Consistent with this, we discovered that MeA Foxp2-lineage cells were responsive to dopamine with differences between males and females. Thus, our findings reveal a central and sex-specific role for Foxp2 in social behavior and MeA function. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32622339  Title: Genetic pathways involved in human speech disorders.  Rare genetic variants that disrupt speech development provide entry points for deciphering the neurobiological foundations of key human capacities. The value of this approach is illustrated by FOXP2, a transcription factor gene that was implicated in speech apraxia, and subsequently investigated using human cell-based systems and animal models. Advances in next-generation sequencing, coupled to de novo paradigms, facilitated discovery of etiological variants in additional genes in speech disorder cohorts. As for other neurodevelopmental syndromes, gene-driven studies show blurring of boundaries between diagnostic categories, with some risk genes shared across speech disorders, intellectual disability and autism. Convergent evidence hints at involvement of regulatory genes co-expressed in early human brain development, suggesting that etiological pathways could be amenable for investigation in emerging neural models such as cerebral organoids. |
| FOXP2, NFIA, NFIB |
| Url: https://pubmed.ncbi.nlm.nih.gov/31067457  Title: Chromatin Decondensation by FOXP2 Promotes Human Neuron Maturation and Expression of Neurodevelopmental Disease Genes.  Forkhead box P2 (FOXP2) is a transcription factor expressed in the human brain that peaks during fetal development, and disruption in its ability to regulate downstream target genes leads to vulnerability to neurodevelopmental disorders. However, the mechanisms by which FOXP2 exerts regulatory control over targets during neuronal maturation have not been fully elucidated. Here, we use genome-wide chromatin accessibility assays and transcriptome-wide expression analyses in differentiating human neurons to show that FOXP2 represses proliferation-promoting genes in a DNA-binding-dependent manner. In contrast, FOXP2 and its cofactors, NFIA and NFIB, activate neuronal maturation genes in a manner that does not require FOXP2 to interact with DNA directly. Moreover, comparisons with expression data from the developing human brain suggest that FOXP2 and NFIA- or NFIB-dependent chromatin alterations drive maturation of excitatory cortical neurons. Thus, FOXP2 and its NFI cofactors may be specifically important for the development of cortical circuits underlying neurodevelopmental disorders. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30816183  Title: Sex Differences in the Effects of Prenatal Bisphenol A Exposure on Genes Associated with Autism Spectrum Disorder in the Hippocampus.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder inexplicably biased towards males. Although prenatal exposure to bisphenol A (BPA) has recently been associated with the ASD risk, whether BPA dysregulates ASD-related genes in the developing brain remains unclear. In this study, transcriptome profiling by RNA-seq analysis of hippocampi isolated from neonatal pups prenatally exposed to BPA was conducted and revealed a list of differentially expressed genes (DEGs) associated with ASD. Among the DEGs, several ASD candidate genes, including Auts2 and Foxp2, were dysregulated and showed sex differences in response to BPA exposure. The interactome and pathway analyses of DEGs using Ingenuity Pathway Analysis software revealed significant associations between the DEGs in males and neurological functions/disorders associated with ASD. Moreover, the reanalysis of transcriptome profiling data from previously published BPA studies consistently showed that BPA-responsive genes were significantly associated with ASD-related genes. The findings from this study indicate that prenatal BPA exposure alters the expression of ASD-linked genes in the hippocampus and suggest that maternal BPA exposure may increase ASD susceptibility by dysregulating genes associated with neurological functions known to be negatively impacted in ASD, which deserves further investigations. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30586385  Title: Comprehensive cross-disorder analyses of CNTNAP2 suggest it is unlikely to be a primary risk gene for psychiatric disorders.  The contactin-associated protein-like 2 (CNTNAP2) gene is a member of the neurexin superfamily. CNTNAP2 was first implicated in the cortical dysplasia-focal epilepsy (CDFE) syndrome, a recessive disease characterized by intellectual disability, epilepsy, language impairments and autistic features. Associated SNPs and heterozygous deletions in CNTNAP2 were subsequently reported in autism, schizophrenia and other psychiatric or neurological disorders. We aimed to comprehensively examine evidence for the role of CNTNAP2 in susceptibility to psychiatric disorders, by the analysis of multiple classes of genetic variation in large genomic datasets. In this study we used: i) summary statistics from the Psychiatric Genomics Consortium (PGC) GWAS for seven psychiatric disorders; ii) examined all reported CNTNAP2 structural variants in patients and controls; iii) performed cross-disorder analysis of functional or previously associated SNPs; and iv) conducted burden tests for pathogenic rare variants using sequencing data (4,483 ASD and 6,135 schizophrenia cases, and 13,042 controls). The distribution of CNVs across CNTNAP2 in psychiatric cases from previous reports was no different from controls of the database of genomic variants. Gene-based association testing did not implicate common variants in autism, schizophrenia or other psychiatric phenotypes. The association of proposed functional SNPs rs7794745 and rs2710102, reported to influence brain connectivity, was not replicated; nor did predicted functional SNPs yield significant results in meta-analysis across psychiatric disorders at either SNP-level or gene-level. Disrupting CNTNAP2 rare variant burden was not higher in autism or schizophrenia compared to controls. Finally, in a CNV mircroarray study of an extended bipolar disorder family with 5 affected relatives we previously identified a 131kb deletion in CNTNAP2 intron 1, removing a FOXP2 transcription factor binding site. Quantitative-PCR validation and segregation analysis of this CNV revealed imperfect segregation with BD. This large comprehensive study indicates that CNTNAP2 may not be a robust risk gene for psychiatric phenotypes. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30357341  Title: Altered social behavior in mice carrying a cortical Foxp2 deletion.  Genetic disruptions of the forkhead box transcription factor FOXP2 in humans cause an autosomal-dominant speech and language disorder. While FOXP2 expression pattern are highly conserved, its role in specific brain areas for mammalian social behaviors remains largely unknown. Here we studied mice carrying a homozygous cortical Foxp2 deletion. The postnatal development and gross morphological architecture of mutant mice was indistinguishable from wildtype (WT) littermates. Unbiased behavioral profiling of adult mice revealed abnormalities in approach behavior towards conspecifics as well as in the reciprocal responses of WT interaction partners. Furthermore mutant mice showed alterations in acoustical parameters of ultrasonic vocalizations, which also differed in function of the social context. Cell type-specific gene expression profiling of cortical pyramidal neurons revealed aberrant regulation of genes involved in social behavior. In particular Foxp2 mutants showed the downregulation of Mint2 (Apba2), a gene involved in approach behavior in mice and autism spectrum disorder in humans. Taken together these data demonstrate that cortical Foxp2 is required for normal social behaviors in mice. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30294994  Title: Aetiology of childhood apraxia of speech: A clinical practice update for paediatricians.  Childhood apraxia of speech (CAS) is a rare disorder of childhood that can leave a watermark of the impacts throughout the lifetime. Since being first described in the 1950s, aetiological insights have been limited. At a neurobiological level, clinical MRI scans fail to reveal overt neural anomalies in individual cases with CAS, although quantitative MRI methods have revealed subtle brain anomalies at a group level. Dramatic insights, however, occurred in the past decade from the discovery of genetic pathways underlying the phenotype. Several single genes and copy number-variant conditions are now associated with CAS either in relative isolation, as in the case of FOXP2 variants, or most typically in association with other neurodevelopmental conditions, such as epilepsy, intellectual disability, motor impairment and autism. CAS requires careful differential diagnosis from other childhood speech disorders, but when a severe and persistent diagnosis is confirmed, a genetic aetiology should increasingly be pursued. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29559544  Title: Vitamin D is crucial for maternal care and offspring social behaviour in rats.  Early life vitamin D plays a prominent role in neurodevelopment and subsequent brain function, including schizophrenic-like outcomes and increasing evidence for an association with autism spectrum disorder (ASD). Here, we investigate how early life vitamin D deficiency during rat pregnancy and lactation alters maternal care and influences neurodevelopment and affective, cognitive and social behaviours in male adult offspring. Sprague-Dawley rats were placed on either a vitamin D control (2195 IU/kg) or deficient diet (0 IU/kg) for five weeks before timed mating, and diet exposure was maintained until weaning of offspring on postnatal day (PND) 23. MRI scans were conducted to assess brain morphology, and plasma corticosterone levels and neural expression of genes associated with language, dopamine and glucocorticoid exposure were characterised at PND1, PND12 and 4 months of age. Compared to controls, vitamin D-deficient dams exhibited decreased licking and grooming of their pups but no differences in pup retrieval. Offspring neurodevelopmental markers were unaltered, but vitamin D-deficient pup ultrasonic vocalisations were atypical. As adults, males that had been exposed to vitamin D deficiency in early life exhibited decreased social behaviour, impaired learning and memory outcomes and increased grooming behaviour, but unaltered affective behaviours. Accompanying these behavioural changes was an increase in lateral ventricle volume, decreased cortical FOXP2 (a protein implicated in language and communication) and altered neural expression of genes involved in dopamine and glucocorticoid-related pathways. These data highlight that early life levels of vitamin D are an important consideration for maternal behavioural adaptations as well as offspring neuropsychiatry. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29123465  Title: Autoimmunity as a Driving Force of Cognitive Evolution.  In the last decades, increasingly robust experimental approaches have formally demonstrated that autoimmunity is a physiological process involved in a large range of functions including cognition. On this basis, the recently enunciated "brain superautoantigens" theory proposes that autoimmunity has been a driving force of cognitive evolution. It is notably suggested that the immune and nervous systems have somehow co-evolved and exerted a mutual selection pressure benefiting to both systems. In this two-way process, the evolutionary-determined emergence of neurons expressing specific immunogenic antigens (brain superautoantigens) has exerted a selection pressure on immune genes shaping the T-cell repertoire. Such a selection pressure on immune genes has translated into the emergence of a finely tuned autoimmune T-cell repertoire that promotes cognition. In another hand, the evolutionary-determined emergence of brain-autoreactive T-cells has exerted a selection pressure on neural genes coding for brain superautoantigens. Such a selection pressure has translated into the emergence of a neural repertoire (defined here as the whole of neurons, synapses and non-neuronal cells involved in cognitive functions) expressing brain superautoantigens. Overall, the brain superautoantigens theory suggests that cognitive evolution might have been primarily driven by internal cues rather than external environmental conditions. Importantly, while providing a unique molecular connection between neural and T-cell repertoires under physiological conditions, brain superautoantigens may also constitute an Achilles heel responsible for the particular susceptibility of Homo sapiens to "neuroimmune co-pathologies" i.e., disorders affecting both neural and T-cell repertoires. These may notably include paraneoplastic syndromes, multiple sclerosis as well as autism, schizophrenia and neurodegenerative diseases. In the context of this theoretical frame, a specific emphasis is given here to the potential evolutionary role exerted by two families of genes, namely the MHC class II genes, involved in antigen presentation to T-cells, and the Foxp genes, which play crucial roles in language (Foxp2) and the regulation of autoimmunity (Foxp3). |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28798667  Title: The FOXP2-Driven Network in Developmental Disorders and Neurodegeneration.  The transcription repressor FOXP2 is a crucial player in nervous system evolution and development of humans and songbirds. In order to provide an additional insight into its functional role we compared target gene expression levels between human neuroblastoma cells (SH-SY5Y) stably overexpressing FOXP2 cDNA of either humans or the common chimpanzee, Rhesus monkey, and marmoset, respectively. RNA-seq led to identification of 27 genes with differential regulation under the control of human FOXP2, which were previously reported to have FOXP2-driven and/or songbird song-related expression regulation. RT-qPCR and Western blotting indicated differential regulation of additional 13 new target genes in response to overexpression of human FOXP2. These genes may be directly regulated by FOXP2 considering numerous matches of established FOXP2-binding motifs as well as publicly available FOXP2-ChIP-seq reads within their putative promoters. Ontology analysis of the new and reproduced targets, along with their interactors in a network, revealed an enrichment of terms relating to cellular signaling and communication, metabolism and catabolism, cellular migration and differentiation, and expression regulation. Notably, terms including the words "neuron" or "axonogenesis" were also enriched. Complementary literature screening uncovered many connections to human developmental (autism spectrum disease, schizophrenia, Down syndrome, agenesis of corpus callosum, trismus-pseudocamptodactyly, ankyloglossia, facial dysmorphology) and neurodegenerative diseases and disorders (Alzheimer's, Parkinson's, and Huntington's diseases, Lewy body dementia, amyotrophic lateral sclerosis). Links to deafness and dyslexia were detected, too. Such relations existed for single proteins (e.g., DCDC2, NURR1, PHOX2B, MYH8, and MYH13) and groups of proteins which conjointly function in mRNA processing, ribosomal recruitment, cell-cell adhesion (e.g., CDH4), cytoskeleton organization, neuro-inflammation, and processing of amyloid precursor protein. Conspicuously, many links pointed to an involvement of the FOXP2-driven network in JAK/STAT signaling and the regulation of the ezrin-radixin-moesin complex. Altogether, the applied phylogenetic perspective substantiated FOXP2's importance for nervous system development, maintenance, and functioning. However, the study also disclosed new regulatory pathways that might prove to be useful for understanding the molecular background of the aforementioned developmental disorders and neurodegenerative diseases. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28687613  Title: Valproic acid induces aberrant development of striatal compartments and corticostriatal pathways in a mouse model of autism spectrum disorder.  The striatum comprises two neurochemical compartments: striosomes and the matrix. Striosomal and matrix compartments receive inputs from limbic system-related and sensorimotor cortices, respectively. Here, we investigate the impact on the corticostriosomal pathway in the valproic acid (VPA)-induced autism spectrum disorder mouse model. VPA administration during the neurogenesis time windows of striosomes, but not the matrix, resulted in aberrant compartmentation [i.e., maternal VPA injections at embryonic day (E)12.75 decreased μ-opioid receptor-positive striosomes, but increased calbindin-positive matrix in the rostral striatum]. VPAE12.75 treatment also impaired the aggregation of cells pulse labeled with 5-bromo-2'-deoxyuridine at E12.75 into striosomal cell clusters, which suggests defective segregation of striosomal cells from matrix cells. This possibility was supported by our findings that VPAE12.75 treatment altered the expression of ephrinA5 and EphA4, two molecules that are related to compartmental segregation. In the VPAE12.75 neocortex, Foxp2-positive neurons were decreased in layer VI, but increased in layer V, which projects to the striosomal compartment. We also investigated VPA effects on the corticostriosomal pathway. VPAE12.75 treatment decreased the putative corticostriosomal synapses of striosomal neurons and induced an aberrant pattern of isolation stress-induced ultrasonic vocalizations. Of interest, risperidone treatments conjointly improved ultrasonic vocalizations and restored the striosomal compartment in VPAE12.75 pups. Collectively, dysfunctional corticostriatal pathways, particularly via the aberrant striosomal compartment, may be involved in autism spectrum disorder pathophysiology.-Kuo, H.-Y., Liu, F.-C. Valproic acid induces aberrant development of striatal compartments and corticostriatal pathways in a mouse model of autism spectrum disorder. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27867345  Title: Language Impairment Resulting from a de novo Deletion of 7q32.1q33.  We report on a girl who presents with hearing loss, behavioral disturbances (according to the Inventory for Client and Agency Planning) as well as motor and cognitive delay (according to Battelle Developmental Inventories) which have a significant impact on her speech and language abilities [according to the Peabody Picture Vocabulary Test (ed 3), and the Prueba de Lenguaje Oral de Navarra-Revisada (Navarra Oral Language Test, Revised)]. Five copy number variations (CNVs) were identified in the child: arr[hg18] 7q32.1q33(127109685-132492196)×1, 8p23.1(7156900-7359099) ×1, 15q13.1(26215673-26884937)×1, Xp22.33(17245- 102434)×3, and Xp22.33(964441-965024)×3. The pathogenicity of similar CNVs is mostly reported as unknown. The largest deletion is found in a hot spot for cognitive disease and language impairment and contains several genes involved in brain development and function, many of which have been related to developmental disorders encompassing language deficits (dyslexia, speech-sound disorder, and autism). Some of these genes interact with FOXP2. The proband's phenotype may result from a reduced expression of some of these genes. |
| FOXP2, MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/27595386  Title: Foxp2 controls synaptic wiring of corticostriatal circuits and vocal communication by opposing Mef2c.  Cortico-basal ganglia circuits are critical for speech and language and are implicated in autism spectrum disorder, in which language function can be severely affected. We demonstrate that in the mouse striatum, the gene Foxp2 negatively interacts with the synapse suppressor gene Mef2c. We present causal evidence that Mef2c inhibition by Foxp2 in neonatal mouse striatum controls synaptogenesis of corticostriatal inputs and vocalization in neonates. Mef2c suppresses corticostriatal synapse formation and striatal spinogenesis, but can itself be repressed by Foxp2 through direct DNA binding. Foxp2 deletion de-represses Mef2c, and both intrastriatal and global decrease of Mef2c rescue vocalization and striatal spinogenesis defects of Foxp2-deletion mutants. These findings suggest that Foxp2-Mef2C signaling is critical to corticostriatal circuit formation. If found in humans, such signaling defects could contribute to a range of neurologic and neuropsychiatric disorders. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26442845  Title: Understanding Language from a Genomic Perspective.  Language is a defining characteristic of the human species, but its foundations remain mysterious. Heritable disorders offer a gateway into biological underpinnings, as illustrated by the discovery that FOXP2 disruptions cause a rare form of speech and language impairment. The genetic architecture underlying language-related disorders is complex, and although some progress has been made, it has proved challenging to pinpoint additional relevant genes with confidence. Next-generation sequencing and genome-wide association studies are revolutionizing understanding of the genetic bases of other neurodevelopmental disorders, like autism and schizophrenia, and providing fundamental insights into the molecular networks crucial for typical brain development. We discuss how a similar genomic perspective, brought to the investigation of language-related phenotypes, promises to yield equally informative discoveries. Moreover, we outline how follow-up studies of genetic findings using cellular systems and animal models can help to elucidate the biological mechanisms involved in the development of brain circuits supporting language. |
| FOXP2, OTX1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24098394  Title: Transcriptome comparison of human neurons generated using induced pluripotent stem cells derived from dental pulp and skin fibroblasts.  Induced pluripotent stem cell (iPSC) technology is providing an opportunity to study neuropsychiatric disorders through the capacity to grow patient-specific neurons in vitro. Skin fibroblasts obtained by biopsy have been the most reliable source of cells for reprogramming. However, using other somatic cells obtained by less invasive means would be ideal, especially in children with autism spectrum disorders (ASD) and other neurodevelopmental conditions. In addition to fibroblasts, iPSCs have been developed from cord blood, lymphocytes, hair keratinocytes, and dental pulp from deciduous teeth. Of these, dental pulp would be a good source for neurodevelopmental disorders in children because obtaining material is non-invasive. We investigated its suitability for disease modeling by carrying out gene expression profiling, using RNA-seq, on differentiated neurons derived from iPSCs made from dental pulp extracted from deciduous teeth (T-iPSCs) and fibroblasts (F-iPSCs). This is the first RNA-seq analysis comparing gene expression profiles in neurons derived from iPSCs made from different somatic cells. For the most part, gene expression profiles were quite similar with only 329 genes showing differential expression at a nominally significant p-value (p<0.05), of which 63 remained significant after correcting for genome-wide analysis (FDR <0.05). The most striking difference was the lower level of expression detected for numerous members of the all four HOX gene families in neurons derived from T-iPSCs. In addition, an increased level of expression was seen for several transcription factors expressed in the developing forebrain (FOXP2, OTX1, and LHX2, for example). Overall, pathway analysis revealed that differentially expressed genes that showed higher levels of expression in neurons derived from T-iPSCs were enriched for genes implicated in schizophrenia (SZ). The findings suggest that neurons derived from T-iPSCs are suitable for disease-modeling neuropsychiatric disorder and may have some advantages over those derived from F-iPSCs. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22272290  Title: Cadm1-expressing synapses on Purkinje cell dendrites are involved in mouse ultrasonic vocalization activity.  Foxp2(R552H) knock-in (KI) mouse pups with a mutation related to human speech-language disorders exhibit poor development of cerebellar Purkinje cells and impaired ultrasonic vocalization (USV), a communication tool for mother-offspring interactions. Thus, human speech and mouse USV appear to have a Foxp2-mediated common molecular basis in the cerebellum. Mutations in the gene encoding the synaptic adhesion molecule CADM1 (RA175/Necl2/SynCAM1/Cadm1) have been identified in people with autism spectrum disorder (ASD) who have impaired speech and language. In the present study, we show that both Cadm1-deficient knockout (KO) pups and Foxp2(R552H) KI pups exhibit impaired USV and smaller cerebellums. Cadm1 was preferentially localized to the apical-distal portion of the dendritic arbor of Purkinje cells in the molecular layer of wild-type pups, and VGluT1 level decreased in the cerebellum of Cadm1 KO mice. In addition, we detected reduced immunoreactivity of Cadm1 and VGluT1 on the poorly developed dendritic arbor of Purkinje cells in the Foxp2(R552H) KI pups. However, Cadm1 mRNA expression was not altered in the Foxp2(R552H) KI pups. These results suggest that although the Foxp2 transcription factor does not target Cadm1, Cadm1 at the synapses of Purkinje cells and parallel fibers is necessary for USV function. The loss of Cadm1-expressing synapses on the dendrites of Purkinje cells may be associated with the USV impairment that Cadm1 KO and Foxp2(R552H) KI mice exhibit. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21711233  Title: The role of the urokinase receptor in epilepsy, in disorders of language, cognition, communication and behavior, and in the central nervous system.  As a key component of the plasminogen activation system, uPAR, the receptor for the plasminogen activator of the urokinase type, is involved in many physiological and pathological processes. Besides its classical roles, there has been increased evidence that uPAR or uPAR-associated pathways, participate in the development, in the functioning and in the pathology of the central nervous system. Qualitative and quantitative changes in the expressions of uPAR and of its canonical ligand uPA have been observed in a large variety of epileptic disorders, either in human or in animal models, as well as in other brain diseases (stroke and brain trauma, multiple sclerosis, Alzheimer's disease, cerebral malaria, HIV-associated leukoencephalopathy and encephalitis). The variety of such pathological conditions and the different brain areas and cell types involved, likely reflects the wide range and the complexity of the multiple and somehow intertwined pathophysiological mechanisms related with uPAR. In the mouse, the knock-out of the Upar-encoding gene (Plaur) leads to significant and nearly complete loss in parvalbumin-containing interneurons during brain development. This is associated with increased susceptibility to spontaneous and chemically-induced seizures and with increased anxiety and impaired social interactions. The recent identification of the novel uPAR ligand SRPX2 (Sushi repeat protein, X-linked 2) and the regulation of both the SRPX2 and PLAUR genes by transcription factor FOXP2 has shed novel and exciting insights into the role of uPAR-related molecular networks in rolandic epilepsy, in developmental verbal dyspraxia, in perisylvian polymicrogyria, and generally in disorders of the speech areas and circuits. uPAR, its regulators and partners, as well as other proteins containing Ly-6/uPAR/alpha-neurotoxin domains, represent key entry points for present and future studies not only on speech-related disorders but also on epilepsy and autism spectrum disorders. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20579107  Title: Ultrasonic vocalizations in mouse models for speech and socio-cognitive disorders: insights into the evolution of vocal communication.  Comparative analyses used to reconstruct the evolution of traits associated with the human language faculty, including its socio-cognitive underpinnings, highlight the importance of evolutionary constraints limiting vocal learning in non-human primates. After a brief overview of this field of research and the neural basis of primate vocalizations, we review studies that have addressed the genetic basis of usage and structure of ultrasonic communication in mice, with a focus on the gene FOXP2 involved in specific language impairments and neuroligin genes (NL-3 and NL-4) involved in autism spectrum disorders. Knockout of FoxP2 leads to reduced vocal behavior and eventually premature death. Introducing the human variant of FoxP2 protein into mice, in contrast, results in shifts in frequency and modulation of pup ultrasonic vocalizations. Knockout of NL-3 and NL-4 in mice diminishes social behavior and vocalizations. Although such studies may provide insights into the molecular and neural basis of social and communicative behavior, the structure of mouse vocalizations is largely innate, limiting the suitability of the mouse model to study human speech, a learned mode of production. Although knockout or replacement of single genes has perceptible effects on behavior, these genes are part of larger networks whose functions remain poorly understood. In humans, for instance, deficiencies in NL-4 can lead to a broad spectrum of disorders, suggesting that further factors (experiential and/or genetic) contribute to the variation in clinical symptoms. The precise nature as well as the interaction of these factors is yet to be determined. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18248790  Title: Maternal infection leads to abnormal gene regulation and brain atrophy in mouse offspring: implications for genesis of neurodevelopmental disorders.  Prenatal viral infection has been associated with development of schizophrenia and autism. Our laboratory has previously shown that viral infection causes deleterious effects on brain structure and function in mouse offspring following late first trimester (E9) administration of influenza virus. We hypothesized that late second trimester infection (E18) in mice may lead to a different pattern of brain gene expression and structural defects in the developing offspring. C57BL6J mice were infected on E18 with a sublethal dose of human influenza virus or sham-infected using vehicle solution. Male offsping of the infected mice were collected at P0, P14, P35 and P56, their brains removed and prefrontal cortex, hippocampus and cerebellum dissected and flash frozen. Microarray, qRT-PCR, DTI and MRI scanning, western blotting and neurochemical analysis were performed to detect differences in gene expression and brain atrophy. Expression of several genes associated with schizophrenia or autism including Sema3a, Trfr2 and Vldlr were found to be altered as were protein levels of Foxp2. E18 infection of C57BL6J mice with a sublethal dose of human influenza virus led to significant gene alterations in frontal, hippocampal and cerebellar cortices of developing mouse progeny. Brain imaging revealed significant atrophy in several brain areas and white matter thinning in corpus callosum. Finally, neurochemical analysis revealed significantly altered levels of serotonin (P14, P35), 5-Hydroxyindoleacetic acid (P14) and taurine (P35). We propose that maternal infection in mouse provides an heuristic animal model for studying the environmental contributions to genesis of schizophrenia and autism, two important examples of neurodevelopmental disorders. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/16764847  Title: Tangled webs: tracing the connections between genes and cognition.  The rise of molecular genetics is having a pervasive influence in a wide variety of fields, including research into neurodevelopmental disorders like dyslexia, speech and language impairments, and autism. There are many studies underway which are attempting to determine the roles of genetic factors in the aetiology of these disorders. Beyond the obvious implications for diagnosis, treatment and understanding, success in these efforts promises to shed light on the links between genes and aspects of cognition and behaviour. However, the deceptive simplicity of finding correlations between genetic and phenotypic variation has led to a common misconception that there exist straightforward linear relationships between specific genes and particular behavioural and/or cognitive outputs. The problem is exacerbated by the adoption of an abstract view of the nature of the gene, without consideration of molecular, developmental or ontogenetic frameworks. To illustrate the limitations of this perspective, I select two cases from recent research into the genetic underpinnings of neurodevelopmental disorders. First, I discuss the proposal that dyslexia can be dissected into distinct components specified by different genes. Second, I review the story of the FOXP2 gene and its role in human speech and language. In both cases, adoption of an abstract concept of the gene can lead to erroneous conclusions, which are incompatible with current knowledge of molecular and developmental systems. Genes do not specify behaviours or cognitive processes; they make regulatory factors, signalling molecules, receptors, enzymes, and so on, that interact in highly complex networks, modulated by environmental influences, in order to build and maintain the brain. I propose that it is necessary for us to fully embrace the complexity of biological systems, if we are ever to untangle the webs that link genes to cognition. |
| FOXP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/15737702  Title: Absence of causative mutations and presence of autism-related allele in FOXP2 in Japanese autistic patients.  We analyzed the FOXP2 gene, which encodes a putative transcription factor containing a polyglutamine tract and a forkhead DNA-binding domain, for a possible causative mutation in autism. FOXP2 was reported to be mutated in patients with a severe speech and language disorder. FOXP2 was located on chromosome 7q31, which is one of the loci involved in autism. Autism and specific language impairment share some of their clinical phenotypes. In addition, FOXP2 was expressed abundantly in the brain. We screened all of the exons of FOXP2 for causative mutations in 53 Japanese autistic patients using denaturing high-performance liquid chromatography and direct sequencing. A delCAA in exon 5 causing one glutamine deletion in the first polyglutamine tract was detected in four patients and in 2 of 50 control individuals. The frequency of the TT allele with the G to T base change in intron 15 was significantly high in the autistic population. The other base changes included one silent base change (A569G) in exon 5 and three in introns. Our results may suggest a relationship between autism and the FOXP2 gene or a gene located nearby. |
| GTF2I |
| Url: https://pubmed.ncbi.nlm.nih.gov/33208191  Title: High-throughput screening identifies histone deacetylase inhibitors that modulate GTF2I expression in 7q11.23 microduplication autism spectrum disorder patient-derived cortical neurons.  Autism spectrum disorder (ASD) is a highly prevalent neurodevelopmental condition affecting almost 1% of children, and represents a major unmet medical need with no effective drug treatment available. Duplication at 7q11.23 (7Dup), encompassing 26-28 genes, is one of the best characterized ASD-causing copy number variations and offers unique translational opportunities, because the hemideletion of the same interval causes Williams-Beuren syndrome (WBS), a condition defined by hypersociability and language strengths, thereby providing a unique reference to validate treatments for the ASD symptoms. In the above-indicated interval at 7q11.23, defined as WBS critical region, several genes, such as GTF2I, BAZ1B, CLIP2 and EIF4H, emerged as critical for their role in the pathogenesis of WBS and 7Dup both from mouse models and human studies. We performed a high-throughput screening of 1478 compounds, including central nervous system agents, epigenetic modulators and experimental substances, on patient-derived cortical glutamatergic neurons differentiated from our cohort of induced pluripotent stem cell lines (iPSCs), monitoring the transcriptional modulation of WBS interval genes, with a special focus on GTF2I, in light of its overriding pathogenic role. The hits identified were validated by measuring gene expression by qRT-PCR and the results were confirmed by western blotting. We identified and selected three histone deacetylase inhibitors (HDACi) that decreased the abnormal expression level of GTF2I in 7Dup cortical glutamatergic neurons differentiated from four genetically different iPSC lines. We confirmed this effect also at the protein level. In this study, we did not address the molecular mechanisms whereby HDAC inhibitors act on GTF2I. The lead compounds identified will now need to be advanced to further testing in additional models, including patient-derived brain organoids and mouse models recapitulating the gene imbalances of the 7q11.23 microduplication, in order to validate their efficacy in rescuing phenotypes across multiple functional layers within a translational pipeline towards clinical use. These results represent a unique opportunity for the development of a specific class of compounds for treating 7Dup and other forms of intellectual disability and autism. |
| GTF2I |
| Url: https://pubmed.ncbi.nlm.nih.gov/29568691  Title: Consistent hypersocial behavior in mice carrying a deletion of Gtf2i but no evidence of hyposocial behavior with Gtf2i duplication: Implications for Williams-Beuren syndrome and autism spectrum disorder.  Williams-Beuren syndrome (WBS) is a developmental disorder caused by hemizygous deletion of human chromosome 7q11.23. Hypersocial behavior is one symptom of WBS and contrasts with hyposociality observed in autism spectrum disorder (ASD). Interestingly, duplications of 7q11.23 have been associated with ASD. The social phenotype of WBS has been linked to GTF2I or general transcription factor IIi (TFII-I). Duplication of GTF2I has also been associated with ASD. We compared mice having either a deletion (Gtf2i+/- ) or duplication (Gtf2i+/dup ) of Gtf2i to wild-type (Gtf2i+/+ ) littermate controls in a series of behavioral tasks including open-field activity monitoring, olfactory probes, a social choice task, social transmission of food preference, habituation-dishabituation, and operant social motivation paradigms. In open-field observations, Gtf2i+/- and Gtf2i+/dup mice demonstrated normal activity and thigmotaxis, and surprisingly, each strain showed a significant preference for a stimulus mouse that was not observed in Gtf2i+/+ siblings. Both Gtf2i+/- and Gtf2i+/dup mice demonstrated normal olfaction in buried food probes, but the Gtf2i+/- mice spent significantly more time investigating urine scent versus water, which was not observed in the other strains. Gtf2i+/- mice also spent significantly more time in nose-to-nose contact compared to Gtf2i+/+ siblings during the open-field encounter of the social transmission of food preference task. In operant tasks of social motivation, Gtf2i+/- mice made significantly more presses for social rewards than Gtf2i+/+ siblings, while there was no difference in presses for the Gtf2i+/dup mice. Results were remarkably consistent across testing paradigms supporting a role for GTF2i in the hypersocial phenotype of WBS and more broadly in the regulation of social behavior. Support was not observed for the role of GTF2i in ASD. |
| GTF2I |
| Url: https://pubmed.ncbi.nlm.nih.gov/25913238  Title: Novel splice variants in the 5'UTR of Gtf2i expressed in the rat brain: alternative 5'UTRs and differential expression in the neuronal dendrites.  General transcription factor II-I (Gtf2i) is a transcription factor and one of the genes implicated in Willams-Beuren syndrome, an autism spectrum disorder. In this study, we investigated splice variants of the Gtf2i gene in both the 5'untranslated region (5'UTR) and the coding region. To search for novel 5'UTRs of Gtf2i, we utilized the cap analysis gene expression database of the mouse. We identified seven novel Gtf2i transcripts with alternatively spliced 5'UTRs in the rat brain. We also identified four novel splice variants in the coding sequence of Gtf2i. Furthermore, we identified a selective usage of certain types of 5'UTR by coding variants. In situ hybridization demonstrated a differential pattern of expression of Gtf2i mRNAs with alternatively spliced 5'UTRs among neuronal cells, and the localization of one of the variants in neuronal dendrites in the rat brain. Immunohistochemistry also demonstrated a distribution of Gtf2i-immunoreactivity in the dendrites. These results suggest multiple pathways of expression of Gtf2i gene in the brain. The expression patterns may be under the control of alternative promoters coupled to the alternative splicing in the coding region. Differential localization of mRNA to neuronal dendrites suggests spatiotemporal-specific translation at the post-synaptic sites that is involved in transfer of synaptic activity to expression of specific sets of genes in the nucleus. Gtf2i is a transcription factor and implicated in Willams-Beuren syndrome. We identified seven novel Gtf2i transcripts with alternatively spliced 5'UTRs in the rat brain. In situ hybridization demonstrated a differential expression of Gtf2i mRNAs with different 5'UTRs in somas and dendrites of neuronal cells. Differential localization of mRNA to neuronal dendrites suggests spatiotemporal-specific translation at the postsynaptic sites. The scheme shows genomic structure showing the positions of the potential transcription start tags (rDEC695, rDEC3D7, rDEC1D3, rDEC104, rDEC072 and rDEBE25). Newly identified exons (1.1-1.6) are shown with the white boxes. The distances from rDEC695-5'end are indicated in bp. |
| GTF2I |
| Url: https://pubmed.ncbi.nlm.nih.gov/21328569  Title: Haploinsufficiency of Gtf2i, a gene deleted in Williams Syndrome, leads to increases in social interactions.  Identifying genes involved in social behavior is important for autism research. Williams-Beuren syndrome (WBS) is a developmental syndrome with unique neurocognitive features, including low IQ, deficits in visuospatial and visual-motor abilities, hypersensitivity to sounds, hypersociability, and increased general anxiety. The syndrome is caused by a recurrent hemizygous deletion of the 7q11.23 region, containing about 28 genes. One of genes in the region, GTF2I, has been implicated in the hypersociability and visuospatial deficits of WBS based on genotype-phenotype correlation studies of patients with atypical deletions. In order to clarify the involvement of GTF2I in neurocognitive function, especially social behavior, we have developed and characterized Gtf2i-deficient mice. We found that homozygous deletion of Gtf2i causes lethality during embryonic development with neural tube closure defects and exencephaly, consistent with other reports. Gtf2i heterozygous animals show no gross changes in brain structure or development. Furthermore, heterozygous animals show no alterations in learning and memory, including spatial memory as assessed by the Morris water maze, but show alterations in the recognition of novel objects. Interestingly, they show increased social interaction with unfamiliar mice and do not show typical social habituation processes, reminiscent of the hypersociability observed in WBS patients. The mice do not appear to show increased anxiety, supporting a specific effect of Gtf2i on defined domains of the WBS phenotype. These data indicate that Gtf2i is involved in several aspects of embryonic development and the development of social neurocircuitry and that GTF2I haploinsufficiency could be a contributor to the hypersociability in WBS patients. |
| HDAC4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25180572  Title: DNA methylation analysis of the autistic brain reveals multiple dysregulated biological pathways.  Autism spectrum disorders (ASD) are a group of neurodevelopmental conditions characterized by dysfunction in social interaction, communication and stereotypic behavior. Genetic and environmental factors have been implicated in the development of ASD, but the molecular mechanisms underlying their interaction are not clear. Epigenetic modifications have been suggested as molecular mechanism that can mediate the interaction between the environment and the genome to produce adaptive or maladaptive behaviors. Here, using the Illumina 450 K methylation array we have determined the existence of many dysregulated CpGs in two cortical regions, Brodmann area 10 (BA10) and Brodmann area 24 (BA24), of individuals who had ASD. In BA10 we found a very significant enrichment for genomic areas responsible for immune functions among the hypomethylated CpGs, whereas genes related to synaptic membrane were enriched among hypermethylated CpGs. By comparing our methylome data with previously published transcriptome data, and by performing real-time PCR on selected genes that were dysregulated in our study, we show that hypomethylated genes are often overexpressed, and that there is an inverse correlation between gene expression and DNA methylation within the individuals. Among these genes there were C1Q, C3, ITGB2 (C3R), TNF-α, IRF8 and SPI1, which have recently been implicated in synaptic pruning and microglial cell specification. Finally, we determined the epigenetic dysregulation of the gene HDAC4, and we confirm that the locus encompassing C11orf21/TSPAN32 has multiple hypomethylated CpGs in the autistic brain, as previously demonstrated. Our data suggest a possible role for epigenetic processes in the etiology of ASD. |
| HDAC4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24095776  Title: Siblings with opposite chromosome constitutions, dup(2q)/del(7q) and del(2q)/dup(7q).  Chromosome 7q36 microdeletion syndrome is a rare genomic disorder characterized by underdevelopment of the brain, microcephaly, anomalies of the sex organs, and language problems. Developmental delay, intellectual disability, autistic spectrum disorders, BDMR syndrome, and unusual facial morphology are the key features of the chromosome 2q37 microdeletion syndrome. A genetic screening for two brothers with global developmental delay using high-resolution chromosomal analysis and subtelomeric multiplex ligation-dependent probe amplification revealed subtelomeric rearrangements on the same sites of 2q37.2 and 7q35, with reversed deletion and duplication. Both of them showed dysmorphic facial features, severe disability of physical and intellectual development, and abnormal genitalia with differential abnormalities in their phenotypes. The family did not have abnormal genetic phenotypes. According to the genetic analysis of their parents, adjacent-1 segregation from their mother's was suggested as a mechanism of their gene mutation. By comparing the phenotypes of our patients with previous reports on similar patients, we tried to obtain the information of related genes and their chromosomal locations. |
| HDAC4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20691407  Title: Haploinsufficiency of HDAC4 causes brachydactyly mental retardation syndrome, with brachydactyly type E, developmental delays, and behavioral problems.  Brachydactyly mental retardation syndrome (BDMR) is associated with a deletion involving chromosome 2q37. BDMR presents with a range of features, including intellectual disabilities, developmental delays, behavioral abnormalities, sleep disturbance, craniofacial and skeletal abnormalities (including brachydactyly type E), and autism spectrum disorder. To date, only large deletions of 2q37 have been reported, making delineation of a critical region and subsequent identification of candidate genes difficult. We present clinical and molecular analysis of six individuals with overlapping deletions involving 2q37.3 that refine the critical region, reducing the candidate genes from >20 to a single gene, histone deacetylase 4 (HDAC4). Driven by the distinct hand and foot anomalies and similar cognitive features, we identified other cases with clinical findings consistent with BDMR but without a 2q37 deletion, and sequencing of HDAC4 identified de novo mutations, including one intragenic deletion probably disrupting normal splicing and one intragenic insertion that results in a frameshift and premature stop codon. HDAC4 is a histone deacetylase that regulates genes important in bone, muscle, neurological, and cardiac development. Reportedly, Hdac4(-/-) mice have severe bone malformations resulting from premature ossification of developing bones. Data presented here show that deletion or mutation of HDAC4 results in reduced expression of RAI1, which causes Smith-Magenis syndrome when haploinsufficient, providing a link to the overlapping findings in these disorders. Considering the known molecular function of HDAC4 and the mouse knockout phenotype, taken together with deletion or mutation of HDAC4 in multiple subjects with BDMR, we conclude that haploinsufficiency of HDAC4 results in brachydactyly mental retardation syndrome. |
| HMGN1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32392183  Title: Genetic regulatory subnetworks and key regulating genes in rat hippocampus perturbed by prenatal malnutrition: implications for major brain disorders.  Many population studies have shown that maternal prenatal nutrition deficiency may increase the risk of neurodevelopmental disorders in their offspring, but its potential transcriptomic effects on brain development are not clear. We aimed to investigate the transcriptional regulatory interactions between genes in particular pathways responding to the prenatal nutritional deficiency and to explore their effects on neurodevelopment and related disorders. We identified three modules in rat hippocampus responding to maternal prenatal nutritional deficiency and found 15 key genes (Hmgn1, Ssbp1, LOC684988, Rpl23, Gga1, Rhobtb2, Dhcr24, Atg9a, Dlgap3, Grm5, Scn2b, Furin, Sh3kbp1, Ubqln1, and Unc13a) related to the rat hippocampus developmental dysregulation, of which Hmgn1, Rhobtb2 and Unc13a related to autism, and Dlgap3, Grm5, Furin and Ubqln1 are related to Alzheimer's disease, and schizophrenia. Transcriptional alterations of the hub genes were confirmed except for Atg9a. Additionally, through modeling miRNA-mRNA-transcription factor interactions for the hub genes, we confirmed a transcription factor, Cebpa, is essential to regulate the expression of Rhobtb2. We did not find singificent singals in the prefrontal cortex responding to maternal prenatal nutritional deficiency. These findings demonstrated that these genes with the three modules in rat hippocampus involved in synaptic development, neuronal projection, cognitive function, and learning function are significantly enriched hippocampal CA1 pyramidal neurons and suggest that three genetic regulatory subnetworks and thirteen key regulating genes in rat hippocampus perturbed by a prenatal nutrition deficiency. These genes and related subnetworks may be prenatally involved in the etiologies of major brain disorders, including Alzheimer's disease, autism, and schizophrenia. We compared the transcriptomic differences in the hippocampus and prefrontal cortex between 10 rats with prenatal nutritional deficiency and 10 rats with prenatal normal chow feeding by differential analysis and co-expression network analysis. A network-driven integrative analysis with microRNAs and transcription factors was performed to define significant modules and hub genes responding to prenatal nutritional deficiency. Meanwhile, the module preservation test was conducted between the hippocampus and prefrontal cortex. Expression levels of the hub genes were further validated with a quantitative real-time polymerase chain reaction based on additional 40 pairs of rats. |
| HMGN1, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22009741  Title: The chromatin-binding protein HMGN1 regulates the expression of methyl CpG-binding protein 2 (MECP2) and affects the behavior of mice.  High mobility group N1 protein (HMGN1), a nucleosomal-binding protein that affects the structure and function of chromatin, is encoded by a gene located on chromosome 21 and is overexpressed in Down syndrome, one of the most prevalent genomic disorders. Misexpression of HMGN1 affects the cellular transcription profile; however, the biological function of this protein is still not fully understood. We report that HMGN1 modulates the expression of methyl CpG-binding protein 2 (MeCP2), a DNA-binding protein known to affect neurological functions including autism spectrum disorders, and whose alterations in HMGN1 levels affect the behavior of mice. Quantitative PCR and Western analyses of cell lines and brain tissues from mice that either overexpress or lack HMGN1 indicate that HMGN1 is a negative regulator of MeCP2 expression. Alterations in HMGN1 levels lead to changes in chromatin structure and histone modifications in the MeCP2 promoter. Behavior analyses by open field test, elevated plus maze, Reciprocal Social Interaction, and automated sociability test link changes in HMGN1 levels to abnormalities in activity and anxiety and to social deficits in mice. Targeted analysis of the Autism Genetic Resource Exchange genotype collection reveals a non-random distribution of genotypes within 500 kbp of HMGN1 in a region affecting its expression in families predisposed to autism spectrum disorders. Our results reveal that HMGN1 affects the behavior of mice and suggest that epigenetic changes resulting from altered HMGN1 levels could play a role in the etiology of neurodevelopmental disorders. |
| JARID2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29216786  Title: A study of single nucleotide polymorphisms in CD157, AIM2 and JARID2 genes in Han Chinese children with autism spectrum disorder.  Autism spectrum disorder (ASD) is a group of developmental brain disorders caused by genetic and environmental factors. The objective of this study was to investigate whether single nucleotide polymorphisms (SNPs) in genes related to immune function were associated with ASD in Chinese Han children. A total of 201 children with ASD and 200 age- and gender-matched healthy controls were recruited from September 2012 to June 2106. A TaqMan probe-based approach was used to genotype SNPs corresponding to rs28532698 and rs4301112 in CD157, rs855867 in AIM2, and rs2237126 in JARID2. Case-control and case-only studies were performed to determine the contribution of SNPs to the predisposition of disease and its severity, respectively. Our results revealed that the genotypes and allele frequencies of these SNPs were not significantly associated with childhood ASD and its severity in this population. Results of our study suggest that these SNPs are not predictors of childhood ASD in the Chinese Han population. The discrepant results suggest the predictor roles of SNPs have to be determined in different ethnic populations due to genetic heterogeneity of ASD. |
| JARID2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23324214  Title: 6p22.3 deletion: report of a patient with autism, severe intellectual disability and electroencephalographic anomalies.  The interstitial 6p deletions, involving the 6p22-p24 chromosomal region, are rare events characterized by variable phenotypes and no clear genotype-phenotype correlation has been established so far. High resolution array-CGH identified 1 Mb de novo interstitial deletion in 6p22.3 chromosomal region in a patient affected by severe Intellectual Disability (ID), Autism Spectrum Disorders (ASDs), and electroencephalographic anomalies. This deletion includes ATXN1, DTNBP1, JARID2 and MYLIP genes, known to play an important role in the brain, and the GMPR gene whose function in the nervous system is unknown. We support the suggestion that ATXN1, DTNBP1, JARID2 and MYLIP are candidate genes for the pathophysiology of ASDs and ID, and we propose that deletion of DTNBP1 and/or JARID2 contributes to the hypotonia phenotype. |
| KDM5A |
| Url: https://pubmed.ncbi.nlm.nih.gov/33350388  Title: KDM5A mutations identified in autism spectrum disorder using forward genetics.  Autism spectrum disorder (ASD) is a constellation of neurodevelopmental disorders with high phenotypic and genetic heterogeneity, complicating the discovery of causative genes. Through a forward genetics approach selecting for defective vocalization in mice, we identified Kdm5a as a candidate ASD gene. To validate our discovery, we generated a Kdm5a knockout mouse model (Kdm5a-/-) and confirmed that inactivating Kdm5a disrupts vocalization. In addition, Kdm5a-/- mice displayed repetitive behaviors, sociability deficits, cognitive dysfunction, and abnormal dendritic morphogenesis. Loss of KDM5A also resulted in dysregulation of the hippocampal transcriptome. To determine if KDM5A mutations cause ASD in humans, we screened whole exome sequencing and microarray data from a clinical cohort. We identified pathogenic KDM5A variants in nine patients with ASD and lack of speech. Our findings illustrate the power and efficacy of forward genetics in identifying ASD genes and highlight the importance of KDM5A in normal brain development and function. |
| KDM5A, KDM5B, KDM5C |
| Url: https://pubmed.ncbi.nlm.nih.gov/30902578  Title: Drosophila Histone Demethylase KDM5 Regulates Social Behavior through Immune Control and Gut Microbiota Maintenance.  Loss-of-function mutations in the histone demethylases KDM5A, KDM5B, or KDM5C are found in intellectual disability (ID) and autism spectrum disorders (ASD) patients. Here, we use the model organism Drosophila melanogaster to delineate how KDM5 contributes to ID and ASD. We show that reducing KDM5 causes intestinal barrier dysfunction and changes in social behavior that correlates with compositional changes in the gut microbiota. Therapeutic alteration of the dysbiotic microbiota through antibiotic administration or feeding with a probiotic Lactobacillus strain partially rescues the behavioral, lifespan, and cellular phenotypes observed in kdm5-deficient flies. Mechanistically, KDM5 was found to transcriptionally regulate component genes of the immune deficiency (IMD) signaling pathway and subsequent maintenance of host-commensal bacteria homeostasis in a demethylase-dependent manner. Together, our study uses a genetic approach to dissect the role of KDM5 in the gut-microbiome-brain axis and suggests that modifying the gut microbiome may provide therapeutic benefits for ID and ASD patients. |
| KDM5A, KDM5B, KDM5C, KMT2A, KMT2C, PHF21A |
| Url: https://pubmed.ncbi.nlm.nih.gov/26077434  Title: Disrupted intricacy of histone H3K4 methylation in neurodevelopmental disorders.  Methylation of histone H3 lysine 4 (H3K4me) is an intricately regulated posttranslational modification, which is broadly associated with enhancers and promoters of actively transcribed genomic loci. Recent advances in next-generation sequencing have identified a number of H3K4me regulators mutated in neurodevelopmental disorders including intellectual disabilities, autism spectrum disorders, and schizophrenia. Here, we aim to summarize the molecular function of H3K4me-regulating enzymes in brain development and function. We describe four H3K4me methyltransferases (KMT2A, KMT2C, KMT2D, KMT2F), four demethylases (KDM1A, KDM5A, KDM5B, KDM5C), and two reader proteins (PHF21A, PHF8) mutated in neurodevelopmental disorders. Understanding the role of these chromatin regulators in the development and maintenance of neural connections will advance therapeutic opportunities for prevention and treatment of these lifelong neurodevelopmental disorders. |
| KDM5C, KMT2A |
| Url: https://pubmed.ncbi.nlm.nih.gov/36831303  Title: Sexually Dimorphic Alterations in the Transcriptome and Behavior with Loss of Histone Demethylase KDM5C.  Chromatin dysregulation has emerged as a major hallmark of neurodevelopmental disorders such as intellectual disability (ID) and autism spectrum disorders (ASD). The prevalence of ID and ASD is higher in males compared to females, with unknown mechanisms. Intellectual developmental disorder, X-linked syndromic, Claes-Jensen type (MRXSCJ), is caused by loss-of-function mutations of lysine demethylase 5C (KDM5C), a histone H3K4 demethylase gene. KDM5C escapes X-inactivation, thereby presenting at a higher level in females. Initially, MRXSCJ was exclusively reported in males, while it is increasingly evident that females with heterozygous KDM5C mutations can show cognitive deficits. The mouse model of MRXSCJ, male Kdm5c-hemizygous knockout animals, recapitulates key features of human male patients. However, the behavioral and molecular traits of Kdm5c-heterozygous female mice remain incompletely characterized. Here, we report that gene expression and behavioral abnormalities are readily detectable in Kdm5c-heterozygous female mice, demonstrating the requirement for a higher KDM5C dose in females. Furthermore, we found both shared and sex-specific consequences of a reduced KDM5C dose in social behavior, gene expression, and genetic interaction with the counteracting enzyme KMT2A. These observations provide an essential insight into the sex-biased manifestation of neurodevelopmental disorders and sex chromosome evolution. |
| KDM5C |
| Url: https://pubmed.ncbi.nlm.nih.gov/31087518  Title: The potential role of a retrotransposed gene and a long noncoding RNA in regulating an X-linked chromatin gene (KDM5C): Novel epigenetic mechanism in autism.  A growing body of evidence supports the potential role of the circadian system and chromatin remodeling genes in autism. Considering the heterogeneity and gender discrepancy in autism, and the complex nature of the epigenetic landscape, identification of biologically relevant epigenetic factors requires reducing heterogeneity using proper subtyping. For this study, we used X chromosome inactivation (XCI) status in females with autism as an epigenetic marker for subtyping and examined the expression level of members of KDM5, a chromatin remodeling gene family. KDM5 are histone demethylases involved in the circadian molecular machinery. We used human blood samples to characterize alternatively spliced KDM5 isoforms and noticed that KDM5C undergoes a complex splicing process. We also identified a KDM5C isoform (KDM5C-3'UTR-lncRNA) containing a novel 3'UTR originated from a retrotransposed gene (retro-SUV39H2) of an autosomal methyltransferase (SUV39H2). This 3'UTR shows 84% sequence homology with long ncRNAs (lncRNAs) and is located 32 kb downstream of KDM5C. The KDM5C-3'UTR-lncRNA isoform was differentially expressed in autistic females with XCI skewness compared with controls. KDM5C plays a crucial role in balancing histone H3K4 methylation states. The identified retro-SUV39H2 originated lncRNA also shows H3K4 marks. By assessing the expression level of alternatively spliced Kdm5 isoforms at different circadian time-points, we showed that some isoforms follow a circadian oscillation pattern in wild type mouse brain.This study provides the first evidence and a suggestive model for the potential role of retrotransposed elements in autism through linking methylases and demethylases, two functionally complementary components of chromatin remodeling, which may collectively contribute to disease etiology through lncRNAs. Autism Res 2019, 12: 1007-1021. © 2019 International Society for Autism Research, Wiley Periodicals, Inc. LAY SUMMARY: Genes do not function in isolated conditions and their proper expression level also depends on a mechanism called gene regulation. An example of gene regulation is when changes outside DNA sequences influence the function of autism susceptibility genes. Alternative splicing is one type of gene regulation, which produces several versions of a gene (called variants) that may slightly differ from each other and be expressed at different levels in response to environmental changes. The circadian clock is an essential timing mechanism that enables organisms to maintain internal processes in sync with the dynamic environment brought about by the day-night cycle. The goal of this study was to assess if a subset of females with autism with certain genetic marker had a unique pattern of alternative splicing of three circadian genes. We identified a novel variant that is differentially expressed in this subset. Our study provides a novel subject stratification strategy, and a suggestive model of how biologically relevant components of a gene regulatory process may be linked and, possibly, collectively contribute to the etiology of autism. |
| KDM5C, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27799067  Title: Identification of a RAI1-associated disease network through integration of exome sequencing, transcriptomics, and 3D genomics.  Smith-Magenis syndrome (SMS) is a developmental disability/multiple congenital anomaly disorder resulting from haploinsufficiency of RAI1. It is characterized by distinctive facial features, brachydactyly, sleep disturbances, and stereotypic behaviors. We investigated a cohort of 15 individuals with a clinical suspicion of SMS who showed neither deletion in the SMS critical region nor damaging variants in RAI1 using whole exome sequencing. A combination of network analysis (co-expression and biomedical text mining), transcriptomics, and circularized chromatin conformation capture (4C-seq) was applied to verify whether modified genes are part of the same disease network as known SMS-causing genes. Potentially deleterious variants were identified in nine of these individuals using whole-exome sequencing. Eight of these changes affect KMT2D, ZEB2, MAP2K2, GLDC, CASK, MECP2, KDM5C, and POGZ, known to be associated with Kabuki syndrome 1, Mowat-Wilson syndrome, cardiofaciocutaneous syndrome, glycine encephalopathy, mental retardation and microcephaly with pontine and cerebellar hypoplasia, X-linked mental retardation 13, X-linked mental retardation Claes-Jensen type, and White-Sutton syndrome, respectively. The ninth individual carries a de novo variant in JAKMIP1, a regulator of neuronal translation that was recently found deleted in a patient with autism spectrum disorder. Analyses of co-expression and biomedical text mining suggest that these pathologies and SMS are part of the same disease network. Further support for this hypothesis was obtained from transcriptome profiling that showed that the expression levels of both Zeb2 and Map2k2 are perturbed in Rai1 -/- mice. As an orthogonal approach to potentially contributory disease gene variants, we used chromatin conformation capture to reveal chromatin contacts between RAI1 and the loci flanking ZEB2 and GLDC, as well as between RAI1 and human orthologs of the genes that show perturbed expression in our Rai1 -/- mouse model. These holistic studies of RAI1 and its interactions allow insights into SMS and other disorders associated with intellectual disability and behavioral abnormalities. Our findings support a pan-genomic approach to the molecular diagnosis of a distinctive disorder. |
| KDM5C |
| Url: https://pubmed.ncbi.nlm.nih.gov/27421841  Title: Patient Mutations of the Intellectual Disability Gene KDM5C Downregulate Netrin G2 and Suppress Neurite Growth in Neuro2a Cells.  The X-linked lysine (K)-specific demethylase 5C (KDM5C) gene plays an important role in brain development and behavior. It encodes a histone demethylase that is involved in gene regulation in neuronal differentiation and morphogenesis. When mutated, it causes neuropsychiatric symptoms, such as intellectual disability, delayed language development, epilepsy, and impulsivity. To better understand how the patient mutations affect neuronal development, we expressed KDM5C mutants in Neuro2a cells, a mouse neuroblastoma cell line. Retinoic acid (RA)-induced neurite growth was suppressed by the mutation KDM5C (Y751C) , KDM5C (H514A) , and KDM5C (F642L) , but not KDM5C (D87G) or KDM5C (A388P) . RNA-seq analysis indicated an upregulation of genes important for neuronal development, such as Ntng2, Enah, Gas1, Slit2, and Dscam, in response to the RA treatment in control Neuro2a cells transfected with GFP or wild-type KDM5C. In contrast, in cells transfected with KDM5C (Y751C) , these genes were not upregulated by RA. Ntng2 was downregulated in cells with KDM5C mutations, concordant with the lower levels of H3K4 methylation at its promoter. Moreover, knocking down Ntng2 in control Neuro2a cells led to the phenotype of short neurites similar to that of cells with KDM5C (Y751C) , whereas Ntng2 overexpression in the mutant cells rescued the morphological phenotype. These findings provide new insight into the pathogenesis of phenotypes associated with KDM5C mutations. |
| KLF16 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17480121  Title: Identification of the imprinted KLF14 transcription factor undergoing human-specific accelerated evolution.  Imprinted genes are expressed in a parent-of-origin manner and are located in clusters throughout the genome. Aberrations in the expression of imprinted genes on human Chromosome 7 have been suggested to play a role in the etiologies of Russell-Silver Syndrome and autism. We describe the imprinting of KLF14, an intronless member of the Krüppel-like family of transcription factors located at Chromosome 7q32. We show that it has monoallelic maternal expression in all embryonic and extra-embryonic tissues studied, in both human and mouse. We examine epigenetic modifications in the KLF14 CpG island in both species and find this region to be hypomethylated. In addition, we perform chromatin immunoprecipitation and find that the murine Klf14 CpG island lacks allele-specific histone modifications. Despite the absence of these defining features, our analysis of Klf14 in offspring from DNA methyltransferase 3a conditional knockout mice reveals that the gene's expression is dependent upon a maternally methylated region. Due to the intronless nature of Klf14 and its homology to Klf16, we suggest that the gene is an ancient retrotransposed copy of Klf16. By sequence analysis of numerous species, we place the timing of this event after the divergence of Marsupialia, yet prior to the divergence of the Xenarthra superclade. We identify a large number of sequence variants in KLF14 and, using several measures of diversity, we determine that there is greater variability in the human lineage with a significantly increased number of nonsynonymous changes, suggesting human-specific accelerated evolution. Thus, KLF14 may be the first example of an imprinted transcript undergoing accelerated evolution in the human lineage. |
| KLF7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36207723  Title: Krüppel-like factor 7 deficiency causes autistic-like behavior in mice via regulating Clock gene.  Krüppel-like factor 7 (klf7), a transcription factor in the nervous system to regulate cell proliferation and differentiation, has been recently identified as a causal gene for autism spectrum disorder (ASD), but the mechanism behind remains unknown. To uncover this mechanism, in this study we characterized the involvement of klf7 in circadian rhythm by knocking down klf7 in N2A cells and examining the rhythmic expression of circadian genes, especially Clock gene. We constructed klf7-/- mice and then investigated into klf7 regulation on the expression of rhythm genes in vivo as well as the use of melatonin to rescue the autism behavior. Our results illustrated that circadian rhythm was disrupted in klf7 knockdown cells and that klf7-/- mice showed autism-like behavior. Also, we found that Clock gene was downregulated in the brain of these klf7-/- mice and that the downstream rhythm genes of Clock were disturbed. Melatonin, as a circadian regulation drug, could regulate the expression level and amplitude of rhythm genes in klf7 knockout cells and further rescue the autistic behavior of klf7-/- mice. Klf7 deficiency causes ASD by disrupting circadian rhythm related genes to trigger rhythm oscillations. To treat ASD, maintaining circadian homeostasis is promising with the use of melatonin. |
| KLF7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35328799  Title: Krüppel-like Transcription Factor 7 Is a Causal Gene in Autism Development.  Autism spectrum disorder (ASD) is a complex neurodevelopmental disease. To date, more than 1000 genes have been shown to be associated with ASD, and only a few of these genes account for more than 1% of autism cases. Klf7 is an important transcription factor of cell proliferation and differentiation in the nervous system, but whether klf7 is involved in autism is unclear. We first performed ChIP-seq analysis of klf7 in N2A cells, then performed behavioral tests and RNA-seq in klf7+/- mice, and finally restored mice with adeno-associated virus (AAV)-mediated overexpression of klf7 in klf7+/- mice. Klf7 targeted genes are enriched with ASD genes, and 631 ASD risk genes are also differentially expressed in klf7+/- mice which exhibited the core symptoms of ASD. When klf7 levels were increased in the central nervous system (CNS) in klf7+/- adult mice, deficits in social interaction, repetitive behavior and majority of dysregulated ASD genes were rescued in the adults, suggesting transcriptional regulation. Moreover, knockdown of klf7 in human brain organoids caused dysregulation of 517 ASD risk genes, 344 of which were shared with klf7+/- mice, including some high-confidence ASD genes. Our findings highlight a klf7 regulation of ASD genes and provide new insights into the pathogenesis of ASD and promising targets for further research on mechanisms and treatments. |
| KMT2A |
| Url: https://pubmed.ncbi.nlm.nih.gov/35444258  Title: Dormant state of quiescent neural stem cells links Shank3 mutation to autism development.  Autism spectrum disorders (ASDs) are common neurodevelopmental disorders characterized by deficits in social interactions and communication, restricted interests, and repetitive behaviors. Despite extensive study, the molecular targets that control ASD development remain largely unclear. Here, we report that the dormancy of quiescent neural stem cells (qNSCs) is a therapeutic target for controlling the development of ASD phenotypes driven by Shank3 deficiency. Using single-cell RNA sequencing (scRNA-seq) and transposase accessible chromatin profiling (ATAC-seq), we find that abnormal epigenetic features including H3K4me3 accumulation due to up-regulation of Kmt2a levels lead to increased dormancy of qNSCs in the absence of Shank3. This result in decreased active neurogenesis in the Shank3 deficient mouse brain. Remarkably, pharmacological and molecular inhibition of qNSC dormancy restored adult neurogenesis and ameliorated the social deficits observed in Shank3-deficient mice. Moreover, we confirmed restored human qNSC activity rescues abnormal neurogenesis and autism-like phenotypes in SHANK3-targeted human NSCs. Taken together, our results offer a novel strategy to control qNSC activity as a potential therapeutic target for the development of autism. |
| KMT2A |
| Url: https://pubmed.ncbi.nlm.nih.gov/33387673  Title: Broad neurodevelopmental features and cortical anomalies associated with a novel de novo KMT2A variant in Wiedemann-Steiner syndrome.  Wiedemann-Steiner syndrome (WDSTS) is a rare genetic disorder including developmental delay/intellectual disability (DD/ID), hypertrichosis cubiti, short stature, and distinctive facial features, caused by mutation in KMT2A gene, which encodes a histone methyltransferase (H3K4) that regulates chromatin-mediated transcription. Different neurodevelopmental phenotypes have been described within the WDSTS spectrum, including a peculiar Autism Spectrum Disorder (ASDs) subtype in some affected individuals. Here, we report a 9-year-old Caucasian male found by next-generation panel sequencing to carry a novel heterozygous de novo KMT2A frameshift variant (NM\_001197104.2:c.4433delG; p. Arg1478LeufsTer108). This boy presented a WDSTS phenotype associated with broad neurodevelopmental features, including an unusual speech difficulty (i.e., palilalia), and brain imaging studies revealed an array of cortical anomalies (e.g., frontal simplified gyration, focal frontal cortical dysplasia). These clinical and radiological observations expand the known WDSTS-related neurodevelopmental phenotypes and further strengthen the important role of KMT2A in brain function and cortical development. |
| KMT2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/35324822  Title: De Novo Mutation in KMT2C Manifesting as Kleefstra Syndrome 2: Case Report and Literature Review.  Diagnosis of pediatric intellectual disability (ID) can be difficult because it is due to a vast number of established and novel causes. Here, we described a full-term female infant affected by Kleefstra syndrome-2 presenting with neurodevelopmental disorder, a history of hypotonia and minor face anomalies. A systematic literature review was also performed. The patient was a 6-year-old Caucasian female. In the family history there was no intellectual disability or genetic conditions. Auxological parameters at birth were adequate for gestational age. Clinical evaluation at 6 months revealed hypotonia and, successively, delay in the acquisition of the stages of psychomotor development. Auditory, visual, somatosensory, and motor-evoked potentials were normal. A brain MRI, performed at 9 months, showed minimal gliotic changes in bilateral occipital periventricular white matter. Neuropsychiatric control, performed at 5 years, established a definitive diagnosis of childhood autism and developmental delay. Molecular analysis of the exome revealed a novel KMT2C missense variant: c.9244C > T (p.Pro3082Ser) at a heterozygous state, giving her a diagnosis of Kleefstra syndrome 2. Parents did not show the variant. Literature review (four retrieved eligible studies, 10 patients) showed that all individuals had mild, moderate, or severe ID; language and motor delay; and autism. Short stature, microcephaly, childhood hypotonia and plagiocephaly were also present. Conclusion. Kleefstra syndrome 2 is a difficult diagnosis of a rare condition with a high clinical phenotypic heterogeneity. This study suggests that it must be taken in account in the work-up of an orphan diagnosis of intellectual disability and/or autism spectrum disorder. |
| KMT2C, ZNF292 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35190550  Title: Analysis of recent shared ancestry in a familial cohort identifies coding and noncoding autism spectrum disorder variants.  Autism spectrum disorder (ASD) is a collection of neurodevelopmental disorders characterized by deficits in social communication and restricted, repetitive patterns of behavior or interests. ASD is highly heritable, but genetically and phenotypically heterogeneous, reducing the power to identify causative genes. We performed whole genome sequencing (WGS) in an ASD cohort of 68 individuals from 22 families enriched for recent shared ancestry. We identified an average of 3.07 million variants per genome, of which an average of 112,512 were rare. We mapped runs of homozygosity (ROHs) in affected individuals and found an average genomic homozygosity of 9.65%, consistent with expectations for multiple generations of consanguineous unions. We identified potentially pathogenic rare exonic or splice site variants in 12 known (including KMT2C, SCN1A, SPTBN1, SYNE1, ZNF292) and 12 candidate (including CHD5, GRB10, PPP1R13B) ASD genes. Furthermore, we annotated noncoding variants in ROHs with brain-specific regulatory elements and identified putative disease-causing variants within brain-specific promoters and enhancers for 5 known ASD and neurodevelopmental disease genes (ACTG1, AUTS2, CTNND2, CNTNAP4, SPTBN4). We also identified copy number variants in two known ASD and neurodevelopmental disease loci in two affected individuals. In total we identified potentially etiological variants in known ASD or neurodevelopmental disease genes for ~61% (14/23) of affected individuals. We combined WGS with homozygosity mapping and regulatory element annotations to identify candidate ASD variants. Our analyses add to the growing number of ASD genes and variants and emphasize the importance of leveraging recent shared ancestry to map disease variants in complex neurodevelopmental disorders. |
| KMT2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/34945726  Title: Identification of a Rare Novel KMT2C Mutation That Presents with Schizophrenia in a Multiplex Family.  Schizophrenia is a complex genetic disorder involving many common variants with modest effects and rare mutations with high penetrance. Rare mutations associated with schizophrenia are highly heterogeneous and private for affected individuals and families. Identifying such mutations can help establish the molecular diagnosis, elucidate the pathogenesis, and provide helpful genetic counseling for affected patients and families. We performed a whole-exome sequencing analysis to search for rare pathogenic mutations co-segregating with schizophrenia transmitted in a dominant inheritance in a two-generation multiplex family. We identified a rare missense mutation H1574R (Histidine1574Arginine, rs199796552) of KMT2C (lysine methyltransferase 2C) co-segregating with affected members in this family. The mutation is a novel deleterious mutation of KMT2C, not reported before in the literature. The KMT2C encodes a histone 3 lysine 4 (H3K4)-specific methyltransferase and involves epigenetic regulation of brain gene expression. Mutations of KMT2C have been found in neurodevelopmental disorders, such as Kleefstra syndrome, intellectual disability, and autism spectrum disorders. Our finding suggests that schizophrenia might be one of the clinical phenotype spectra of KMT2C mutations, and KMT2C might be a novel risk gene for schizophrenia. Nevertheless, the co-segregation of this mutation with schizophrenia in this family might also be due to chance; functional assays of this mutation are needed to address this issue. |
| LMX1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/35417751  Title: Maternal Herpesviridae infection during pregnancy alters midbrain dopaminergic signatures in adult offspring.  Motor symptoms of Parkinson's disease (PD) are apparent after a high proportion of dopamine neurons in the substantia nigra have degenerated. The vast majority of PD cases are sporadic, and the underlying pathobiological causes are poorly understood. Adults exhibit great variability in the numbers of nigral dopamine neurons, suggesting that factors during embryonic or early life regulate the development and physiology of dopaminergic neurons. Furthermore, exposure to infections and inflammation in utero has been shown to affect fetal brain development in models of schizophrenia and autism. Here, we utilize a mouse maternal infection model to examine how maternal herpesvirus infection impacts dopaminergic neuron-related gene and protein expression in the adult offspring. Pregnant mice were injected with murine cytomegalovirus (MCMV), murine gamma herpes virus-68 (MHV68) or phosphate buffered saline (PBS) at embryonic day 8.5. Offspring were sacrificed at eight weeks of age and midbrains were processed for whole genome RNA sequencing, DNA methylation analysis, targeted protein expression and high-performance liquid chromatography for quantification of dopamine and its metabolites. The midbrain of adult offspring from MHV68 infected dams had significantly decreased expression of genes linked to dopamine neurons (Th, Lmx1b, and Foxa1) and increased Lrrk2, a gene involved in familial PD and PD risk that associates with neuroinflammation. Deconvolution analysis revealed that the proportion of dopamine neuron genes in the midbrain was reduced. There was an overall increase in DNA methylation in the midbrain of animals from MHV68-infected dams and pathway analyses indicated mitochondrial dysfunction, with reductions in genes associated with ATP synthesis, mitochondrial respiratory chain, and mitochondrial translation in the offspring of dams infected with MHV68. TIGAR (a negative regulator of mitophagy) and SDHA (mitochondrial complex II subunit) protein levels were increased, and the levels of 3,4-dihydroxyphenylacetic acid (DOPAC) in the striatum were increased in these offspring compared to offspring from uninfected control dams. No such changes were observed in the offspring of dams infected with MCMV. Our data suggest that maternal infection with Herpesviridae, specifically MHV68, can trigger changes in the development of the midbrain that impact dopamine neuron physiology in adulthood. Our work is of importance for the understanding of neuronal susceptibility underlying neurodegenerative disease, with particular relevance for PD. |
| LMX1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/26216300  Title: Direct conversion of human fibroblasts to induced serotonergic neurons.  Serotonergic (5HT) neurons exert diverse and widespread functions in the brain. Dysfunction of the serotonergic system gives rise to a variety of mental illnesses including depression, anxiety, obsessive compulsive disorder, autism and eating disorders. Here we show that human primary fibroblasts were directly converted to induced serotonergic (i5HT) neurons by the expression of Ascl1, Foxa2, Lmx1b and FEV. The transdifferentiation was enhanced by p53 knockdown and appropriate culture conditions including hypoxia. The i5HT neurons expressed markers for mature serotonergic neurons, had Ca(2+)-dependent 5HT release and selective 5HT uptake, exhibited spontaneous action potentials and spontaneous excitatory postsynaptic currents. Application of serotonin significantly increased the firing rate of spontaneous action potentials, demonstrating the functional utility of i5HT neurons for studying serotonergic neurotransmission. The availability of human i5HT neurons will be very useful for research and drug discovery on many serotonin-related mental disorders. |
| LMX1B |
| Url: https://pubmed.ncbi.nlm.nih.gov/21901133  Title: Association of transcription factor gene LMX1B with autism.  Multiple lines of evidence suggest a serotoninergic dysfunction in autism. The role of LMX1B in the development and maintenance of serotoninergic neurons is well known. In order to examine the role, if any, of LMX1B with autism pathophysiology, a trio-based SNP association study using 252 family samples from the AGRE was performed. Using pair-wise tagging method, 24 SNPs were selected from the HapMap data, based on their location and minor allele frequency. Two SNPs (rs10732392 and rs12336217) showed moderate association with autism with p values 0.018 and 0.022 respectively in transmission disequilibrium test. The haplotype AGCGTG also showed significant association (p = 0.008). Further, LMX1B mRNA expressions were studied in the postmortem brain tissues of autism subjects and healthy controls samples. LMX1B transcripts was found to be significantly lower in the anterior cingulate gyrus region of autism patients compared with controls (p = 0.049). Our study suggests a possible role of LMX1B in the pathophysiology of autism. Based on previous reports, it is likely to be mediated through a seretoninergic mechanism. This is the first report on the association of LMX1B with autism, though it should be viewed with some caution considering the modest associations we report. |
| MBD1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28100736  Title: Methyl-CpG-Binding Protein MBD1 Regulates Neuronal Lineage Commitment through Maintaining Adult Neural Stem Cell Identity.  Methyl-CpG-binding domain 1 (MBD1) belongs to a family of methyl-CpG-binding proteins that are epigenetic "readers" linking DNA methylation to transcriptional regulation. MBD1 is expressed in neural stem cells residing in the dentate gyrus of the adult hippocampus (aNSCs) and MBD1 deficiency leads to reduced neuronal differentiation, impaired neurogenesis, learning deficits, and autism-like behaviors in mice; however, the precise function of MBD1 in aNSCs remains unexplored. Here, we show that MBD1 is important for maintaining the integrity and stemness of NSCs, which is critical for their ability to generate neurons. MBD1 deficiency leads to the accumulation of undifferentiated NSCs and impaired transition into the neuronal lineage. Transcriptome analysis of neural stem and progenitor cells isolated directly from the dentate gyrus of MBD1 mutant (KO) and WT mice showed that gene sets related to cell differentiation, particularly astrocyte lineage genes, were upregulated in KO cells. We further demonstrated that, in NSCs, MBD1 binds and represses directly specific genes associated with differentiation. Our results suggest that MBD1 maintains the multipotency of NSCs by restraining the onset of differentiation genes and that untimely expression of these genes in MBD1-deficient stem cells may interfere with normal cell lineage commitment and cause the accumulation of undifferentiated cells. Our data reveal a novel role for MBD1 in stem cell maintenance and provide insight into how epigenetic regulation contributes to adult neurogenesis and the potential impact of its dysregulation. Adult neural stem cells (aNSCs) in the hippocampus self-renew and generate neurons throughout life. We show that methyl-CpG-binding domain 1 (MBD1), a DNA methylation "reader," is important for maintaining the integrity of NSCs, which is critical for their neurogenic potency. Our data reveal a novel role for MBD1 in stem cell maintenance and provide insight into how epigenetic regulation preserves the multipotency of stem cells for subsequent differentiation. |
| MBD1, MBD4, MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19000991  Title: Reciprocal co-regulation of EGR2 and MECP2 is disrupted in Rett syndrome and autism.  Mutations in MECP2, encoding methyl-CpG-binding protein 2 (MeCP2), cause the neurodevelopmental disorder Rett syndrome (RTT). Although MECP2 mutations are rare in idiopathic autism, reduced MeCP2 levels are common in autism cortex. MeCP2 is critical for postnatal neuronal maturation and a modulator of activity-dependent genes such as Bdnf (brain-derived neurotropic factor) and JUNB. The activity-dependent early growth response gene 2 (EGR2), required for both early hindbrain development and mature neuronal function, has predicted binding sites in the promoters of several neurologically relevant genes including MECP2. Conversely, MeCP2 family members MBD1, MBD2 and MBD4 bind a methylated CpG island in an enhancer region located in EGR2 intron 1. This study was designed to test the hypothesis that MECP2 and EGR2 regulate each other's expression during neuronal maturation in postnatal brain development. Chromatin immunoprecipitation analysis showed EGR2 binding to the MECP2 promoter and MeCP2 binding to the enhancer region in EGR2 intron 1. Reduction in EGR2 and MeCP2 levels in cultured human neuroblastoma cells by RNA interference reciprocally reduced expression of both EGR2 and MECP2 and their protein products. Consistent with a role of MeCP2 in enhancing EGR2, Mecp2-deficient mouse cortex samples showed significantly reduced EGR2 by quantitative immunofluorescence. Furthermore, MeCP2 and EGR2 show coordinately increased levels during postnatal development of both mouse and human cortex. In contrast to age-matched Controls, RTT and autism postmortem cortex samples showed significant reduction in EGR2. Together, these data support a role of dysregulation of an activity-dependent EGR2/MeCP2 pathway in RTT and autism. |
| MBD1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18385101  Title: The loss of methyl-CpG binding protein 1 leads to autism-like behavioral deficits.  Methyl-CpG binding proteins (MBDs) are central components of DNA methylation-mediated epigenetic gene regulation. Alterations of epigenetic pathways are known to be associated with several neurodevelopmental disorders, particularly autism. Our previous studies showed that the loss of Mbd1 led to reduced hippocampal neurogenesis and impaired learning in mice. However, whether MBD1 regulates the autism-related cognitive functions remains unknown. Here we show that Mbd1 mutant (Mbd1(-/-)) mice exhibit several core deficits frequently associated with autism, including reduced social interaction, learning deficits, anxiety, defective sensory motor gating, depression and abnormal brain serotonin activity. Furthermore, we find that Mbd1 can directly regulate the expression of Htr2c, one of the serotonin receptors, by binding to its promoter, and the loss of Mbd1 led to elevated expression of Htr2c. Our results, therefore, demonstrate the importance of epigenetic regulation in mammalian brain development and cognitive functions. Understanding how the loss of Mbd1 could lead to autism-like behavioral phenotypes would reveal much-needed information about the molecular pathogenesis of autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36724557  Title: Human MECP2 transgenic rats show increased anxiety, severe social deficits, and abnormal prefrontal neural oscillation stability.  Methylated CpG binding protein 2 (MeCP2) plays an important role in the development and normal function of the neural system. Abnormally high expression of MECP2 leads to a subtype of autism called MECP2 duplication syndrome and MECP2 is considered one of the key pathogenic genes for autism spectrum disorders. However, the effect of MECP2 overexpression on neural activity is still not fully understood. Thus, transgenic (TG) animals that abnormally overexpress MeCP2 are important disease models in research on neurological function and autism. To create an animal model with a stronger and more stable autism phenotype, this study established a human MECP2 TG rat model and evaluated its movement ability, anxiety, and social behavior through behavioral tests. The results showed that MECP2 TG rats had an abnormally increased anxiety phenotype and social deficits in terms of abnormal social approach and social novelty preference, but no movement disorder. These autism-like behavioral phenotypes suggest that human MECP2 TG rats are suitable models for studying autism as they show more severe social deficit phenotypes and without interference from movement disorders affecting other phenotypes, which is an issue for mouse models with MECP2 duplication. In addition, this study performed preliminary exploration of the influence of the human MECP2 transgene on neural oscillation stability of the medial prefrontal cortex (mPFC), which is an important brain region for social interactions. Oscillation stability in MECP2 TG rats showed abnormal responses to social conditions. Overall, the results of this study provide a new research tool for understanding the mechanism of social impairment and treatment of autism. The results also provide evidence for the influence of MECP2 duplication on mPFC neural activity. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36441779  Title: Linking neuroanatomical abnormalities in autism spectrum disorder with gene expression of candidate ASD genes: A meta-analytic and network-oriented approach.  Autism spectrum disorder (ASD) is a set of developmental conditions with widespread neuroanatomical abnormalities and a strong genetic basis. Although neuroimaging studies have indicated anatomical changes in grey matter (GM) morphometry, their associations with gene expression remain elusive. Here, we aim to understand how gene expression correlates with neuroanatomical atypicalities in ASD. To do so, we performed a coordinate-based meta-analysis to determine the common GM variation pattern in the autistic brain. From the Allen Human Brain Atlas, we selected eight genes from the SHANK, NRXN, NLGN family and MECP2, which have been implicated with ASD, particularly in regards to altered synaptic transmission and plasticity. The gene expression maps for each gene were built. We then assessed the correlation between the gene expression maps and the GM alteration maps. Lastly, we projected the obtained clusters of GM alteration-gene correlations on top of the canonical resting state networks, in order to provide a functional characterization of the structural evidence. We found that gene expression of most genes correlated with GM alteration (both increase and decrease) in regions located in the default mode network. Decreased GM was also correlated with gene expression of some ASD genes in areas associated with the dorsal attention and cerebellar network. Lastly, single genes were found to be significantly correlated with increased GM in areas located in the somatomotor, limbic and ganglia/thalamus networks. This approach allowed us to combine the well beaten path of genetic and brain imaging in a novel way, to specifically investigate the relation between gene expression and brain with structural damage, and individuate genes of potential interest for further investigation in the functional domain. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36156984  Title: Brain structural alterations in young girls with Rett syndrome: A voxel-based morphometry and tract-based spatial statistics study.  Rett syndrome (RTT) is a neurodevelopmental disorder caused by loss-of-function variants in the MECP2 gene, currently with no cure. Neuroimaging is an important tool for obtaining non-invasive structural and functional information about the in vivo brain. Multiple approaches to magnetic resonance imaging (MRI) scans have been utilized effectively in RTT patients to understand the possible pathological basis. This study combined developmental evaluations with clinical severity, T1-weighted imaging, and diffusion tensor imaging, aiming to explore the structural alterations in cohorts of young girls with RTT, idiopathic autism spectrum disorder (ASD), or typical development. Voxel-based morphometry (VBM) was used to determine the voxel-wised volumetric characteristics of gray matter, while tract-based spatial statistics (SPSS) was used to obtain voxel-wised properties of white matter. Finally, a correlation analysis between the brain structural alterations and the clinical evaluations was performed. In the RTT group, VBM revealed decreased gray matter volume in the insula, frontal cortex, calcarine, and limbic/paralimbic regions; TBSS demonstrated decreased fractional anisotropy (FA) and increased mean diffusivity (MD) mainly in the corpus callosum and other projection and association fibers such as superior longitudinal fasciculus and corona radiata. The social impairment quotient and clinical severity were associated with these morphometric alterations. This monogenic study with an early stage of RTT may provide some valuable guidance for understanding the disease pathogenesis. At the same time, the pediatric-adjusted analytic pipelines for VBM and TBSS were introduced for significant improvement over classical approaches for MRI scans in children. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36142719  Title: Epilepsy Characteristics in Neurodevelopmental Disorders: Research from Patient Cohorts and Animal Models Focusing on Autism Spectrum Disorder.  Epilepsy, a heterogeneous group of brain-related diseases, has continued to significantly burden society and families. Epilepsy comorbid with neurodevelopmental disorders (NDDs) is believed to occur due to multifaceted pathophysiological mechanisms involving disruptions in the excitation and inhibition (E/I) balance impeding widespread functional neuronal circuitry. Although the field has received much attention from the scientific community recently, the research has not yet translated into actionable therapeutics to completely cure epilepsy, particularly those comorbid with NDDs. In this review, we sought to elucidate the basic causes underlying epilepsy as well as those contributing to the association of epilepsy with NDDs. Comprehensive emphasis is put on some key neurodevelopmental genes implicated in epilepsy, such as MeCP2, SYNGAP1, FMR1, SHANK1-3 and TSC1, along with a few others, and the main electrophysiological and behavioral deficits are highlighted. For these genes, the progress made in developing appropriate and valid rodent models to accelerate basic research is also detailed. Further, we discuss the recent development in the therapeutic management of epilepsy and provide a briefing on the challenges and caveats in identifying and testing species-specific epilepsy models. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36063192  Title: A perspective on molecular signalling dysfunction, its clinical relevance and therapeutics in autism spectrum disorder.  Intellectual disability (ID) and autism spectrum disorder (ASD) are neurodevelopmental disorders that have become a primary clinical and social concern, with a prevalence of 2-3% in the population. Neuronal function and behaviour undergo significant malleability during the critical period of development that is found to be impaired in ID/ASD. Human genome sequencing studies have revealed many genetic variations associated with ASD/ID that are further verified by many approaches, including many mouse and other models. These models have facilitated the identification of fundamental mechanisms underlying the pathogenesis of ASD/ID, and several studies have proposed converging molecular pathways in ASD/ID. However, linking the mechanisms of the pathogenic genes and their molecular characteristics that lead to ID/ASD has progressed slowly, hampering the development of potential therapeutic strategies. This review discusses the possibility of recognising the common molecular causes for most ASD/ID based on studies from the available models that may enable a better therapeutic strategy to treat ID/ASD. We also reviewed the potential biomarkers to detect ASD/ID at early stages that may aid in diagnosis and initiating medical treatment, the concerns with drug failure in clinical trials, and developing therapeutic strategies that can be applied beyond a particular mutation associated with ASD/ID. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35985556  Title: Aberrant brain functional and structural developments in MECP2 duplication rats.  Transgenic animal models with homologous etiology provide a promising way to pursue the neurobiological substrates of the behavioral deficits in autism spectrum disorder (ASD). Gain-of-function mutations of MECP2 cause MECP2 duplication syndrome, a severe neurological disorder with core symptoms of ASD. However, abnormal brain developments underlying the autistic-like behavioral deficits of MECP2 duplication syndrome are rarely investigated. To this end, a human MECP2 duplication (MECP2-DP) rat model was created by the bacterial artificial chromosome transgenic method. Functional and structural magnetic resonance imaging (MRI) with high-field were performed on 16 male MECP2-DP rats and 15 male wildtype rats at postnatal 28 days, 42 days, and 56 days old. Multimodal fusion analyses guided by locomotor-relevant metrics and social novelty time separately were applied to identify abnormal brain networks associated with diverse behavioral deficits induced by MECP2 duplication. Aberrant functional developments of a core network primarily composed of the dorsal medial prefrontal cortex (dmPFC) and retrosplenial cortex (RSP) were detected to associate with diverse behavioral phenotypes in MECP2-DP rats. Altered developments of gray matter volume were detected in the hippocampus and thalamus. We conclude that gain-of-function mutations of MECP2 induce aberrant functional activities in the default-mode-like network and aberrant volumetric changes in the brain, resulting in autistic-like behavioral deficits. Our results gain critical insights into the biomarker of MECP2 duplication syndrome and the neurobiological underpinnings of the behavioral deficits in ASD. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35883206  Title: Behavioral manifestations in rodent models of autism spectrum disorder: protocol for a systematic review and network meta-analysis.  Autism spectrum disorder (ASD) is a neurodevelopmental condition associated with severe social communication, interaction, and sensory processing impairments. Efforts to understand its etiology and pathophysiology are crucial for improving treatment and prevention measures. Preclinical models of ASD are essential for investigating the biological mechanisms and should present translatability potential. We aim to evaluate the consistency of the most commonly used rodent models of ASD in displaying autistic-like behavior through a systematic review and meta-analysis. This review will focus on the most frequently used autism models, surveying studies of six genetic (Ube3a, Pten, Nlgn3, Shank3, Mecp2, and Fmr1), three chemically induced (valproic acid (VPA), lipopolysaccharide (LPS), and polyinosinic:polycytidylic acid (poly(I:C))), and one inbred model (BTBR T+ Itpr3tf/J mouse strain). Two independent reviewers will screen the records. Data extraction of behavioral outcomes and risk of bias evaluation will be performed. We will conduct a meta-analysis whenever at least five studies investigate the same model and behavioral outcome. We will also explore the heterogeneity and publication bias. Network meta-analyses are planned to compare different models. By shortening the gap between animal behavior and human endophenotypes or specific clinical symptoms, we expect to help researchers on which rodent models are adequate for research of specific behavioral manifestations of autism, which potentially require a combination of them depending on the research interest. PROSPERO CRD42021226299 . |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35831173  Title: Increased Reliability of Visually-Evoked Activity in Area V1 of the MECP2-Duplication Mouse Model of Autism.  Atypical sensory processing is now thought to be a core feature of the autism spectrum. Influential theories have proposed that both increased and decreased neural response reliability within sensory systems could underlie altered sensory processing in autism. Here, we report evidence for abnormally increased reliability of visual-evoked responses in layer 2/3 neurons of adult male and female primary visual cortex in the MECP2-duplication syndrome animal model of autism. Increased response reliability was due in part to decreased response amplitude, decreased fluctuations in endogenous activity, and an abnormal decoupling of visual-evoked activity from endogenous activity. Similar to what was observed neuronally, the optokinetic reflex occurred more reliably at low contrasts in mutant mice compared with controls. Retinal responses did not explain our observations. These data suggest that the circuit mechanisms for combining sensory-evoked and endogenous signal and noise processes may be altered in this form of syndromic autism.SIGNIFICANCE STATEMENT Atypical sensory processing is now thought to be a core feature of the autism spectrum. Influential theories have proposed that both increased and decreased neural response reliability within sensory systems could underlie altered sensory processing in autism. Here, we report evidence for abnormally increased reliability of visual-evoked responses in primary visual cortex of the animal model for MECP2-duplication syndrome, a high-penetrance single-gene cause of autism. Visual-evoked activity was abnormally decoupled from endogenous activity in mutant mice, suggesting in line with the influential "hypo-priors" theory of autism that sensory priors embedded in endogenous activity may have less influence on perception in autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35460749  Title: TAT-MeCP2 protein variants rescue disease phenotypes in human and mouse models of Rett syndrome.  Rett syndrome (RTT) is a neurodevelopmental disorder caused by pathogenic variants leading to functional impairment of the MeCP2 protein. Here, we used purified recombinant MeCP2e1 and MeCP2e2 protein variants fused to a TAT protein transduction domain (PTD) to evaluate their transduction ability into RTT patient-derived fibroblasts and the ability to carry out their cellular function. We then assessed their transduction ability and therapeutic effects in a RTT mouse model. In vitro, TAT-MeCP2e2-eGFP reversed the pathological hyperacetylation of histones H3K9 and H4K16, a hallmark of abolition of MeCP2 function. In vivo, intraperitoneal administration of TAT-MeCP2e1 and TAT-MeCP2e2 extended the lifespan of Mecp2-/y mice by >50%. This was accompanied by rescue of hippocampal CA2 neuron size in animals treated with TAT-MeCP2e1. Taken together, these findings provide a strong indication that recombinant TAT-MeCP2 can reach mouse brains following peripheral injection and can ameliorate the phenotype of RTT mouse models. Thus, our study serves as a first step in the development of a potentially novel RTT therapy. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35456477  Title: Electrophysiological and Behavioral Evidence for Hyper- and Hyposensitivity in Rare Genetic Syndromes Associated with Autism.  Our study reviewed abnormalities in spontaneous, as well as event-related, brain activity in syndromes with a known genetic underpinning that are associated with autistic symptomatology. Based on behavioral and neurophysiological evidence, we tentatively subdivided the syndromes on primarily hyper-sensitive (Fragile X, Angelman) and hypo-sensitive (Phelan-McDermid, Rett, Tuberous Sclerosis, Neurofibromatosis 1), pointing to the way of segregation of heterogeneous idiopathic ASD, that includes both hyper-sensitive and hypo-sensitive individuals. This segmentation links abnormalities in different genes, such as FMR1, UBE3A, GABRB3, GABRA5, GABRG3, SHANK3, MECP2, TSC1, TSC2, and NF1, that are causative to the above-mentioned syndromes and associated with synaptic transmission and cell growth, as well as with translational and transcriptional regulation and with sensory sensitivity. Excitation/inhibition imbalance related to GABAergic signaling, and the interplay of tonic and phasic inhibition in different brain regions might underlie this relationship. However, more research is needed. As most genetic syndromes are very rare, future investigations in this field will benefit from multi-site collaboration with a common protocol for electrophysiological and event-related potential (EEG/ERP) research that should include an investigation into all modalities and stages of sensory processing, as well as potential biomarkers of GABAergic signaling (such as 40-Hz ASSR). |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35410641  Title: Autistic-like social deficits in hippocampal MeCP2 knockdown rat models are rescued by ketamine.  Autism or autism spectrum disorder (ASD) is a behavioral syndrome characterized by persistent deficits in social interaction, and repetitive patterns of behavior, interests, or activities. The gene encoding Methyl-CpG binding protein 2 (MeCP2) is one of a few exceptional genes of established causal effect in ASD. Although genetically engineered mice studies may shed light on how MeCP2 loss affects synaptic activity patterns across the whole brain, such studies are not considered practical in ASD patients due to the overall level of impairment, and are technically challenging in mice. For the first time, we show that hippocampal MeCP2 knockdown produces behavioral abnormalities associated with autism-like traits in rats, providing a new strategy to investigate the efficacy of therapeutics in ASD. Ketamine, an N-Methyl-D-aspartate (NMDA) blocker, has been proposed as a possible treatment for autism. Using the MeCP2 knockdown rats in conjunction with a rat model of valproic acid (VPA)-induced ASD, we examined gene expression and ASD behaviors upon ketamine treatment. We report that the core symptoms of autism in MeCP2 knockdown rats with social impairment recovered dramatically following a single treatment with ketamine. [BMB Reports 2022; 55(5): 238-243]. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35365601  Title: Loss of neurodevelopmental-associated miR-592 impairs neurogenesis and causes social interaction deficits.  microRNA-592 (miR-592) has been linked to neurogenesis, but the influence of miR-592 knockout in vivo remains unknown. Here, we report that miR-592 knockout represses IPC-to-mature neuron transition, impairs motor coordination and reduces social interaction. Combining the RNA-seq and tandem mass tagging-based quantitative proteomics analysis (TMT protein quantification) and luciferase reporter assays, we identified MeCP2 as the direct targetgene of miR-592 in the mouse cortex. In Tg(MECP2) mice, lipofection of miR-592 efficiently reduced MECP2 expression in the brains of Tg(MECP2) mice at E14.5. Furthermore, treatment with miR-592 partially ameliorated the autism-like phenotypes observed in adult Tg(MECP2) mice. The findings demonstrate that miR-592 might play a novel role in treating the neurodevelopmental-associated disorder. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35235788  Title: Placenta and fetal brain share a neurodevelopmental disorder DNA methylation profile in a mouse model of prenatal PCB exposure.  Polychlorinated biphenyls (PCBs) are developmental neurotoxicants implicated as environmental risk factors for neurodevelopmental disorders (NDDs). Here, we report the effects of prenatal exposure to a human-relevant mixture of PCBs on the DNA methylation profiles of mouse placenta and fetal brain. Thousands of differentially methylated regions (DMRs) distinguish placenta and fetal brain from PCB-exposed mice from sex-matched vehicle controls. In both placenta and fetal brain, PCB-associated DMRs are enriched for functions related to neurodevelopment and cellular signaling and enriched within regions of bivalent chromatin. The placenta and brain PCB DMRs overlap significantly and map to a shared subset of genes enriched for Wnt signaling, Slit/Robo signaling, and genes differentially expressed in NDD models. The consensus PCB DMRs also significantly overlap with DMRs from human NDD brain and placenta. These results demonstrate that PCB-exposed placenta contains a subset of DMRs that overlap fetal brain DMRs relevant to an NDD. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35147909  Title: Astrocytic Gap Junctions Contribute to Aberrant Neuronal Synchronization in a Mouse Model of MeCP2 Duplication Syndrome.  Abnormal synchronous neuronal activity has been widely detected by brain imaging of autistic patients, but its underlying neural mechanism remains unclear. Compared with wild-type mice, our in vivo two-photon imaging showed that transgenic (Tg1) mice over-expressing human autism risk gene MeCP2 exhibited higher neuronal synchrony in the young but lower synchrony in the adult stage. Whole-cell recording of neuronal pairs in brain slices revealed that higher neuronal synchrony in young postnatal Tg1 mice was attributed mainly to more prevalent giant slow inward currents (SICs). Both in vivo and slice imaging further demonstrated more dynamic activity and higher synchrony in astrocytes from young Tg1 mice. Blocking astrocytic gap junctions markedly decreased the generation of SICs and overall cell synchrony in the Tg1 brain. Furthermore, the expression level of Cx43 protein and the coupling efficiency of astrocyte gap junctions remained unchanged in Tg1 mice. Thus, astrocytic gap junctions facilitate but do not act as a direct trigger for the abnormal neuronal synchrony in young Tg1 mice, revealing the potential role of the astrocyte network in the pathogenesis of MeCP2 duplication syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35074918  Title: Disruption of MeCP2-TCF20 complex underlies distinct neurodevelopmental disorders.  MeCP2 is associated with Rett syndrome (RTT), MECP2 duplication syndrome, and a number of conditions with isolated features of these diseases, including autism, intellectual disability, and motor dysfunction. MeCP2 is known to broadly bind methylated DNA, but the precise molecular mechanism driving disease pathogenesis remains to be determined. Using proximity-dependent biotinylation (BioID), we identified a transcription factor 20 (TCF20) complex that interacts with MeCP2 at the chromatin interface. Importantly, RTT-causing mutations in MECP2 disrupt this interaction. TCF20 and MeCP2 are highly coexpressed in neurons and coregulate the expression of key neuronal genes. Reducing Tcf20 partially rescued the behavioral deficits caused by MECP2 overexpression, demonstrating a functional relationship between MeCP2 and TCF20 in MECP2 duplication syndrome pathogenesis. We identified a patient exhibiting RTT-like neurological features with a missense mutation in the PHF14 subunit of the TCF20 complex that abolishes the MeCP2-PHF14-TCF20 interaction. Our data demonstrate the critical role of the MeCP2-TCF20 complex for brain function. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34840252  Title: Challenging Case: The Role of Genetic Testing in Complex Autism.  S is a 12-year-old boy with autism spectrum disorder (ASD), seizure disorder, cerebral palsy, and intellectual disability who presented to the primary care clinician for a preventative care visit.S was born at full term after an unremarkable pregnancy. His developmental delays were first noted at around 8 months, when he could not sit independently and had intermittently poor eye contact. He was referred to Part C Early Intervention and subsequently evaluated by a neurodevelopmental pediatrician, where he was noted to be hypotonic, with delayed motor and cognitive skills. Initial genetics evaluation included karyotype, fragile X testing, Angelman and Prader-Willi DNA fluorescence in situ hybridization probes, POLG sequencing, MECP2 testing, a microarray, creatinine kinase, very long-chain fatty acids, lymphocyte arylsulfatase, urine organic acids, and plasma amino acids, all of which were normal.As time progressed, S continued to have motor and communication delays and developed choreic movements. He also developed episodes concerning for seizure, including periods of staring while awake and episodes of extremity shaking lasting a few seconds with associated eye deviation, which eventually progressed to generalized seizures. He also developed periods of lethargy. Outpatient workup included several EEGs, which were notable for foci in the right frontal and left temporal regions. He has had several brain MRIs showing generalized volume loss and had critical laboratory tests during a period of lethargy, which were unconcerning. He was treated with multiple antiseizure medications. He was diagnosed with ASD at age 5 years because of delayed language, poor social communication, and repetitive behaviors.Over time, S continued to experience global developmental delays and autistic-like behaviors and remained minimally verbal. However, clinicians noted a number of developmental strengths, including a generally positive mood, a willingness to participate in therapy, improved receptive language skills, attachment to his mother, and a love of nature and the outdoors. He participated in a number of therapy modalities including speech/language therapy, occupational therapy, physical therapy, applied behavioral analysis, aqua therapy, partner-assisted scanning, and therapeutic horseback riding.In 2019, whole-exome sequencing was newly covered by the state Medicaid program, and testing was obtained in 2020. Whole-exome sequencing revealed a de novo STXBP1 pathogenic variant c.874C>T (p.Arg292Cys), which is associated with developmental and epileptic encephalopathy. His presentation is consistent with STXBP1 encephalopathy including refractory epilepsy, ASD, intellectual disability, and movement disorders.What are important considerations in genetic testing for children with autism? How does a genetic testing result alter management for clinicians and families? |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34485605  Title: Gene therapy using an ortholog of human fragile X mental retardation protein partially rescues behavioral abnormalities and EEG activity.  Fragile X syndrome (FXS), a neurodevelopmental disorder with no known cure, is caused by a lack of expression of the fragile X mental retardation protein (FMRP). As a single-gene disorder, FXS is an excellent candidate for viral-vector-based gene therapy, although that is complicated by the existence of multiple isoforms of FMRP, whose individual cellular functions are unknown. We studied the effects of rat and mouse orthologs of human isoform 17, a major expressed isoform of FMRP. Injection of neonatal Fmr1 knockout rats and mice with adeno-associated viral vectors (AAV9 serotype) under the control of an MeCP2 mini-promoter resulted in widespread distribution of the FMRP transgenes throughout the telencephalon and diencephalon. Transgene expression occurred mainly in non-GABAergic neurons, with little expression in glia. Early postnatal treatment resulted in partial rescue of the Fmr1 KO rat phenotype, including improved social dominance in treated Fmr1 KO females and partial rescue of locomotor activity in males. Electro-encephalogram (EEG) recordings showed correction of abnormal slow-wave activity during the sleep-like state in male Fmr1 KO rats. These findings support the use of AAV-based gene therapy as a treatment for FXS and specifically demonstrate the potential therapeutic benefit of human FMRP isoform 17 orthologs. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34021030  Title: Inhibition of Elevated Ras-MAPK Signaling Normalizes Enhanced Motor Learning and Excessive Clustered Dendritic Spine Stabilization in the MECP2-Duplication Syndrome Mouse Model of Autism.  The inflexible repetitive behaviors and "insistence on sameness" seen in autism imply a defect in neural processes controlling the balance between stability and plasticity of synaptic connections in the brain. It has been proposed that abnormalities in the Ras-ERK/MAPK pathway, a key plasticity-related cell signaling pathway known to drive consolidation of clustered synaptic connections, underlie altered learning phenotypes in autism. However, a link between altered Ras-ERK signaling and clustered dendritic spine plasticity has yet to be explored in an autism animal model in vivo The formation and stabilization of dendritic spine clusters is abnormally increased in the MECP2-duplication syndrome mouse model of syndromic autism, suggesting that ERK signaling may be increased. Here, we show that the Ras-ERK pathway is indeed hyperactive following motor training in MECP2-duplication mouse motor cortex. Pharmacological inhibition of ERK signaling normalizes the excessive clustered spine stabilization and enhanced motor learning behavior in MECP2-duplication mice. We conclude that hyperactive ERK signaling may contribute to abnormal clustered dendritic spine consolidation and motor learning in this model of syndromic autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36654241  Title: Induction of core symptoms of autism spectrum disorder by in vivo CRISPR/Cas9-based gene editing in the brain of adolescent rhesus monkeys.  Although CRISPR/Cas9-mediated gene editing is widely applied to mimic human disorders, whether acute manipulation of disease-causing genes in the brain leads to behavioral abnormalities in non-human primates remains to be determined. Here we induced genetic mutations in MECP2, a critical gene linked to Rett syndrome (RTT) and autism spectrum disorders (ASD), in the hippocampus (DG and CA1-4) of adolescent rhesus monkeys (Macaca mulatta) in vivo via adeno-associated virus (AAV)-delivered Staphylococcus aureus Cas9 with small guide RNAs (sgRNAs) targeting MECP2. In comparison to monkeys injected with AAV-SaCas9 alone (n = 4), numerous autistic-like behavioral abnormalities were identified in the AAV-SaCas9-sgMECP2-injected monkeys (n = 7), including social interaction deficits, abnormal sleep patterns, insensitivity to aversive stimuli, abnormal hand motions, and defective social reward behaviors. Furthermore, some aspects of ASD and RTT, such as stereotypic behaviors, did not appear in the MECP2 gene-edited monkeys, suggesting that different brain areas likely contribute to distinct ASD symptoms. This study showed that acute manipulation of disease-causing genes via in vivo gene editing directly led to behavioral changes in adolescent primates, paving the way for the rapid generation of genetically engineered non-human primate models for neurobiological studies and therapeutic development. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33373327  Title: The DNA repair protein ATM as a target in autism spectrum disorder.  Impairment of the GABAergic system has been reported in epilepsy, autism, attention deficit hyperactivity disorder, and schizophrenia. We recently demonstrated that ataxia telangiectasia mutated (ATM) directly shapes the development of the GABAergic system. Here, we show for the first time to our knowledge how the abnormal expression of ATM affects the pathological condition of autism. We exploited 2 different animal models of autism, the methyl CpG binding protein 2-null (Mecp2y/-) mouse model of Rett syndrome and mice prenatally exposed to valproic acid, and found increased ATM levels. Accordingly, treatment with the specific ATM kinase inhibitor KU55933 (KU) normalized molecular, functional, and behavioral defects in these mouse models, such as (a) delayed GABAergic development, (b) hippocampal hyperexcitability, (c) low cognitive performances, and (d) social impairments. Mechanistically, we demonstrate that KU administration to WT hippocampal neurons leads to (a) higher early growth response 4 activity on Kcc2b promoter, (b) increased expression of Mecp2, and (c) potentiated GABA transmission. These results provide evidence and molecular substrates for the pharmacological development of ATM inhibition in autism spectrum disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33168618  Title: Excessive Formation and Stabilization of Dendritic Spine Clusters in the MECP2-Duplication Syndrome Mouse Model of Autism.  Autism-associated genetic mutations may perturb the balance between stability and plasticity of synaptic connections in the brain. Here, we report an increase in the formation and stabilization of dendritic spines in the cerebral cortex of the mouse model of MECP2-duplication syndrome, a high-penetrance form of syndromic autism. Increased stabilization is mediated entirely by spines that form cooperatively in 10-μm clusters and is observable across multiple cortical areas both spontaneously and following motor training. Excessive stability of dendritic spine clusters could contribute to behavioral rigidity and other phenotypes in syndromic autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33084871  Title: Interregulation between fragile X mental retardation protein and methyl CpG binding protein 2 in the mouse posterior cerebral cortex.  Several X-linked neurodevelopmental disorders including Rett syndrome, induced by mutations in the MECP2 gene, and fragile X syndrome (FXS), caused by mutations in the FMR1 gene, share autism-related features. The mRNA coding for methyl CpG binding protein 2 (MeCP2) has previously been identified as a substrate for the mRNA-binding protein, fragile X mental retardation protein (FMRP), which is silenced in FXS. Here, we report a homeostatic relationship between these two key regulators of gene expression in mouse models of FXS (Fmr1 Knockout (KO)) and Rett syndrome (MeCP2 KO). We found that the level of MeCP2 protein in the cerebral cortex was elevated in Fmr1 KO mice, whereas MeCP2 KO mice displayed reduced levels of FMRP, implicating interplay between the activities of MeCP2 and FMRP. Indeed, knockdown of MeCP2 with short hairpin RNAs led to a reduction of FMRP in mouse Neuro2A and in human HEK-293 cells, suggesting a reciprocal coupling in the expression level of these two regulatory proteins. Intra-cerebroventricular injection of an adeno-associated viral vector coding for FMRP led to a concomitant reduction in MeCP2 expression in vivo and partially corrected locomotor hyperactivity. Additionally, the level of MeCP2 in the posterior cortex correlated with the severity of the hyperactive phenotype in Fmr1 KO mice. These results demonstrate that MeCP2 and FMRP operate within a previously undefined homeostatic relationship. Our findings also suggest that MeCP2 overexpression in Fmr1 KO mouse posterior cerebral cortex may contribute to the fragile X locomotor hyperactivity phenotype. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32749543  Title: Male-specific features are reduced in Mecp2-null mice: analyses of vasopressinergic innervation, pheromone production and social behaviour.  Deficits in arginine vasopressin (AVP) and oxytocin (OT), two neuropeptides closely implicated in the modulation of social behaviours, have been reported in some early developmental disorders and autism spectrum disorders. Mutations in the X-linked methyl-CpG-binding protein 2 (MECP2) gene are associated to Rett syndrome and other neuropsychiatric conditions. Thus, we first analysed AVP and OT expression in the brain of Mecp2-mutant mice by immunohistochemistry. Our results revealed no significant differences in these systems in young adult Mecp2-heterozygous females, as compared to WT littermates. By contrast, we found a significant reduction in the sexually dimorphic, testosterone-dependent, vasopressinergic innervation in several nuclei of the social brain network and oxytocinergic innervation in the lateral habenula of Mecp2-null males, as compared to WT littermates. Analysis of urinary production of pheromones shows that Mecp2-null males lack the testosterone-dependent pheromone darcin, strongly suggesting low levels of androgens in these males. In addition, resident-intruder tests revealed lack of aggressive behaviour in Mecp2-null males and decreased chemoinvestigation of the intruder. By contrast, Mecp2-null males exhibited enhanced social approach, as compared to WT animals, in a 3-chamber social interaction test. In summary, Mecp2-null males, which display internal testicles, display a significant reduction of some male-specific features, such as vasopressinergic innervation within the social brain network, male pheromone production and aggressive behaviour. Thus, atypical social behaviours in Mecp2-null males may be caused, at least in part, by the effect of lack of MeCP2 over sexual differentiation. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32539526  Title: Diagnostic Classification for Human Autism and Obsessive-Compulsive Disorder Based on Machine Learning From a Primate Genetic Model.  Psychiatric disorders commonly comprise comorbid symptoms, such as autism spectrum disorder (ASD), obsessive-compulsive disorder (OCD), and attention deficit hyperactivity disorder (ADHD), raising controversies over accurate diagnosis and overlap of their neural underpinnings. The authors used noninvasive neuroimaging in humans and nonhuman primates to identify neural markers associated with DSM-5 diagnoses and quantitative measures of symptom severity. Resting-state functional connectivity data obtained from both wild-type and methyl-CpG binding protein 2 (MECP2) transgenic monkeys were used to construct monkey-derived classifiers for diagnostic classification in four human data sets (ASD: Autism Brain Imaging Data Exchange [ABIDE-I], N=1,112; ABIDE-II, N=1,114; ADHD-200 sample: N=776; OCD local institutional database: N=186). Stepwise linear regression models were applied to examine associations between functional connections of monkey-derived classifiers and dimensional symptom severity of psychiatric disorders. Nine core regions prominently distributed in frontal and temporal cortices were identified in monkeys and used as seeds to construct the monkey-derived classifier that informed diagnostic classification in human autism. This same set of core regions was useful for diagnostic classification in the OCD cohort but not the ADHD cohort. Models based on functional connections of the right ventrolateral prefrontal cortex with the left thalamus and right prefrontal polar cortex predicted communication scores of ASD patients and compulsivity scores of OCD patients, respectively. The identified core regions may serve as a basis for building markers for ASD and OCD diagnoses, as well as measures of symptom severity. These findings may inform future development of machine-learning models for psychiatric disorders and may improve the accuracy and speed of clinical assessments. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32269107  Title: MECP2 Duplication Causes Aberrant GABA Pathways, Circuits and Behaviors in Transgenic Monkeys: Neural Mappings to Patients with Autism.  MECP2 gain-of-function and loss-of-function in genetically engineered monkeys recapitulates typical phenotypes in patients with autism, yet where MECP2 mutation affects the monkey brain and whether/how it relates to autism pathology remain unknown. Here we report a combination of gene-circuit-behavior analyses including MECP2 coexpression network, locomotive and cognitive behaviors, and EEG and fMRI findings in 5 MECP2 overexpressed monkeys (Macaca fascicularis; 3 females) and 20 wild-type monkeys (Macaca fascicularis; 11 females). Whole-genome expression analysis revealed MECP2 coexpressed genes significantly enriched in GABA-related signaling pathways, whereby reduced β-synchronization within fronto-parieto-occipital networks was associated with abnormal locomotive behaviors. Meanwhile, MECP2-induced hyperconnectivity in prefrontal and cingulate networks accounted for regressive deficits in reversal learning tasks. Furthermore, we stratified a cohort of 49 patients with autism and 72 healthy controls of 1112 subjects using functional connectivity patterns, and identified dysconnectivity profiles similar to those in monkeys. By establishing a circuit-based construct link between genetically defined models and stratified patients, these results pave new avenues to deconstruct clinical heterogeneity and advance accurate diagnosis in psychiatric disorders.SIGNIFICANCE STATEMENT Autism spectrum disorder (ASD) is a complex disorder with co-occurring symptoms caused by multiple genetic variations and brain circuit abnormalities. To dissect the gene-circuit-behavior causal chain underlying ASD, animal models are established by manipulating causative genes such as MECP2 However, it is unknown whether such models have captured any circuit-level pathology in ASD patients, as demonstrated by human brain imaging studies. Here, we use transgenic macaques to examine the causal effect of MECP2 overexpression on gene coexpression, brain circuits, and behaviors. For the first time, we demonstrate that the circuit abnormalities linked to MECP2 and autism-like traits in the monkeys can be mapped to a homogeneous ASD subgroup, thereby offering a new strategy to deconstruct clinical heterogeneity in ASD. |
| MECP2, TCF4, TCF7L2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32015540  Title: A myelin-related transcriptomic profile is shared by Pitt-Hopkins syndrome models and human autism spectrum disorder.  Autism spectrum disorder (ASD) is genetically heterogeneous with convergent symptomatology, suggesting common dysregulated pathways. In this study, we analyzed brain transcriptional changes in five mouse models of Pitt-Hopkins syndrome (PTHS), a syndromic form of ASD caused by mutations in the TCF4 gene, but not the TCF7L2 gene. Analyses of differentially expressed genes (DEGs) highlighted oligodendrocyte (OL) dysregulation, which we confirmed in two additional mouse models of syndromic ASD (Ptenm3m4/m3m4 and Mecp2tm1.1Bird). The PTHS mouse models showed cell-autonomous reductions in OL numbers and myelination, functionally confirming OL transcriptional signatures. We also integrated PTHS mouse model DEGs with human idiopathic ASD postmortem brain RNA-sequencing data and found significant enrichment of overlapping DEGs and common myelination-associated pathways. Notably, DEGs from syndromic ASD mouse models and reduced deconvoluted OL numbers distinguished human idiopathic ASD cases from controls across three postmortem brain data sets. These results implicate disruptions in OL biology as a cellular mechanism in ASD pathology. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31947619  Title: Comprehensive Analysis of GABAA-A1R Developmental Alterations in Rett Syndrome: Setting the Focus for Therapeutic Targets in the Time Frame of the Disease.  Rett syndrome, a serious neurodevelopmental disorder, has been associated with an altered expression of different synaptic-related proteins and aberrant glutamatergic and γ-aminobutyric acid (GABA)ergic neurotransmission. Despite its severity, it lacks a therapeutic option. Through this work we aimed to define the relationship between MeCP2 and GABAA.-A1 receptor expression, emphasizing the time dependence of such relationship. For this, we analyzed the expression of the ionotropic receptor subunit in different MeCP2 gene-dosage and developmental conditions, in cells lines, and in primary cultured neurons, as well as in different developmental stages of a Rett mouse model. Further, RNAseq and systems biology analysis was performed from post-mortem brain biopsies of Rett patients. We observed that the modulation of the MeCP2 expression in cellular models (both Neuro2a (N2A) cells and primary neuronal cultures) revealed a MeCP2 positive effect on the GABAA.-A1 receptor subunit expression, which did not occur in other proteins such as KCC2 (Potassium-chloride channel, member 5). In the Mecp2+/- mouse brain, both the KCC2 and GABA subunits expression were developmentally regulated, with a decreased expression during the pre-symptomatic stage, while the expression was variable in the adult symptomatic mice. Finally, the expression of the gamma-aminobutyric acid (GABA) receptor-related synaptic proteins from the postmortem brain biopsies of two Rett patients was evaluated, specifically revealing the GABA A1R subunit overexpression. The identification of the molecular changes along with the Rett syndrome prodromic stages strongly endorses the importance of time frame when addressing this disease, supporting the need for a neurotransmission-targeted early therapeutic intervention. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31398341  Title: Targeting Peripheral Somatosensory Neurons to Improve Tactile-Related Phenotypes in ASD Models.  Somatosensory over-reactivity is common among patients with autism spectrum disorders (ASDs) and is hypothesized to contribute to core ASD behaviors. However, effective treatments for sensory over-reactivity and ASDs are lacking. We found distinct somatosensory neuron pathophysiological mechanisms underlie tactile abnormalities in different ASD mouse models and contribute to some ASD-related behaviors. Developmental loss of ASD-associated genes Shank3 or Mecp2 in peripheral mechanosensory neurons leads to region-specific brain abnormalities, revealing links between developmental somatosensory over-reactivity and the genesis of aberrant behaviors. Moreover, acute treatment with a peripherally restricted GABAA receptor agonist that acts directly on mechanosensory neurons reduced tactile over-reactivity in six distinct ASD models. Chronic treatment of Mecp2 and Shank3 mutant mice improved body condition, some brain abnormalities, anxiety-like behaviors, and some social impairments but not memory impairments, motor deficits, or overgrooming. Our findings reveal a potential therapeutic strategy targeting peripheral mechanosensory neurons to treat tactile over-reactivity and select ASD-related behaviors. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31389199  Title: Social-valence-related increased attention in rett syndrome cynomolgus monkeys: An eye-tracking study.  The cognitive phenotypes of Rett syndrome (RTT) remain unclarified compared with the well-defined genetic etiology. Recent clinical studies suggest the eye-tracking method as a promising avenue to quantify the visual phenotypes of the syndrome. The present study explored various aspects of visual attention of the methyl-CpG-binding protein 2 gene mutant RTT monkeys with the eye-tracking procedure. Comprehensive testing paradigms, including social valence comparison (SVC), visual paired comparison (VPC), and social recognition memory (SRM), were utilized to investigate their attentional features to social stimuli with differential valence, the novelty preferences, and short-term recognition memory, respectively. To explore the neurobiological mechanisms underlying the eye-tracking findings, we assessed changes of the brain subregion volumes and neurotransmitter concentrations. Compared with control monkeys, RTT monkeys demonstrated increased viewing on the more salient stare faces than profile faces in the SVC test, and increased viewing on the whole presented images composed of monkey faces in the VPC and SRM tests. Brain imaging revealed reduced bilateral occipital gyrus in RTT monkeys. The exploratory neurotransmitter analyses revealed no significant changes of various neurotransmitter concentrations in the cerebrospinal fluid and blood of RTT monkeys. The eye-tracking results suggested social-valence-related increased attention in RTT monkeys, supplementing the cognitive phenotypes associated with the syndrome. Further investigations from broader perspectives are required to uncover the underlying neurobiological mechanisms. Autism Res 2019, 00: 1-13. © 2019 International Society for Autism Research, Wiley Periodicals, Inc. LAY SUMMARY: Altered expressions of the methyl-CpG-binding protein 2 (MECP2) gene are usually associated with neurodevelopmental disorders, such as autism spectrum disorders, Rett syndrome (RTT), and so forth. The present eye-tracking study found social-valence-related increased attention in our firstly established MECP2 mutant RTT monkeys. The novel findings supplement the cognitive phenotypes and potentially benefit the behavioral interventions of the RTT syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31333414  Title: MeCP2 Deficiency Disrupts Kainate-Induced Presynaptic Plasticity in the Mossy Fiber Projections in the Hippocampus.  Methyl cytosine binding protein 2 (MeCP2) is a structural chromosomal protein involved in the regulation of gene expression. Mutations in the gene encoding MeCP2 result in Rett Syndrome (RTT), a pervasive neurodevelopmental disorder. RTT is one of few autism spectrum disorders whose cause was identified as a single gene mutation. Remarkably, abnormal levels of MeCP2 have been associated to other neurodevelopmental disorders, as well as neuropsychiatric disorders. Therefore, many studies have been oriented to investigate the role of MeCP2 in the nervous system. In the present work, we explore cellular and molecular mechanisms affecting synaptic plasticity events in vivo in the hippocampus of MeCP2 mutant mice. While most studies addressed postsynaptic defects in the absence of MeCP2, we took advantage of an in vivo activity-paradigm (seizures), two models of MeCP2 deficiency, and neurobiological assays to reveal novel defects in presynaptic structural plasticity in the hippocampus in RTT rodent models. These approaches allowed us to determine that MeCP2 mutations alter presynaptic components, i.e., disrupts the plastic response of mossy fibers to synaptic activity and results in reduced axonal growth which is correlated with imbalanced trophic and guidance support, associated with aberrant expression of brain-derived neurotrophic factor and semaphorin 3F. Our results also revealed that adult-born granule cells recapitulate maturational defects that have been only shown at early postnatal ages. As these cells do not mature timely, they may not integrate properly into the adult hippocampal circuitry. Finally, we performed a hippocampal-dependent test that revealed defective spatial memory in these mice. Altogether, our studies establish a model that allows us to evaluate the effect of the manipulation of specific pathways involved in axonal guidance, synaptogenesis, or maturation in specific circuits and correlate it with changes in behavior. Understanding the mechanisms underlying the neuronal compromise caused by mutations in MeCP2 could provide information on the pathogenic mechanism of autistic spectrum disorders and improve our understanding of brain development and molecular basis of behavior. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31332003  Title: Deep learning of spontaneous arousal fluctuations detects early cholinergic defects across neurodevelopmental mouse models and patients.  Neurodevelopmental spectrum disorders like autism (ASD) are diagnosed, on average, beyond age 4 y, after multiple critical periods of brain development close and behavioral intervention becomes less effective. This raises the urgent need for quantitative, noninvasive, and translational biomarkers for their early detection and tracking. We found that both idiopathic (BTBR) and genetic (CDKL5- and MeCP2-deficient) mouse models of ASD display an early, impaired cholinergic neuromodulation as reflected in altered spontaneous pupil fluctuations. Abnormalities were already present before the onset of symptoms and were rescued by the selective expression of MeCP2 in cholinergic circuits. Hence, we trained a neural network (ConvNetACh) to recognize, with 97% accuracy, patterns of these arousal fluctuations in mice with enhanced cholinergic sensitivity (LYNX1-deficient). ConvNetACh then successfully detected impairments in all ASD mouse models tested except in MeCP2-rescued mice. By retraining only the last layers of ConvNetACh with heart rate variation data (a similar proxy of arousal) directly from Rett syndrome patients, we generated ConvNetPatients, a neural network capable of distinguishing them from typically developing subjects. Even with small cohorts of rare patients, our approach exhibited significant accuracy before (80% in the first and second year of life) and into regression (88% in stage III patients). Thus, transfer learning across species and modalities establishes spontaneous arousal fluctuations combined with deep learning as a robust noninvasive, quantitative, and sensitive translational biomarker for the rapid and early detection of neurodevelopmental disorders before major symptom onset. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31323913  Title: High Functioning Autism with Missense Mutations in Synaptotagmin-Like Protein 4 (SYTL4) and Transmembrane Protein 187 (TMEM187) Genes: SYTL4- Protein Modeling, Protein-Protein Interaction, Expression Profiling and MicroRNA Studies.  We describe a 7-year-old male with high functioning autism spectrum disorder (ASD) and maternally-inherited rare missense variant of Synaptotagmin-like protein 4 (SYTL4) gene (Xq22.1; c.835C>T; p.Arg279Cys) and an unknown missense variant of Transmembrane protein 187 (TMEM187) gene (Xq28; c.708G>T; p. Gln236His). Multiple in-silico predictions described in our study indicate a potentially damaging status for both X-linked genes. Analysis of predicted atomic threading models of the mutant and the native SYTL4 proteins suggest a potential structural change induced by the R279C variant which eliminates the stabilizing Arg279-Asp60 salt bridge in the N-terminal half of the SYTL4, affecting the functionality of the protein's critical RAB-Binding Domain. In the European (Non-Finnish) population, the allele frequency for this variant is 0.00042. The SYTL4 gene is known to directly interact with several members of the RAB family of genes, such as, RAB27A, RAB27B, RAB8A, and RAB3A which are known autism spectrum disorder genes. The SYTL4 gene also directly interacts with three known autism genes: STX1A, SNAP25 and STXBP1. Through a literature-based analytical approach, we identified three of five (60%) autism-associated serum microRNAs (miRs) with high predictive power among the total of 298 mouse Sytl4 associated/predicted microRNA interactions. Five of 13 (38%) miRs were differentially expressed in serum from ASD individuals which were predicted to interact with the mouse equivalent Sytl4 gene. TMEM187 gene, like SYTL4, is a protein-coding gene that belongs to a group of genes which host microRNA genes in their introns or exons. The novel Q236H amino acid variant in the TMEM187 in our patient is near the terminal end region of the protein which is represented by multiple sequence alignments and hidden Markov models, preventing comparative structural analysis of the variant harboring region. Like SYTL4, the TMEM187 gene is expressed in the brain and interacts with four known ASD genes, namely, HCFC1; TMLHE; MECP2; and GPHN. TMM187 is in linkage with MECP2, which is a well-known determinant of brain structure and size and is a well-known autism gene. Other members of the TMEM gene family, TMEM132E and TMEM132D genes are associated with bipolar and panic disorders, respectively, while TMEM231 is a known syndromic autism gene. Together, TMEM187 and SYTL4 genes directly interact with recognized important ASD genes, and their mRNAs are found in extracellular vesicles in the nervous system and stimulate target cells to translate into active protein. Our evidence shows that both these genes should be considered as candidate genes for autism. Additional biological testing is warranted to further determine the pathogenicity of these gene variants in the causation of autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31273723  Title: Behavioral Characterization of MeCP2 Dysfunction-Associated Rett Syndrome and Neuropsychiatric Disorders.  The methyl-CpG-binding protein 2 (MECP2) gene has been implicated in multiple neuropsychiatric disorders such as autism and schizophrenia and, most notably, Rett syndrome (RTT). Mouse models of MeCP2 dysfunction that have been developed are thus important not only for examining the protein's contribution to RTT, but also for elucidating the etiologies of other MECP2-associated neuropsychiatric disorders. In this chapter, we present protocols for three behavioral assays for characterizing major functional domains of MeCP2 dysfunction-the open field test for measuring general locomotor activity and anxiety-like behavior, the three-chambered Crawley box test for assessing social preference and social novelty, and the rotarod assay for testing locomotor coordination. It is hoped that these information facilitate systematic characterization of mouse models that may aid in elucidating the role of MeCP2 in neurological disorders, as well as assessing the effects of putative mechanistic and therapeutic interventions. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31273722  Title: MeCP2 Dysfunction in Rett Syndrome and Neuropsychiatric Disorders.  Elucidating the functions of a particular gene is paramount to the understanding of how its dysfunction contributes to disease. This is especially important when the gene is implicated in multiple different disorders. One such gene is methyl-CpG-binding protein 2 (MECP2), which has been most prominently associated with the neurodevelopmental disorder Rett syndrome, as well as major neuropsychiatric disorders such as autism and schizophrenia. Being initially identified as a transcriptional regulator that modulates gene expression and subsequently also shown to be involved in other molecular events, dysfunction of the MeCP2 protein has the potential to affect many cellular processes. In this chapter, we will briefly review the functions of the MeCP2 protein and how its mutations are implicated in Rett syndrome and other neuropsychiatric disorders. We will further discuss about the mouse models that have been generated to specifically dissect the function of MeCP2 in different cell types and brain regions. It is envisioned that such thorough and targeted examination of MeCP2 functions can aid in enlightening the role that it plays in normal and dysfunctional physiological systems. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31193946  Title: LINE1 and Mecp2 methylation of the adult striatum and prefrontal cortex exposed to prenatal immune activation.  Prenatal exposure to infection and inflammation increases the risk of neurodevelopmental disorders such as schizophrenia and autism. The etiology could be partly through transgenerational and modifiable DNA methylation changes in the adult offspring's brain. This data descriptor presents a dataset of global DNA methylation (using LINE1 assay) and Mecp2 promoter methylation in adolescent and adult brain tissue of offspring exposed to prenatal immune activation on gestation day 9 and offspring of saline exposed mice. PCR based methylation assays using Sequenom EpiTYPER was used to quantify DNA methylation at promoter CpG methylation of Long Interspersed Elements-1 (LINE1 or L1) and Mecp2. The dataset also includes global DNA methylation and Mecp2 promoter methylation profile at 6 and 12 weeks following early dietary intervention with omega-3 (n-3) PUFA. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31185809  Title: Understanding intellectual disability and autism spectrum disorders from common mouse models: synapses to behaviour.  Normal brain development is highly dependent on the timely coordinated actions of genetic and environmental processes, and an aberration can lead to neurodevelopmental disorders (NDDs). Intellectual disability (ID) and autism spectrum disorders (ASDs) are a group of co-occurring NDDs that affect between 3% and 5% of the world population, thus presenting a great challenge to society. This problem calls for the need to understand the pathobiology of these disorders and to design new therapeutic strategies. One approach towards this has been the development of multiple analogous mouse models. This review discusses studies conducted in the mouse models of five major monogenic causes of ID and ASDs: Fmr1, Syngap1, Mecp2, Shank2/3 and Neuroligins/Neurnexins. These studies reveal that, despite having a diverse molecular origin, the effects of these mutations converge onto similar or related aetiological pathways, consequently giving rise to the typical phenotype of cognitive, social and emotional deficits that are characteristic of ID and ASDs. This convergence, therefore, highlights common pathological nodes that can be targeted for therapy. Other than conventional therapeutic strategies such as non-pharmacological corrective methods and symptomatic alleviation, multiple studies in mouse models have successfully proved the possibility of pharmacological and genetic therapy enabling functional recovery. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31133783  Title: Atypical Response Properties of the Auditory Cortex of Awake MECP2-Overexpressing Mice.  Methyl-CpG binding protein 2 (MECP2) is a gene associated with DNA methylation and has been found to be important for maintaining brain function. In humans, overexpression of MECP2 can cause a severe developmental disorder known as MECP2 duplication syndrome. However, it is still unclear whether MECP2 overexpression also causes auditory abnormalities, which are common in people with autism. MECP2-TG is a mouse model of MECP2 duplication syndrome and has been widely used for research on social difficulty and other autism-like disorders. In this study, we used a combination of multiple electrophysiological techniques to document the response properties of the auditory cortex of awake MECP2-TG mice. Our results showed that while the auditory brainstem responses are similar, cortical activity patterns including local field potentials (LFPs), multiunit activity (MUA), and single-neuron responses differ between MECP2-TG and wild-type (WT) mice. At the single-neuron level, the spike waveform of fast-spiking (FS) neurons from MECP2-TG mice is different from that of WT mice, as reflected by reduced peak/trough ratios in the transgenic mice. Both regular-spiking (RS) and FS neurons exhibited atypical response properties in MECP2-TG mice compared with WT mice, such as prolonged latency and an elevated intensity threshold; furthermore, regarding the response strength to different stimuli, MECP2-TG mice exhibited stronger responses to noise than to pure tone, while this pattern was not observed in WT mice. Our findings suggest that MECP2 overexpression can cause the auditory cortex to have atypical response properties, an implication that could be helpful for further understanding the nature of auditory deficits in autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31112129  Title: Ventral hippocampal projections to the medial prefrontal cortex regulate social memory.  Inputs from the ventral hippocampus (vHIP) to the medial prefrontal cortex (mPFC) are implicated in several neuropsychiatric disorders. Here, we show that the vHIP-mPFC projection is hyperactive in the Mecp2 knockout mouse model of the autism spectrum disorder Rett syndrome, which has deficits in social memory. Long-term excitation of mPFC-projecting vHIP neurons in wild-type mice impaired social memory, whereas their long-term inhibition in Rett mice rescued social memory deficits. The extent of social memory improvement was negatively correlated with vHIP-evoked responses in mPFC slices, on a mouse-per-mouse basis. Acute manipulations of the vHIP-mPFC projection affected social memory in a region and behavior selective manner, suggesting that proper vHIP-mPFC signaling is necessary to recall social memories. In addition, we identified an altered pattern of vHIP innervation of mPFC neurons, and increased synaptic strength of vHIP inputs onto layer five pyramidal neurons as contributing factors of aberrant vHIP-mPFC signaling in Rett mice. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31013990  Title: DNA Methylation Contributes to the Differential Expression Levels of Mecp2 in Male Mice Neurons and Astrocytes.  Methyl CpG binding protein-2 (MeCP2) isoforms (E1 and E2) are important epigenetic regulators in brain cells. Accordingly, MeCP2 loss- or gain-of-function mutation causes neurodevelopmental disorders, including Rett syndrome (RTT), MECP2 duplication syndrome (MDS), and autism spectrum disorders (ASD). Within different types of brain cells, highest MeCP2 levels are detected in neurons and the lowest in astrocytes. However, our current knowledge of Mecp2/MeCP2 regulatory mechanisms remains largely elusive. It appears that there is a sex-dependent effect in X-linked MeCP2-associated disorders, as RTT primarily affects females, whereas MDS is found almost exclusively in males. This suggests that Mecp2 expression levels in brain cells might be sex-dependent. Here, we investigated the sex- and cell type-specific expression of Mecp2 isoforms in male and female primary neurons and astrocytes isolated from the murine forebrain. Previously, we reported that DNA methylation of six Mecp2 regulatory elements correlated with Mecp2 levels in the brain. We now show that in male brain cells, DNA methylation is significantly correlated with the transcript expression of these two isoforms. We show that both Mecp2 isoforms are highly expressed in male neurons compared to male astrocytes, with Mecp2e1 expressed at higher levels than Mecp2e2. Our data indicate that higher DNA methylation at the Mecp2 regulatory element(s) is associated with lower levels of Mecp2 isoforms in male astrocytes compared to male neurons. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30636904  Title: Striatal Inhibition of MeCP2 or TSC1 Produces Sociability Deficits and Repetitive Behaviors.  Autism spectrum disorder (ASD) is a heterogeneous group of neurobehavioral disorders characterized by the two core domains of behavioral deficits, including sociability deficits and stereotyped repetitive behaviors. It is not clear whether the core symptoms of ASD are produced by dysfunction of the overall neural network of the brain or that of a limited brain region. Recent studies reported that excessive glutamatergic or dopaminergic inputs in the dorsal striatum induced sociability deficits and repetitive behaviors. These findings suggest that the dorsal striatum plays a crucial role in autistic-like behaviors. The present study addresses whether functional deficits of well-known ASD-related genes in the dorsal striatum also produce ASD core symptoms. This study also examines whether these behavioral changes can be modulated by rebalancing glutamate and/or dopamine receptor activity in the dorsal striatum. First, we found that the siRNA-mediated inhibition of Shank3, Nlgn3, Fmr1, Mecp2, or Tsc1 in the dorsal striatum produced mild to severe behavioral changes in sociability, cognition, and/or repetitive behaviors. The knockdown effects of Mecp2 and Tsc1 on behavioral changes were the most prominent. Next, we demonstrated that behavioral changes induced by striatal inhibition of MeCP2 and TSC1 were rescued by D-cycloserine (an NMDA agonist), fenobam (an mGluR5 antagonist), SCH23390 (a D1 antagonist), and/or ecopipam (a D1 partial antagonist), pharmacological drugs that are known to regulate ASD-like symptoms in animal models. Collectively, these results suggest that the dorsal striatum is a critical brain region that, when dysfunctional, produces the core symptoms of ASD. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30560934  Title: MicroRNA-197 controls ADAM10 expression to mediate MeCP2's role in the differentiation of neuronal progenitors.  Duplication of MECP2 (Methyl-CpG-binding protein 2) causes severe mental illness called MECP2 duplication syndrome (MDS), yet the underlying mechanism remains elusive. Here we show, in Tg(MECP2) transgenic mouse brain or cultured neural progenitor cells (NPCs), that elevated MeCP2 expression promotes NPC differentiation into neurons. Ectopic expression of MeCP2 inhibits ADAM10 and thus the NOTCH pathway during NPC differentiation. In human cells, this downregulation on ADAM10 was mediated by miRNA-197, which is upregulated by MeCP2. Surprisingly, miR-197 binds to the ADAM10 3'-UTR via its 3' side, not the canonical seed sequence on the 5' side. In mouse cells, a noncoding RNA Gm28836 is used to replace the function of miR-197 between MeCP2 and ADAM10. Similar to MeCP2, overexpressing miR-197 also promotes NPCs differentiation into neurons. Interestingly, three rare missense mutations (H371R, E394K, and G428S) in MECP2, which we identified in a Han Chinese autism spectrum disorders (ASD) cohort showed loss-of-function effects in NPC differentiation assay. These mutations cannot upregulate miR-197. Overexpressing miR-197 together with these MeCP2 mutations could rescue the downregulation on ADAM10. Not only the inhibitor of miR-197 could reverse the effect of overexpressed MeCP2 on NPCs differentiation, but also overexpression of miR-197 could reverse the NPCs differentiation defects caused by MECP2 mutations. Our results revealed that a regulatory axis involving MeCP2, miR-197, ADAM10, and NOTCH signaling is critical for NPC differentiation, which is affected by both MeCP2 duplication and mutation. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30402028  Title: Dual mechanisms for the regulation of brain-derived neurotrophic factor by valproic acid in neural progenitor cells.  Autism spectrum disorders (ASDs) are neurodevelopmental disorders that share behavioral features, the results of numerous studies have suggested that the underlying causes of ASDs are multifactorial. Behavioral and/or neurobiological analyses of ASDs have been performed extensively using a valid model of prenatal exposure to valproic acid (VPA). Abnormal synapse formation resulting from altered neurite outgrowth in neural progenitor cells (NPCs) during embryonic brain development has been observed in both the VPA model and ASD subjects. Although several mechanisms have been suggested, the actual mechanism underlying enhanced neurite outgrowth remains unclear. In this study, we found that VPA enhanced the expression of brain-derived neurotrophic factor (BDNF), particularly mature BDNF (mBDNF), through dual mechanisms. VPA increased the mRNA and protein expression of BDNF by suppressing the nuclear expression of methyl-CpG-binding protein 2 (MeCP2), which is a transcriptional repressor of BDNF. In addition, VPA promoted the expression and activity of the tissue plasminogen activator (tPA), which induces BDNF maturation through proteolytic cleavage. Trichostatin A and sodium butyrate also enhanced tPA activity, but tPA activity was not induced by valpromide, which is a VPA analog that does not induce histone acetylation, indicating that histone acetylation activity was required for tPA regulation. VPA-mediated regulation of BDNF, MeCP2, and tPA was not observed in astrocytes or neurons. Therefore, these results suggested that VPA-induced mBDNF upregulation was associated with the dysregulation of MeCP2 and tPA in developing cortical NPCs. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30341178  Title: FABP3 in the Anterior Cingulate Cortex Modulates the Methylation Status of the Glutamic Acid Decarboxylase67 Promoter Region.  Polyunsaturated fatty acids (PUFAs) are essential for brain development and function. Increasing evidence has shown that an imbalance of PUFAs is associated with various human psychiatric disorders, including autism and schizophrenia. Fatty acid-binding proteins (FABPs), cellular chaperones of PUFAs, are involved in PUFA intracellular trafficking, signal transduction, and gene transcription. In this study, we show that FABP3 is strongly expressed in the GABAergic inhibitory interneurons of the male mouse anterior cingulate cortex (ACC), which is a component of the limbic cortex and is important for the coordination of cognitive and emotional behaviors. Interestingly, Fabp3 KO male mice show an increase in the expression of the gene encoding the GABA-synthesizing enzyme glutamic acid decarboxylase 67 (Gad67) in the ACC. In the ACC of Fabp3 KO mice, Gad67 promoter methylation and the binding of methyl-CpG binding protein 2 (MeCP2) and histone deacetylase 1 (HDAC1) to the Gad67 promoter are significantly decreased compared with those in WT mice. The abnormal cognitive and emotional behaviors of Fabp3 KO mice are restored by methionine administration. Notably, methionine administration normalizes Gad67 promoter methylation and its mRNA expression in the ACC of Fabp3 KO mice. These findings demonstrate that FABP3 is involved in the control of DNA methylation of the Gad67 promoter and activation of GABAergic neurons in the ACC, thus suggesting the importance of PUFA homeostasis in the ACC for cognitive and emotional behaviors.SIGNIFICANCE STATEMENT The ACC is important for emotional and cognitive processing. However, the mechanisms underlying its involvement in the control of behavioral responses are largely unknown. We show the following new observations: (1) FABP3, a PUFA cellular chaperone, is exclusively expressed in GABAergic interneurons in the ACC; (2) an increase in Gad67 expression is detected in the ACC of Fabp3 KO mice; (3) the Gad67 promoter is hypomethylated and the binding of transcriptional repressor complexes is decreased in the ACC of Fabp3 KO mice; and (4) elevated Gad67 expression and abnormal behaviors seen in Fabp3 KO mice are mostly recovered by methionine treatment. These suggest that FABP3 regulates GABA synthesis through transcriptional regulation of Gad67 in the ACC. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30312858  Title: Deletion of TLR-4 attenuates fetal alcohol exposure-induced gene expression and social interaction deficits.  Fetal alcohol spectrum disorders (FASD) are associated with social interaction behavior and gastrointestinal (GI) abnormalities. These abnormal behaviors and GI abnormalities overlap with autism spectrum disorder (ASD). We investigated the effect of fetal alcohol exposure (FAE) on social interaction deficits (hallmark of autism) in mice. Evidence indicates that exogenous lipopolysaccharide (LPS) administration during gestation induces autism-like behavior in the offspring. LPS regulates the expression of genes underlying differentiation, immune function, myelination, and synaptogenesis in fetal brain by the LPS receptor, TLR-4-dependent mechanism. In this study, we evaluated the role of TLR-4 in FAE-induced social behavior deficit. WT and TLR4-/- pregnant mice were fed Lieber-DeCarli liquid diet with or without ethanol. The control group was pair-fed with an isocaloric diet. Social behavior was tested in the adult offspring at postnatal day 60. Frontal cortex mRNA expression of autistic candidate genes (Ube3a, Gabrb3, Mecp2) and inflammatory cytokine genes (IL-1β, IL-6, TNF-α) were measured by RT-qPCR. Adult male offspring of ethanol-fed WT dams showed low birth weight compared to offspring of pair-fed WT dams. However, their body weights at adulthood were greater compared to the body weights of offspring of pair-fed WT dams. There were no body weight differences in offspring of TLR4-/- dams. Social interaction deficit was observed only in male offspring of ethanol-fed WT dams, but it was not observed in both male and female offspring of ethanol-fed TLR4-/- dams. Expressions of autism candidate genes, Gabrb3 and Ube3a, were elevated, while that of the Mecp2 gene was suppressed in the frontal cortex of male, but not female, offspring of ethanol-fed WT mice. The expressions of inflammatory cytokine genes, IL-1β, IL-6, and TNF-α, were also significantly increased in the frontal cortex of male, but not female, offspring of ethanol-fed dams. The changes in the expression of autistic and cytokine genes were unaffected in the offspring of ethanol-fed TLR4-/- dams. These data also indicate that TLR4 mediates FAE-induced changes in social interactions and gene expression in brain, suggesting that ethanol-induced LPS absorption from the maternal gut may be involved in gene expression changes in the fetal brain. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30304570  Title: Application of Human-Induced Pluripotent Stem Cells (hiPSCs) to Study Synaptopathy of Neurodevelopmental Disorders.  Synapses are the basic structural and functional units for information processing and storage in the brain. Their diverse properties and functions ultimately underlie the complexity of human behavior. Proper development and maintenance of synapses are essential for normal functioning of the nervous system. Disruption in synaptogenesis and the consequent alteration in synaptic function have been strongly implicated to cause neurodevelopmental disorders such as autism spectrum disorders (ASDs) and schizophrenia (SCZ). The introduction of human-induced pluripotent stem cells (hiPSCs) provides a new path to elucidate disease mechanisms and potential therapies. In this review, we will discuss the advantages and limitations of using hiPSC-derived neurons to study synaptic disorders. Many mutations in genes encoding for proteins that regulate synaptogenesis have been identified in patients with ASDs and SCZ. We use Methyl-CpG binding protein 2 (MECP2), SH3 and multiple ankyrin repeat domains 3 (SHANK3) and Disrupted in schizophrenia 1 (DISC1) as examples to illustrate the promise of using hiPSCs as cellular models to elucidate the mechanisms underlying disease-related synaptopathy. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30111726  Title: Alterations in the MicroRNA of the Blood of Autism Spectrum Disorder Patients: Effects on Epigenetic Regulation and Potential Biomarkers.  Aims: Autism spectrum disorder (ASD) refers to a group of heterogeneous brain-based neurodevelopmental disorders with different levels of symptom severity. Given the challenges, the clinical diagnosis of ASD is based on information gained from interviews with patients' parents. The heterogeneous pathogenesis of this disorder appears to be driven by genetic and environmental interactions, which also plays a vital role in predisposing individuals to ASD with different commitment levels. In recent years, it has been proposed that epigenetic modifications directly contribute to the pathogenesis of several neurodevelopmental disorders, such as ASD. The microRNAs (miRNAs) comprises a species of short noncoding RNA that regulate gene expression post-transcriptionally and have an essential functional role in the brain, particularly in neuronal plasticity and neuronal development, and could be involved in ASD pathophysiology. The aim of this study is to evaluate the expression of blood miRNA in correlation with clinical findings in patients with ASD, and to find possible biomarkers for the disorder. Results: From a total of 26 miRNA studied, seven were significantly altered in ASD patients, when compared to the control group: miR34c-5p, miR92a-2-5p, miR-145-5p and miR199a-5p were up-regulated and miR27a-3p, miR19-b-1-5p and miR193a-5p were down-regulated in ASD patients. Discussion: The main targets of these miRNAs are involved in immunological developmental, immune response and protein synthesis at transcriptional and translational levels. The up-regulation of both miR-199a-5p and miR92a-2a and down-regulation of miR-193a and miR-27a was observed in AD patients, and may in turn affect the SIRT1, HDAC2, and PI3K/Akt-TSC:mTOR signaling pathways. Furthermore, MeCP2 is a target of miR-199a-5p, and is involved in Rett Syndrome (RTT), which possibly explains the autistic phenotype in male patients with this syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30105297  Title: Increased Axonal Bouton Stability during Learning in the Mouse Model of MECP2 Duplication Syndrome.  MECP2 duplication syndrome is an X-linked form of syndromic autism caused by genomic duplication of the region encoding methyl-CpG-binding protein 2 (MECP2). Mice overexpressing MECP2 demonstrate social impairment, behavioral inflexibility, and altered patterns of learning and memory. Previous work showed abnormally increased stability of dendritic spines formed during motor training in the apical tuft of primary motor cortex (area M1) corticospinal neurons in the MECP2 duplication mouse model. In the current study, we measure the structural plasticity of axonal boutons in layer 5 pyramidal neuron projections to layer 1 of area M1 during motor training. In wild-type littermate control mice, we find that during rotarod training the bouton formation rate changes minimally, if at all, while the bouton elimination rate more than doubles. Notably, the observed upregulation in bouton elimination with training is absent in MECP2 duplication mice. This result provides further evidence of an imbalance between structural stability and plasticity in this form of syndromic autism. Furthermore, the observation that axonal bouton elimination more than doubles with motor training in wild-type animals contrasts with the increase of dendritic spine consolidation observed in corticospinal neurons at the same layer. This dissociation suggests that different area M1 microcircuits may manifest different patterns of structural synaptic plasticity during motor training. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29984470  Title: Imaging genetics in neurodevelopmental psychopathology.  Neurodevelopmental disorders are defined by highly heritable problems during development and brain growth. Attention-deficit/hyperactivity disorder (ADHD), autism spectrum disorders (ASDs), and intellectual disability (ID) are frequent neurodevelopmental disorders, with common comorbidity among them. Imaging genetics studies on the role of disease-linked genetic variants on brain structure and function have been performed to unravel the etiology of these disorders. Here, we reviewed imaging genetics literature on these disorders attempting to understand the mechanisms of individual disorders and their clinical overlap. For ADHD and ASD, we selected replicated candidate genes implicated through common genetic variants. For ID, which is mainly caused by rare variants, we included genes for relatively frequent forms of ID occurring comorbid with ADHD or ASD. We reviewed case-control studies and studies of risk variants in healthy individuals. Imaging genetics studies for ADHD were retrieved for SLC6A3/DAT1, DRD2, DRD4, NOS1, and SLC6A4/5HTT. For ASD, studies on CNTNAP2, MET, OXTR, and SLC6A4/5HTT were found. For ID, we reviewed the genes FMR1, TSC1 and TSC2, NF1, and MECP2. Alterations in brain volume, activity, and connectivity were observed. Several findings were consistent across studies, implicating, for example, SLC6A4/5HTT in brain activation and functional connectivity related to emotion regulation. However, many studies had small sample sizes, and hypothesis-based, brain region-specific studies were common. Results from available studies confirm that imaging genetics can provide insight into the link between genes, disease-related behavior, and the brain. However, the field is still in its early stages, and conclusions about shared mechanisms cannot yet be drawn. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29970983  Title: Common Defects of Spine Dynamics and Circuit Function in Neurodevelopmental Disorders: A Systematic Review of Findings From in Vivo Optical Imaging of Mouse Models.  In vivo optical imaging is a powerful tool for revealing brain structure and function at both the circuit and cellular levels. Here, we provide a systematic review of findings obtained from in vivo imaging studies of mouse models of neurodevelopmental disorders, including the monogenic disorders fragile X syndrome, Rett syndrome, and Angelman syndrome, which are caused by genetic abnormalities of FMR1, MECP2, and UBE3A, as well as disorders caused by copy number variations (15q11-13 duplication and 22q11.2 deletion) and BTBR mice as an inbred strain model of autism spectrum disorder (ASD). Most studies visualize the structural and functional responsiveness of cerebral cortical neurons to sensory stimuli and the developmental and experience-dependent changes in these responses as a model of brain functions affected by these disorders. The optical imaging techniques include two-photon microscopy of fluorescently labeled dendritic spines or neurons loaded with fluorescent calcium indicators and macroscopic imaging of cortical activity using calcium indicators, voltage-sensitive dyes or intrinsic optical signals. Studies have revealed alterations in the density, stability, and turnover of dendritic spines, aberrant cortical sensory responses, impaired inhibitory function, and concomitant failure of circuit maturation as common causes for neurological deficits. Mechanistic hypotheses derived from in vivo imaging also provide new directions for therapeutic interventions. For instance, it was recently demonstrated that early postnatal administration of a selective serotonin reuptake inhibitor (SSRI) restores impaired cortical inhibitory function and ameliorates the aberrant social behaviors in a mouse model of ASD. We discuss the potential use of SSRIs for treating ASDs in light of these findings. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29967385  Title: Prenatal immune activation alters the adult neural epigenome but can be partly stabilised by a n-3 polyunsaturated fatty acid diet.  An unstable epigenome is implicated in the pathophysiology of neurodevelopmental disorders such as schizophrenia and autism. This is important because the epigenome is potentially modifiable. We have previously reported that adult offspring exposed to maternal immune activation (MIA) prenatally have significant global DNA hypomethylation in the hypothalamus. However, what genes had altered methylation state, their functional effects on gene expression and whether these changes can be moderated, have not been addressed. In this study, we used next-generation sequencing (NGS) for methylome profiling in a MIA rodent model of neurodevelopmental disorders. We assessed whether differentially methylated regions (DMRs) affected the chromatin state by mapping known DNase I hypersensitivity sites (DHSs), and selected overlapping genes to confirm a functional effect of MIA on gene expression using qPCR. Finally, we tested whether methylation differences elicited by MIA could be limited by post-natal dietary (omega) n-3 polyunsaturated fatty acid (PUFA) supplementation. These experiments were conducted using hypothalamic brain tissue from 12-week-old offspring of mice injected with viral analogue PolyI:C on gestation day 9 of pregnancy or saline on gestation day 9. Half of the animals from each group were fed a diet enriched with n-3 PUFA from weaning (MIA group, n = 12 units, n = 39 mice; Control group, n = 12 units, n = 38 mice). The results confirmed our previous finding that adult offspring exposed to MIA prenatally had significant global DNA hypomethylation. Furthermore, genes linked to synaptic plasticity were over-represented among differentially methylated genes following MIA. More than 80% of MIA-induced hypomethylated sites, including those affecting chromatin state and MECP2 binding, were stabilised by the n-3 PUFA intervention. MIA resulted in increased expression of two of the 'top five' genes identified from an integrated analysis of DMRs, DHSs and MECP2 binding sites, namely Abat (t = 2.46, p < 0.02) and Gnas9 (t = 2.96, p < 0.01), although these changes were not stabilised by dietary intervention. Thus, prenatal MIA exposure impacts upon the epigenomic regulation of gene pathways linked to neurodevelopmental conditions; and many of the changes can be attenuated by a low-cost dietary intervention. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29738885  Title: Regulation of seizure-induced MeCP2 Ser421 phosphorylation in the developing brain.  Neonatal seizures disrupt normal synaptic maturation and often lead to later-life epilepsy and cognitive deficits. During early life, the brain exhibits heightened synaptic plasticity, in part due to a developmental overabundance of CaV1.2 L-type voltage gated calcium (Ca2+) channels (LT-VGCCs) and Ca2+-permeable AMPARs (CP-AMPARs) lacking GluA2 subunits. We hypothesized that early-life seizures overactivate these channels, in turn dysregulating Ca2+-dependent signaling pathways including that of methyl CPG binding protein 2 (MeCP2), a transcription factor implicated in the autism spectrum disorder (ASD) Rett Syndrome. Here, we show that in vivo hypoxia-induced seizures (HS) in postnatal day (P)10 rats acutely induced phosphorylation of the neuronal-specific target of activity-dependent MeCP2 phosphorylation, S421, as well as its upstream activator CaMKII T286. We next identified mechanisms by which activity-dependent Ca2+ influx induced MeCP2 phosphorylation using in vitro cortical and hippocampal neuronal cultures at embryonic day (E)18 + 10 days in vitro (DIV). In contrast to the prevalent role of NMDARs in the adult brain, we found that both CP-AMPARs and LT-VGCCs mediated MeCP2 S421 and CaMKII T286 phosphorylation induced by kainic acid (KA) or high potassium chloride (KCl) stimulation. Furthermore, in vivo post-seizure treatment with the broad-spectrum AMPAR antagonist NBQX, the CP-AMPAR blocker IEM-1460, or the LT-VGCC antagonist nimodipine blocked seizure-induced MeCP2 phosphorylation. Collectively, these results demonstrate that early-life seizures dysregulate critical activity-dependent developmental signaling pathways, in part via CP-AMPAR and LT-VGCC activation, providing novel age-specific therapeutic targets for convergent pathways underlying epilepsy and ASDs. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29698767  Title: Regulation of neural differentiation, synaptic scaling and animal behavior by MeCP2 phophorylation.  Highly expressed in the mammalian brain and widely distributed across the genome, MeCP2 is a key player in recognizing modified DNA and interpreting the epigenetic information encoded in different DNA methylation/hydroxymethylation patterns. Alterations in sequence or copy number of the X-linked human MECP2 gene cause either Rett syndrome (RTT) or MECP2 duplication syndrome. Alterations in MECP2 levels have also been identified in patients with autism. To fully understand the significant role of MECP2 in regulating the development and function of the nervous system, it is important to study all aspects of MeCP2 function. Stimulus-induced MeCP2 phosphorylation has been shown to influence the proliferation and differentiation of neural progenitor cells, synaptic scaling, excitatory synaptogenesis, and animal behavior. However, all of the previous functional evidence is from studying phospho-dead mutations. In addition, the relationship between phosphorylation events at multiple sites on the MeCP2 protein is not well understood. Here, we report the generation of a phospho-mimic knockin Mecp2 mouse line. At the synaptic and behavioral levels, the phospho-mimic Mecp2 mice show phenotypes opposite to those observed in phospho-dead mutation at the same phosphorylation site. Moreover, we report opposite phenotypes between phospho-mutants of two sites on the MeCP2 protein. Our new data further confirm the functional significance of specific MeCP2 phosphorylation event and support the opposing regulatory role between different MeCP2 phosphorylation events. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29534967  Title: Mitochondrial dysfunction in dopaminergic neurons differentiated from exfoliated deciduous tooth-derived pulp stem cells of a child with Rett syndrome.  Rett syndrome is an X-linked neurodevelopmental disorder associated with psychomotor impairments, autonomic dysfunctions and autism. Patients with Rett syndrome have loss-of-function mutations in MECP2, the gene encoding methyl-CpG-binding protein 2 (MeCP2). Abnormal biogenic amine signaling and mitochondrial function have been found in patients with Rett syndrome; however, few studies have analyzed the association between these factors. This study investigated the functional relationships between mitochondria and the neuronal differentiation of the MeCP2-deficient stem cells from the exfoliated deciduous teeth of a child with Rett syndrome. An enrolled subject in this study was a 5-year-old girl carrying a large deletion that included the methyl-CpG-binding domain, transcriptional repression domain, and nuclear localization signal of MECP2. Using the single-cell isolation technique, we found that the two populations of MeCP2-expressing and MeCP2-deficient stem cells kept their MECP2 expression profiles throughout the stages of cell proliferation and neuronal differentiation in vitro. Neurite outgrowth and branching were attenuated in MeCP2-deficient dopaminergic neurons. MeCP2-deficient cells showed reduced mitochondrial membrane potential, ATP production, restricted mitochondrial distribution in neurites, and lower expression of a central mitochondrial fission factor, dynamin-related protein 1 than MeCP2-expressing cells. These data indicated that MeCP2-deficiency dysregulates the expression of mitochondrial factors required for the maturation of dopaminergic neurons. This study also provides insight into the pathogenic mechanism underlying dysfunction of the intracerebral dopaminergic signaling pathway in Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29333487  Title: Activation of the Medial Prefrontal Cortex Reverses Cognitive and Respiratory Symptoms in a Mouse Model of Rett Syndrome.  Rett syndrome (RTT) is a severe neurodevelopmental disorder caused by loss-of-function mutations in the gene encoding methyl-CpG-binding protein 2 (MeCP2; Amir et al., 1999), a transcriptional regulatory protein (Klose et al., 2005). Mouse models of RTT (Mecp2 mutants) exhibit excitatory hypoconnectivity in the medial prefrontal cortex (mPFC; Sceniak et al., 2015), a region critical for functions that are abnormal in RTT patients, ranging from learning and memory to regulation of visceral homeostasis (Riga et al., 2014). The present study was designed to test the hypothesis that increasing the activity of mPFC pyramidal neurons in heterozygous female Mecp2 mutants (Hets) would ameliorate RTT-like symptoms, including deficits in respiratory control and long-term retrieval of auditory conditioned fear. Selective activation of mPFC pyramidal neurons in adult animals was achieved by bilateral infection with an AAV8 vector expressing excitatory hm3D(Gq) DREADD (Designer Receptors Exclusively Activated by Designer Drugs) (Armbruster et al., 2007) under the control of the CamKIIa promoter. DREADD activation in Mecp2 Hets completely restored long-term retrieval of auditory conditioned fear, eliminated respiratory apneas, and reduced respiratory frequency variability to wild-type (Wt) levels. Reversal of respiratory symptoms following mPFC activation was associated with normalization of Fos protein levels, a marker of neuronal activity, in a subset of brainstem respiratory neurons. Thus, despite reduced levels of MeCP2 and severe neurological deficits, mPFC circuits in Het mice are sufficiently intact to generate normal behavioral output when pyramidal cell activity is increased. These findings highlight the contribution of mPFC hypofunction to the pathophysiology of RTT and raise the possibility that selective activation of cortical regions such as the mPFC could provide therapeutic benefit to RTT patients. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29274743  Title: ANKRD11 associated with intellectual disability and autism regulates dendrite differentiation via the BDNF/TrkB signaling pathway.  Haploinsufficiency of ANKRD11 due to deletion or truncation mutations causes KBG syndrome, a rare genetic disorder characterized by intellectual disability, autism spectrum disorder, and craniofacial abnormalities. However, little is known about the neurobiological role of ANKRD11 during brain development. Here we show that ANKRD11 regulates pyramidal neuron migration and dendritic differentiation in the developing mouse cerebral cortex. Using an in utero manipulation approach, we found that Ankrd11 knockdown delayed radial migration of cortical neurons. ANKRD11-deficient neurons displayed markedly reduced dendrite growth and branching as well as abnormal dendritic spine morphology. Ankrd11 knockdown suppressed acetylation of epigenetic molecules such as p53 and Histone H3. Furthermore, the mRNA levels of Trkb, Bdnf, and neurite growth-related genes were downregulated in ANKRD11-deficient cortical neurons. The Trkb promoter region was largely devoid of acetylated Histone H3 and p53, and was instead occupied with MeCP2 and DNMT1. Overexpression of TrkB rescued abnormal dendrite growth in these cells. Our findings demonstrate a novel role for ANKRD11 in neuron differentiation during brain development and suggest an epigenetic modification as a potential key molecular feature underlying KBG syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29203410  Title: Breathing disturbances in a model of Rett syndrome: A potential involvement of the glycine receptor α3 subunit?  The glycine receptor α3 subunit is known to be a target for cAMP/PKA-mediated phosphorylation and regulation. Mice that lack this subunit are apparently normal but the 5-HT1A-receptor mediated modulation of respiratory network activity is disturbed. Since the intracellular cAMP-concentration is reduced in mice that lack the transcriptional modulator methyl-CpG-binding protein 2 (MeCP2) gene, we aimed to test if the α3 subunit of the glycine receptor is involved in the development of the breathing phenotype of MeCP2-deficient mice (Mecp2-/y). Therefore, we generated a double knock-out mouse line that lacks both the Mecp2 gene as well as the gene (Glra3) for the α3 subunit of the ionotropic glycine receptor. As compared to WT and Glra3-/- mice, both Mecp2-/y mice and Mecp2-/y; Glra3-/- mice (DKO) showed a slower respiratory rate and a tendency towards higher numbers of apneas. Interestingly, the irregularity of the breathing was significantly reduced in DKO as compared to Mecp2-/y littermates. In the light of the unaltered survival of DKO mice, however, the contribution of the glycine receptor α3 subunit for development and progression of the breathing disturbances in the mouse model of Rett syndrome appears to be only of minor relevance. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29074463  Title: Disruption of AT-hook 1 domain in MeCP2 protein caused behavioral abnormality in mice.  MECP2 is the causative gene for autism spectrum disorders, including Rett syndrome, a regressive neurodevelopmental rare disease mainly occurring in girls. Except for the distinct methyl-CpG binding domain and the transcriptional repression domain in MeCP2, three AT-hook-like domains have recently been identified. Several mutations in AT-hook 1 domain have been reported in autism cases or Rett database. However, the role of AT-hook 1 domain is still unclear. In this study, we generated a mouse line carrying deletion of eight conserved amino acids in AT-hook 1 domain by clustered regularly interspaced short palindromic repeats (CRISPR)/Cas9 technology. Mecp2ΔAT-hook1/y mutant male mice exhibited low locomotor activity, motor incoordination and cognitive deficit. In addition, these mutant mice exhibited increased anxiety. Moreover, pain insensitivity was noted in the mutant males. However, the social interactions were unaffected in AT-hook 1 mutant mice. Thinner CA1 region of the hippocampus was observed in the mutant mice. On the molecular basis, Western blot analysis showed increased expression of mutant MeCP2 protein in the cortex. Additionally, several genes expressed specifically in inhibitory neurons were markedly changed in the cerebrum. Taken together, these data demonstrate that disruption of AT-hook 1 domain in MeCP2 caused behavioral abnormality in mice, which suggests that AT-hook 1 is a critical region for the function of MeCP2 protein. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28961504  Title: Deciphering MECP2-associated disorders: disrupted circuits and the hope for repair.  MECP2 is a critical gene for neural development, mutations or duplication of which led to severe neurodevelopmental disorders, such as Rett syndrome (RTT) and autism spectrum disorders (ASD). Extensive works during the past decade yield ample insights into the molecular and cellular functions of MeCP2 in neural development. Furthermore, genetic manipulations in Mecp2 mouse models strongly suggested that deficiency in synaptic plasticity and various behaviors of Mecp2 null or transgenic mice could be rescued in adulthood. Further studies elucidating neural circuits responsible for symptoms in MECP2-associated disorders in rodent and non-human primate models will shed light on the development of potential therapeutic interventions. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28835516  Title: An RNA interference screen identifies druggable regulators of MeCP2 stability.  Alterations in gene dosage due to copy number variation are associated with autism spectrum disorder, intellectual disability (ID), and other psychiatric disorders. The nervous system is so acutely sensitive to the dose of methyl-CpG-binding protein 2 (MeCP2) that even a twofold change in MeCP2 protein-either increased or decreased-results in distinct disorders with overlapping features including ID, autistic behavior, and severe motor dysfunction. Rett syndrome is caused by loss-of-function mutations in MECP2, whereas duplications spanning the MECP2 locus result in MECP2 duplication syndrome (MDS), which accounts for ~1% of X-linked ID. Despite evidence from mouse models that restoring MeCP2 can reverse the course of disease, there are currently no U.S. Food and Drug Administration-approved therapies available to clinically modulate MeCP2 abundance. We used a forward genetic screen against all known human kinases and phosphatases to identify druggable regulators of MeCP2 stability. Two putative modulators of MeCP2, HIPK2 (homeodomain-interacting protein kinase 2) and PP2A (protein phosphatase 2A), were validated as stabilizers of MeCP2 in vivo. Further, pharmacological inhibition of PP2A in vivo reduced MeCP2 in the nervous system and rescued both overexpression and motor abnormalities in a mouse model of MDS. Our findings reveal potential therapeutic targets for treating disorders of altered MECP2 dosage. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28785396  Title: Identification of autism-related MECP2 mutations by whole-exome sequencing and functional validation.  Methyl-CpG-binding protein-2 (MeCP2) is a critical regulator for neural development. Either loss- or gain-of-function leads to severe neurodevelopmental disorders, such as Rett syndrome (RTT) and autism spectrum disorder (ASD). We set out to screen for MECP2 mutations in patients of ASD and determine whether these autism-related mutations may compromise the proper function of MeCP2. Whole-exome sequencing was performed to screen MECP2 and other ASD candidate genes for 120 patients diagnosed with ASD. The parents of patients who were identified with MECP2 mutation were selected for further Sanger sequencing. Each patient accomplished the case report form including general information and clinical scales applied to assess their clinical features. Mouse cortical neurons and HEK-293 cells were cultured and transfected with MeCP2 wild-type (WT) or mutant to examine the function of autism-associated MeCP2 mutants. HEK-293 cells were used to examine the expression of MeCP2 mutant constructs with Western blot. Mouse cortical neurons were used to analyze neurites and axon outgrowth by immunofluorescence experiments. We identified three missense mutations of MECP2 from three autism patients by whole-exome sequencing: p.P152L (c.455C>T), p.P376S (c.1162C>T), and p.R294X (c.880C>T). Among these mutations, p.P152L and p.R294X were de novo mutations, whereas p.P376S was inherited maternally. The diagnosis of RTT was excluded in all three autism patients. Abnormalities of dendritic and axonal growth were found after autism-related MeCP2 mutants were expressed in mouse cortical neurons; suggesting that autism-related MECP2 mutations impair the proper development of neurons. Our study identified genetic mutations of the MECP2 gene in autism patients, which were previously considered to be associated primarily with RTT. This finding suggests that loss-of-function mutations of MECP2 may also lead to autism spectrum disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28743991  Title: Altered visual cortical processing in a mouse model of MECP2 duplication syndrome.  As an epigenetic modulator of gene expression, Methyl-CpG binding protein 2 (MeCP2) is essential for normal neurological function. Dysfunction of MeCP2 is associated with a variety of neurological disorders. MECP2 gene duplication in human causes neuropsychiatric symptoms such as mental retardation and autism. MeCP2 overexpression in mice results in neurobehavioural disorders, dendritic abnormalities, and synaptic defects. However, how gain of MeCP2 function influences cortical processing of sensory information remains unclear. In this study, we examined visual processing in a mouse model of MECP2 duplication syndrome (MECP2 Tg1 mouse) at 8 and 14 weeks, which were before and after the onset of behavioural symptoms, respectively. In vivo extracellular recordings from primary visual cortex (V1) showed that neurons in Tg1 mice at both adult ages preferred higher spatial frequencies (SFs) than those in wild-type (WT) littermate controls, and the semi-saturation contrasts of neurons were lower in Tg1 mice at 8 weeks but not at 14 weeks. Behavioural experiments showed that the performance for visual detection at high SFs and low contrasts was higher in MECP2 Tg1 mice. Thus, MeCP2 gain-of-function in mice leads to higher visual acuity and contrast sensitivity, both at the levels of cortical response and behavioural performance. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28674485  Title: The Pathophysiological Role of Microglia in Dynamic Surveillance, Phagocytosis and Structural Remodeling of the Developing CNS.  In vertebrates, during an early wave of hematopoiesis in the yolk sac between embryonic day E7.0 and E9.0, cells of mesodermal leaflet addressed to macrophage lineage enter in developing central nervous system (CNS) and originate the developing native microglial cells. Depending on the species, microglial cells represent 5-20% of glial cells resident in adult brain. Here, we briefly discuss some canonical functions of the microglia, i.e., cytokine secretion and functional transition from M1 to M2 phenotype. In addition, we review studies on the non-canonical functions of microglia such as regulation of phagocytosis, synaptic pruning, and sculpting postnatal neural circuits. In this latter context the contribution of microglia to some neurodevelopmental disorders is now well established. Nasu-Hakola (NHD) disease is considered a primary microgliopathy with alterations of the DNAX activation protein 12 (DAP12)-Triggering receptor expressed on myeloid cells 2 (TREM-2) signaling and removal of macromolecules and apoptotic cells followed by secondary microglia activation. In Rett syndrome Mecp2-/- microglia shows a substantial impairment of phagocytic ability, although the role of microglia is not yet clear. In a mouse model of Tourette syndrome (TS), microglia abnormalities have also been described, and deficient microglia-mediated neuroprotection is obvious. Here we review the role of microglial cells in neurodevelopmental disorders without inflammation and on the complex role of microglia in developing CNS. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28673581  Title: Environmental contributors to modulation of brain estrogen signaling and male gender bias in autism: A reply to the oral contraceptive use hypothesis by Strifert (2015).  Strifert has recently put forward an interesting hypothesis regarding the role of oral contraceptive (OC) use in mothers and risk of offspring autism spectrum disorder (ASD). First, the author reports that combined oral contraceptives (COCs), containing both estrogen and progesterone, were developed in the late 1950s and early 60s, which is a time-frame distinct from Leo Kanner's documentation of infantile ASD in 1943 that Strifert just briefly mentions. While this important temporal inconsistency of ASD origin does not invalidate the potential role of OC use in contributing to the rise of ASD, it does support the likely possibility of other environmental exposures at play. Second, the epigenetic basis of the hypothesis is that the endocrine-disrupting components (i.e., ethinylestradiol) of OC perturb estrogenic signaling in the fetal brain by triggering aberrant DNA methylation of the estrogen receptor β (ERβ) gene, and such methylation patterns may be imprinted to future generations and could theoretically increase subsequent ASD offspring risk. The premise of the hypothesis is challenged, however, with the recognition that MeCP2, a "reader" of DNA methylation sites, is not only associated with age-dependent alteration in ERβ in females but is also significantly reduced in ASD brain. Furthermore, Strifert does not clearly address how the OC hypothesis accounts for the male bias in ASD. Therefore, the purpose of this correspondence is to address these inconsistencies by proposing a hypothesis that challenges these points. That is, gestational exposure to the agricultural and combustion air pollutant, nitrous oxide (N2O), may be a leading contributor to the development of an ASD phenotype. The mechanism undergirding this hypothesis suggests that compensatory estrogenic activity may mitigate the effects of fetal N2O exposure and thereby confer a protective effect against ASD development in a sex-dependent manner (i.e., male bias in ASD). |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28535982  Title: Altered gene expression in early postnatal monoamine oxidase A knockout mice.  We reported previously that monoamine oxidase (MAO) A knockout (KO) mice show increased serotonin (5-hydroxytryptamine, 5-HT) levels and autistic-like behaviors characterized by repetitive behaviors, and anti-social behaviors. We showed that administration of the serotonin synthesis inhibitor para-chlorophenylalanine (pCPA) from post-natal day 1 (P1) through 7 (P7) in MAO A KO mice reduced the serotonin level to normal and reverses the repetitive behavior. These results suggested that the altered gene expression at P1 and P7 may be important for the autistic-like behaviors seen in MAO A KO mice and was studied here. In this study, Affymetrix mRNA array data for P1 and P7 MAO A KO mice were analyzed using Partek Genomics Suite and Ingenuity Pathways Analysis to identify genes differentially expressed versus wild-type and assess their functions and relationships. The number of significant differentially expressed genes (DEGs) varied with age: P1 (664) and P7 (3307) [false discovery rate (FDR) 1.5 for autism-linked genes and >2.0 for functionally categorized genes]. Eight autism-linked genes were differentially expressed in P1 (upregulated: NLGN3, SLC6A2; down-regulated: HTR2C, MET, ADSL, MECP2, ALDH5A1, GRIN3B) while four autism-linked genes were differentially expressed at P7 (upregulated: HTR2B; downregulated: GRIN2D, GRIN2B, CHRNA4). Many other genes involved in neurodevelopment, apoptosis, neurotransmission, and cognitive function were differentially expressed at P7 in MAO A KO mice. This result suggests that modulation of these genes by the increased serotonin may lead to neurodevelopmental alteration in MAO A KO mice and results in autistic-like behaviors. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28497075  Title: Development of a Novel AAV Gene Therapy Cassette with Improved Safety Features and Efficacy in a Mouse Model of Rett Syndrome.  Rett syndrome (RTT), caused by loss-of-function mutations in the MECP2 gene, is a neurological disorder characterized by severe impairment of motor and cognitive functions. The aim of this study was to investigate the impact of vector design, dosage, and delivery route on the efficacy and safety of gene augmentation therapy in mouse models of RTT. Our results show that AAV-mediated delivery of MECP2 to Mecp2 null mice by systemic administration, and utilizing a minimal endogenous promoter, was associated with a narrow therapeutic window and resulted in liver toxicity at higher doses. Lower doses of this vector significantly extended the survival of mice lacking MeCP2 or expressing a mutant T158M allele but had no impact on RTT-like neurological phenotypes. Modifying vector design by incorporating an extended Mecp2 promoter and additional regulatory 3' UTR elements significantly reduced hepatic toxicity after systemic administration. Moreover, direct cerebroventricular injection of this vector into neonatal Mecp2-null mice resulted in high brain transduction efficiency, increased survival and body weight, and an amelioration of RTT-like phenotypes. Our results show that controlling levels of MeCP2 expression in the liver is achievable through modification of the expression cassette. However, it also highlights the importance of achieving high brain transduction to impact the RTT-like phenotypes. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28439102  Title: MeCP2-regulated miRNAs control early human neurogenesis through differential effects on ERK and AKT signaling.  Rett syndrome (RTT) is an X-linked, neurodevelopmental disorder caused primarily by mutations in the methyl-CpG-binding protein 2 (MECP2) gene, which encodes a multifunctional epigenetic regulator with known links to a wide spectrum of neuropsychiatric disorders. Although postnatal functions of MeCP2 have been thoroughly investigated, its role in prenatal brain development remains poorly understood. Given the well-established importance of microRNAs (miRNAs) in neurogenesis, we employed isogenic human RTT patient-derived induced pluripotent stem cell (iPSC) and MeCP2 short hairpin RNA knockdown approaches to identify novel MeCP2-regulated miRNAs enriched during early human neuronal development. Focusing on the most dysregulated miRNAs, we found miR-199 and miR-214 to be increased during early brain development and to differentially regulate extracellular signal-regulated kinase (ERK)/mitogen-activated protein kinase and protein kinase B (PKB/AKT) signaling. In parallel, we characterized the effects on human neurogenesis and neuronal differentiation brought about by MeCP2 deficiency using both monolayer and three-dimensional (cerebral organoid) patient-derived and MeCP2-deficient neuronal culture models. Inhibiting miR-199 or miR-214 expression in iPSC-derived neural progenitors deficient in MeCP2 restored AKT and ERK activation, respectively, and ameliorated the observed alterations in neuronal differentiation. Moreover, overexpression of miR-199 or miR-214 in the wild-type mouse embryonic brains was sufficient to disturb neurogenesis and neuronal migration in a similar manner to Mecp2 knockdown. Taken together, our data support a novel miRNA-mediated pathway downstream of MeCP2 that influences neurogenesis via interactions with central molecular hubs linked to autism spectrum disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28367307  Title: Transcriptome analysis of microglia in a mouse model of Rett syndrome: differential expression of genes associated with microglia/macrophage activation and cellular stress.  Rett syndrome (RTT) is a severe, neurodevelopmental disorder primarily affecting girls, characterized by progressive loss of cognitive, social, and motor skills after a relatively brief period of typical development. It is usually due to de novo loss of function mutations in the X-linked gene, MeCP2, which codes for the gene expression and chromatin regulator, methyl-CpG binding protein 2. Although the behavioral phenotype appears to be primarily due to neuronal Mecp2 deficiency in mice, other cell types, including astrocytes and oligodendrocytes, also appear to contribute to some aspects of the RTT phenotype. In addition, microglia may also play a role. However, the effect of Mecp2 deficiency in microglia on RTT pathogenesis is controversial. In the current study, we applied whole transcriptome analysis using RNA-seq to gain insight into molecular pathways in microglia that might be dysregulated during the transition, in female mice heterozygous for an Mecp2-null allele (Mecp2+/-; Het), from the pre-phenotypic (5 weeks) to the phenotypic phases (24 weeks). We found a significant overlap in differentially expressed genes (DEGs) with genes involved in regulating the extracellular matrix, and those that are activated or inhibited when macrophages and microglia are stimulated towards the M1 and M2 activation states. However, the M1- and M2-associated genes were different in the 5- and 24-week samples. In addition, a substantial decrease in the expression of nine members of the heat shock protein (HSP) family was found in the 5-week samples, but not at 24 weeks. These findings suggest that microglia from pre-phenotypic and phenotypic female mice are activated in a manner different from controls and that pre-phenotypic female mice may have alterations in their capacity to response to heat stress and other stressors that function through the HSP pathway. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28317218  Title: Rodent models of genetic and chromosomal variations in psychiatric disorders.  Elucidating the molecular basis of complex human psychiatric disorders is challenging due to the multitude of factors that underpin these disorders. Genetic and chromosomal changes are two factors that have been suggested to be involved in psychiatric disorders. Indeed, numerous risk loci have been identified in autism spectrum disorders, schizophrenia, and related psychiatric disorders. Here, we introduce genetic animal models that disturb excitatory-inhibitory balance in the brain and animal models mirroring human chromosomal abnormalities, both of which may be implicated in autism spectrum disorder pathophysiology. In addition, we discuss recent unique translational research using rodent models, such as Cntnap2 knockout mouse, Mecp2 mutant mouse, Pick1 knockout mouse, and neonatal ventral hippocampal lesion rat. By using these models, several types of drugs are administered during the developmental period to see the effect on psychotic symptoms and neural activities in adults. The accumulating evidence from recent animal studies provides an informative intervention strategy as a translational research. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28258509  Title: Distinct Defects in Spine Formation or Pruning in Two Gene Duplication Mouse Models of Autism.  Autism spectrum disorder (ASD) encompasses a complex set of developmental neurological disorders, characterized by deficits in social communication and excessive repetitive behaviors. In recent years, ASD is increasingly being considered as a disease of the synapse. One main type of genetic aberration leading to ASD is gene duplication, and several mouse models have been generated mimicking these mutations. Here, we studied the effects of MECP2 duplication and human chromosome 15q11-13 duplication on synaptic development and neural circuit wiring in the mouse sensory cortices. We showed that mice carrying MECP2 duplication had specific defects in spine pruning, while the 15q11-13 duplication mouse model had impaired spine formation. Our results demonstrate that spine pathology varies significantly between autism models and that distinct aspects of neural circuit development may be targeted in different ASD mutations. Our results further underscore the importance of gene dosage in normal development and function of the brain. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28229923  Title: Epigenetic regulation of RELN and GAD1 in the frontal cortex (FC) of autism spectrum disorder (ASD) subjects.  Both Reelin (RELN) and glutamate decarboxylase 67 (GAD1) have been implicated in the pathophysiology of Autism Spectrum Disorders (ASD). We have previously shown that both mRNAs are reduced in the cerebella (CB) of ASD subjects through a mechanism that involves increases in the amounts of MECP2 binding to the corresponding promoters. In the current study, we examined the expression of RELN, GAD1, GAD2, and several other mRNAs implicated in this disorder in the frontal cortices (FC) of ASD and CON subjects. We also focused on the role that epigenetic processes play in the regulation of these genes in ASD brain. Our goal is to better understand the molecular basis for the down-regulation of genes expressed in GABAergic neurons in ASD brains. We measured mRNA levels corresponding to selected GABAergic genes using qRT-PCR in RNA isolated from both ASD and CON groups. We determined the extent of binding of MECP2 and DNMT1 repressor proteins by chromatin immunoprecipitation (ChIP) assays. The amount of 5-methylcytosine (5mC) and 5-hydroxymethylcytosine (5hmC) present in the promoters of the target genes was quantified by methyl DNA immunoprecipitation (MeDIP) and hydroxyl MeDIP (hMeDIP). We detected significant reductions in the mRNAs associated with RELN and GAD1 and significant increases in mRNAs encoding the Ten-eleven Translocation (TET) enzymes 1, 2, and 3. We also detected increased MECP2 and DNMT1 binding to the corresponding promoter regions of GAD1, RELN, and GAD2. Interestingly, there were decreased amounts of 5mC at both promoters and little change in 5hmC content in these same DNA fragments. Our data demonstrate that RELN, GAD1, and several other genes selectively expressed in GABAergic neurons, are down-regulated in post-mortem ASD FC. In addition, we observed increased DNMT1 and MECP2 binding at the corresponding promoters of these genes. The finding of increased MECP2 binding to the RELN, GAD1 and GAD2 promoters, with reduced amounts of 5mC and unchanged amounts of 5hmC present in these regions, suggests the possibility that DNMT1 interacts with and alters MECP2 binding properties to selected promoters. Comparisons between data obtained from the FC with CB studies showed some common themes between brain regions which are discussed. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28220356  Title: Overexpression of LINE-1 Retrotransposons in Autism Brain.  Long interspersed nuclear elements-1 (LINE-1 or L1) are mobile DNA sequences that are capable of duplication and insertion (retrotransposition) within the genome. Recently, retrotransposition of L1 was shown to occur within human brain leading to somatic mosaicism in hippocampus and cerebellum. Because unregulated L1 activity can promote genomic instability and mutagenesis, multiple mechanisms including epigenetic chromatin condensation have evolved to effectively repress L1 expression. Nonetheless, L1 expression has been shown to be increased in patients with Rett syndrome and schizophrenia. Based on this evidence and our reports of oxidative stress and epigenetic dysregulation in autism cerebellum, we sought to determine whether L1 expression was increased in autism brain. The results indicated that L1 expression was significantly elevated in the autism cerebellum but not in BA9, BA22, or BA24. The binding of repressive MeCP2 and histone H3K9me3 to L1 sequences was significantly lower in autism cerebellum suggesting that relaxation of epigenetic repression may have contributed to increased expression. Further, the increase in L1 expression was inversely correlated with glutathione redox status consistent with reports indicating that L1 expression is increased under pro-oxidant conditions. Finally, the expression of transcription factor FOXO3, sensor of oxidative stress, was significantly increased and positively associated with L1 expression and negatively associated with glutathione redox status. While these novel results are an important first step, future understanding of the contribution of elevated L1 expression to neuronal CNVs and genomic instability in autism will depend on emerging cell-specific genomic technologies, a challenge that warrants future investigation. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28139724  Title: Accumulated quiescent neural stem cells in adult hippocampus of the mouse model for the MECP2 duplication syndrome.  Duplications of Methyl CpG binding protein 2 (MECP2) -containing segments lead to the MECP2 duplication syndrome, in which severe autistic symptoms were identified. Whether adult neurogenesis may play a role in pathogenesis of autism and the role of MECP2 on state determination of adult neural stem cells (NSCs) remain largely unclear. Using a MECP2 transgenic (TG) mouse model for the MECP2 duplication syndrome, we found that adult hippocampal quiescent NSCs were significantly accumulated in TG mice comparing to wild type (WT) mice, the neural progenitor cells (NPCs) were reduced and the neuroblasts were increased in adult hippocampi of MECP2 TG mice. Interestingly, we found that parvalbumin (PV) positive interneurons were significantly decreased in MECP2 TG mice, which were critical for determining fates of adult hippocampal NSCs between the quiescence and activation. In summary, we found that MeCP2 plays a critical role in regulating fate determination of adult NSCs. These evidences further suggest that abnormal development of NSCs may play a role in the pathogenesis of the MECP2 duplication syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28093257  Title: Selective preservation of cholinergic MeCP2 rescues specific Rett-syndrome-like phenotypes in MeCP2stop mice.  RTT is a neurodevelopmental disorder characterized by growth regression, motor dysfunction, stereotypic hand movements, and autism features. Typical Rett syndrome (RTT) is predominantly caused by mutations in X-linked MeCP2 gene which encodes methyl-CpG-binding protein 2 (MeCP2). The brain-abundant MeCP2 protein mainly functions as a transcriptional regulator for neurodevelopment-associated genes. Specific functions of MeCP2 in certain neuron types remain to be known. Although cholinergic system is an important modulating system in brain, how MeCP2 in cholinergic neurons contribute to RTT has not been clearly understood. Here we use a mouse model with selectively activated endogenous MeCP2 in cholinergic neurons in otherwise MeCP2stop mice to determine the cholinergic MeCP2 effects on rescuing the RTT-like phenotypes. We found cholinergic MeCP2 preservation could reverse some aspects of the RTT-like phenotypes in mice including hypolocomotion and increased anxiety level, and delay the onset of underweight, instead of improving the hypersocial abnormality and the poor general conditions such as short lifespan, low brain weight, and increasing severity score. Our findings suggest that selective activation of cholinergic MeCP2 is sufficient to reverse the locomotor impairment and increased anxiety-like behaviors at least in early symptomatic stage, supporting future development of RTT therapies associated with cholinergic system. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28041965  Title: Analysis of gene expression in Ca2+-dependent activator protein for secretion 2 (Cadps2) knockout cerebellum using GeneChip and KEGG pathways.  In the mouse cerebellum, Ca2+-dependent activator protein for secretion 2 (CADPS2, CAPS2) is involved in regulated secretion from dense-core vesicles (DCVs), which contain neuropeptides including brain-derived neurotrophic factor (BDNF) and neurotrophin-3 (NT-3). Capds2 knockout (KO) mice show impaired cerebellar development in addition to autistic-like behavioral phenotypes. To understand the molecular impact caused by loss of Capds2, we analyzed gene expression profiles in the Capds2 KO cerebellum using a GeneChip microarray and the KEGG Pathway database. Significant differential expression was observed in 1211 of 22,690 (5.34%) genes represented on the chip. The expression levels of exocytosis-related genes (Stx5a, Syt6), genes encoding secretory (Fgf2, Fgf4, Edn2) and synaptic proteins (Grin2b, Gabbr1), neurotrophin signaling-associated genes (Sos1, Shc1, Traf6, Psen2), and a gene for Rett syndrome (Mecp2) were significantly changed. Taken together, these results suggest that deregulated gene expression caused by loss of Capds2 may cause developmental deficits and/or pathological symptoms, resulting in autistic-like phenotypes. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27881780  Title: Negative Allosteric Modulation of mGluR5 Partially Corrects Pathophysiology in a Mouse Model of Rett Syndrome.  Rett syndrome (RTT) is caused by mutations in the gene encoding methyl-CpG binding protein 2 (MECP2), an epigenetic regulator of mRNA transcription. Here, we report a test of the hypothesis of shared pathophysiology of RTT and fragile X, another monogenic cause of autism and intellectual disability. In fragile X, the loss of the mRNA translational repressor FMRP leads to exaggerated protein synthesis downstream of metabotropic glutamate receptor 5 (mGluR5). We found that mGluR5- and protein-synthesis-dependent synaptic plasticity were similarly altered in area CA1 of Mecp2 KO mice. CA1 pyramidal cell-type-specific, genome-wide profiling of ribosome-bound mRNAs was performed in wild-type and Mecp2 KO hippocampal CA1 neurons to reveal the MeCP2-regulated "translatome." We found significant overlap between ribosome-bound transcripts overexpressed in the Mecp2 KO and FMRP mRNA targets. These tended to encode long genes that were functionally related to either cytoskeleton organization or the development of neuronal connectivity. In the Fmr1 KO mouse, chronic treatment with mGluR5-negative allosteric modulators (NAMs) has been shown to ameliorate many mutant phenotypes by correcting excessive protein synthesis. In Mecp2 KO mice, we found that mGluR5 NAM treatment significantly reduced the level of overexpressed ribosome-associated transcripts, particularly those that were also FMRP targets. Some Rett phenotypes were also ameliorated by treatment, most notably hippocampal cell size and lifespan. Together, these results suggest a potential mechanistic link between MeCP2-mediated transcription regulation and mGluR5/FMRP-mediated protein translation regulation through coregulation of a subset of genes relevant to synaptic functions. Altered regulation of synaptic protein synthesis has been hypothesized to contribute to the pathophysiology that underlies multiple forms of intellectual disability and autism spectrum disorder. Here, we show in a mouse model of Rett syndrome (Mecp2 KO) that metabotropic glutamate receptor 5 (mGluR5)- and protein-synthesis-dependent synaptic plasticity are abnormal in the hippocampus. We found that a subset of ribosome-bound mRNAs was aberrantly upregulated in hippocampal CA1 neurons of Mecp2 KO mice, that these significantly overlapped with FMRP direct targets and/or SFARI human autism genes, and that chronic treatment of Mecp2 KO mice with an mGluR5-negative allosteric modulator tunes down upregulated ribosome-bound mRNAs and partially improves mutant mice phenotypes. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27604541  Title: FMRP Expression Levels in Mouse Central Nervous System Neurons Determine Behavioral Phenotype.  Fragile X mental retardation protein (FMRP) is absent or highly reduced in Fragile X Syndrome, a genetic disorder causing cognitive impairment and autistic behaviors. Previous proof-of-principle studies have demonstrated that restoring FMRP in the brain using viral vectors can improve pathological abnormalities in mouse models of fragile X. However, unlike small molecule drugs where the dose can readily be adjusted during treatment, viral vector-based biological therapeutic drugs present challenges in terms of achieving optimal dosing and expression levels. The objective of this study was to investigate the consequences of expressing varying levels of FMRP selectively in neurons of Fmr1 knockout and wild-type (WT) mice. A wide range of neuronal FMRP transgene levels was achieved in individual mice after intra-cerebroventricular administration of adeno-associated viral vectors coding for FMRP. In all treated knockout mice, prominent FMRP transgene expression was observed in forebrain structures, whereas lower levels were present in more caudal regions of the brain. Reduced levels of the synaptic protein PSD-95, elevated levels of the transcriptional modulator MeCP2, and abnormal motor activity, anxiety, and acoustic startle responses in Fmr1 knockout mice were fully or partially rescued after expression of FMRP at about 35-115% of WT expression, depending on the brain region examined. In the WT mouse, moderate FMRP over-expression of up to about twofold had little or no effect on PSD-95 and MeCP2 levels or on behavioral endophenotypes. In contrast, excessive over-expression in the Fmr1 knockout mouse forebrain (approximately 2.5-6-fold over WT) induced pathological motor hyperactivity and suppressed the startle response relative to WT mice. These results delineate a range of FMRP expression levels in the central nervous system that confer phenotypic improvement in fragile X mice. Collectively, these findings are pertinent to the development of long-term curative gene therapy strategies for treating Fragile X Syndrome and other neurodevelopmental disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27672368  Title: GlyT2-Dependent Preservation of MECP2-Expression in Inhibitory Neurons Improves Early Respiratory Symptoms but Does Not Rescue Survival in a Mouse Model of Rett Syndrome.  Mutations in methyl-CpG-binding protein 2 (MECP2) gene have been shown to manifest in a neurodevelopmental disorder that is called Rett syndrome. A typical problem that occurs during development is a disturbance of breathing. To address the role of inhibitory neurons, we generated a mouse line that restores MECP2 in inhibitory neurons in the brainstem by crossbreeding a mouse line that expresses the Cre-recombinase (Cre) in inhibitory neurons under the control of the glycine transporter 2 (GlyT2, slc6a5) promotor (GlyT2-Cre) with a mouse line that has a floxed-stop mutation of the Mecp2 gene (Mecp2 (stop/y)). Unrestrained whole-body-plethysmography at postnatal day P60 revealed a low respiratory rate and prolonged respiratory pauses in Mecp2 (stop/y) mice. In contrast, GlyT2-Cre positive Mecp2 (stop/y) mice (Cre(+) ; Mecp2 (stop/y)) showed greatly improved respiration and were indistinguishable from wild type littermates. These data support the concept that alterations in inhibitory neurons are important for the development of the respiratory phenotype in Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27623092  Title: Kynurenine pathway metabolism following prenatal KMO inhibition and in Mecp2+/- mice, using liquid chromatography-tandem mass spectrometry.  To quantify the full range of tryptophan metabolites along the kynurenine pathway, a liquid chromatography - tandem mass spectrometry method was developed and used to analyse brain extracts of rodents treated with the kynurenine-3-mono-oxygenase (KMO) inhibitor Ro61-8048 during pregnancy. There were significant increases in the levels of kynurenine, kynurenic acid, anthranilic acid and 3-hydroxy-kynurenine (3-HK) in the maternal brain after 5 h but not 24 h, while the embryos exhibited high levels of kynurenine, kynurenic acid and anthranilic acid after 5 h which were maintained at 24 h post-treatment. At 24 h there was also a strong trend to an increase in quinolinic acid levels (P = 0.055). No significant changes were observed in any of the other kynurenine metabolites. The results confirm the marked increase in the accumulation of some neuroactive kynurenines when KMO is inhibited, and re-emphasise the potential importance of changes in anthranilic acid. The prolonged duration of metabolite accumulation in the embryo brains indicates a trapping of compounds within the embryonic CNS independently of maternal levels. When brains were examined from young mice heterozygous for the meCP2 gene - a potential model for Rett syndrome - no differences were noted from control mice, suggesting that the proposed roles for kynurenines in autism spectrum disorder are not relevant to Rett syndrome, supporting its recognition as a distinct, independent, condition. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27399968  Title: Haploinsufficiency of MeCP2-interacting transcriptional co-repressor SIN3A causes mild intellectual disability by affecting the development of cortical integrity.  Numerous genes are associated with neurodevelopmental disorders such as intellectual disability and autism spectrum disorder (ASD), but their dysfunction is often poorly characterized. Here we identified dominant mutations in the gene encoding the transcriptional repressor and MeCP2 interactor switch-insensitive 3 family member A (SIN3A; chromosome 15q24.2) in individuals who, in addition to mild intellectual disability and ASD, share striking features, including facial dysmorphisms, microcephaly and short stature. This phenotype is highly related to that of individuals with atypical 15q24 microdeletions, linking SIN3A to this microdeletion syndrome. Brain magnetic resonance imaging showed subtle abnormalities, including corpus callosum hypoplasia and ventriculomegaly. Intriguingly, in vivo functional knockdown of Sin3a led to reduced cortical neurogenesis, altered neuronal identity and aberrant corticocortical projections in the developing mouse brain. Together, our data establish that haploinsufficiency of SIN3A is associated with mild syndromic intellectual disability and that SIN3A can be considered to be a key transcriptional regulator of cortical brain development. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27328325  Title: Manipulations of MeCP2 in glutamatergic neurons highlight their contributions to Rett and other neurological disorders.  Many postnatal onset neurological disorders such as autism spectrum disorders (ASDs) and intellectual disability are thought to arise largely from disruption of excitatory/inhibitory homeostasis. Although mouse models of Rett syndrome (RTT), a postnatal neurological disorder caused by loss-of-function mutations in MECP2, display impaired excitatory neurotransmission, the RTT phenotype can be largely reproduced in mice simply by removing MeCP2 from inhibitory GABAergic neurons. To determine what role excitatory signaling impairment might play in RTT pathogenesis, we generated conditional mouse models with Mecp2 either removed from or expressed solely in glutamatergic neurons. MeCP2 deficiency in glutamatergic neurons leads to early lethality, obesity, tremor, altered anxiety-like behaviors, and impaired acoustic startle response, which is distinct from the phenotype of mice lacking MeCP2 only in inhibitory neurons. These findings reveal a role for excitatory signaling impairment in specific neurobehavioral abnormalities shared by RTT and other postnatal neurological disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27323888  Title: Brain phosphorylation of MeCP2 at serine 164 is developmentally regulated and globally alters its chromatin association.  MeCP2 is a transcriptional regulator whose functional alterations are responsible for several autism spectrum and mental disorders. Post-translational modifications (PTMs), and particularly differential phosphorylation, modulate MeCP2 function in response to diverse stimuli. Understanding the detailed role of MeCP2 phosphorylation is thus instrumental to ascertain how MeCP2 integrates the environmental signals and directs its adaptive transcriptional responses. The evolutionarily conserved serine 164 (S164) was found phosphorylated in rodent brain but its functional role has remained uncharacterized. We show here that phosphorylation of S164 in brain is dynamically regulated during neuronal maturation. S164 phosphorylation highly impairs MeCP2 binding to DNA in vitro and largely affects its nucleosome binding and chromatin affinity in vivo. Strikingly, the chromatin-binding properties of the global MeCP2 appear also extensively altered during the course of brain maturation. Functional assays reveal that proper temporal regulation of S164 phosphorylation controls the ability of MeCP2 to regulate neuronal morphology. Altogether, our results support the hypothesis of a complex PTM-mediated functional regulation of MeCP2 potentially involving a still poorly characterized epigenetic code. Furthermore, they demonstrate the relevance of the Intervening Domain of MeCP2 for binding to DNA. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27313794  Title: Characterization of Rett Syndrome-like phenotypes in Mecp2-knockout rats.  Rett Syndrome (RTT) is a neurodevelopmental disease caused by the disruption of the MECP2 gene. Several mouse models of RTT have been developed with Mecp2 disruptions. Although the mouse models are widely used in RTT research, results obtained need to be validated in other species. Therefore, we performed these studies to characterize phenotypes of a novel Mecp2 (-/Y) rat model and compared them with the Mecp2 (tm1.1Bird) mouse model of RTT. RTT-like phenotypes were systematically studied and compared between Mecp2 (-/Y) rats and Mecp2 (-/Y) mice. In-cage conditions of the rats were monitored. Grip strength and spontaneous locomotion were used to evaluate the motor function. Three-chamber test was performed to show autism-type behaviors. Breathing activity was recorded with the plethysmograph. Individual neurons in the locus coeruleus (LC) were studied in the whole-cell current clamp. The lifespan of the rats was determined with their survival time. Mecp2 (-/Y) rats displayed growth retardation, malocclusion, and lack of movements, while hindlimb clasping was not seen. They had weaker forelimb grip strength and a lower rate of locomotion than the WT littermates. Defects in social interaction with other rats were obvious. Breathing frequency variation and apnea in the null rats were significantly higher than in the WT. LC neurons in the null rats showed excessive firing activity. A half of the null rats died in 2 months. Most of the RTT-like symptoms were comparable to those seen in Mecp2 (-/Y) mice, while some appeared more or less severe. The findings that most RTT-like symptoms exist in the rat model with moderate variations and differences from the mouse models support the usefulness of both Mecp2 (-/Y) rodent models. The novel Mecp2 (-/Y) rat model recapitulated numerous RTT-like symptoms as Mecp2 (-/Y) mouse models did, which makes it a valuable alternative model in the RTT studies when the body size matters. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27293187  Title: Peripheral Mechanosensory Neuron Dysfunction Underlies Tactile and Behavioral Deficits in Mouse Models of ASDs.  Patients with autism spectrum disorders (ASDs) commonly experience aberrant tactile sensitivity, yet the neural alterations underlying somatosensory dysfunction and the extent to which tactile deficits contribute to ASD characteristics are unknown. We report that mice harboring mutations in Mecp2, Gabrb3, Shank3, and Fmr1 genes associated with ASDs in humans exhibit altered tactile discrimination and hypersensitivity to gentle touch. Deletion of Mecp2 or Gabrb3 in peripheral somatosensory neurons causes mechanosensory dysfunction through loss of GABAA receptor-mediated presynaptic inhibition of inputs to the CNS. Remarkably, tactile defects resulting from Mecp2 or Gabrb3 deletion in somatosensory neurons during development, but not in adulthood, cause social interaction deficits and anxiety-like behavior. Restoring Mecp2 expression exclusively in the somatosensory neurons of Mecp2-null mice rescues tactile sensitivity, anxiety-like behavior, and social interaction deficits, but not lethality, memory, or motor deficits. Thus, mechanosensory processing defects contribute to anxiety-like behavior and social interaction deficits in ASD mouse models. PAPERCLIP. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27080430  Title: Expression of nuclear Methyl-CpG binding protein 2 (Mecp2) is dependent on neuronal stimulation and application of Insulin-like growth factor 1.  Methyl-CpG binding protein 2 (MECP2) is a chromosome-binding protein that regulates the development and maintenance of brain circuits. Altered function of the protein product of MECP2 plays an important role in the etiology of many neurodevelopmental disorders. Mutations involving a loss of function are implicated in the etiology of Rett syndrome, intellectual disability, psychosis and severe encephalopathy. Conversely, MECP2 duplications have been identified in autism and intellectual disability. MECP2 action is dependent on neuronal function, as the DNA binding is modulated by activity, and it is phosphorylated in response to stimulation. Although MECP2 is considered a major risk factor for neurodevelopmental disorders, and it is a mediator of activity-dependent mechanisms, the expression levels in response to neuronal activity have never been measured. We studied the expression of Mecp2 protein and RNA in mice neuronal cultures in response to different stimulation conditions and in the presence of insulin-like growth factor1 (IGF1): a growth factor involved in brain development and plasticity. The stimulation protocols were selected according to their ability to induce different forms of synaptic plasticity: rapid depolarization, feed-forward plasticity (LTP, LTD) and feedback forms of plasticity (TTX, KCl). We find a significant reduction of Mecp2 protein nuclear expression in neurons in response to stimuli that induce a potentiation of neuronal response, suggesting that Mecp2 protein expression is modulated by neuronal activation. Application of IGF1 to the cultures induces an increase in the expression of Mecp2 transcript and nuclear Mecp2 protein in neurons. These results show that Mecp2 is responsive to neuronal stimulation and IGF1, and different stimuli have different effects on Mecp2 expression; this differential response may have downstream effects on functional mechanisms regulating brain development and plasticity. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26949739  Title: Identification of Human Neuronal Protein Complexes Reveals Biochemical Activities and Convergent Mechanisms of Action in Autism Spectrum Disorders.  The prevalence of autism spectrum disorders (ASDs) is rapidly growing, yet its molecular basis is poorly understood. We used a systems approach in which ASD candidate genes were mapped onto the ubiquitous human protein complexes and the resulting complexes were characterized. The studies revealed the role of histone deacetylases (HDAC1/2) in regulating the expression of ASD orthologs in the embryonic mouse brain. Proteome-wide screens for the co-complexed subunits with HDAC1 and six other key ASD proteins in neuronal cells revealed a protein interaction network, which displayed preferential expression in fetal brain development, exhibited increased deleterious mutations in ASD cases, and were strongly regulated by FMRP and MECP2 causal for Fragile X and Rett syndromes, respectively. Overall, our study reveals molecular components in ASD, suggests a shared mechanism between the syndromic and idiopathic forms of ASDs, and provides a systems framework for analyzing complex human diseases. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26936821  Title: mGlu5 positive allosteric modulation normalizes synaptic plasticity defects and motor phenotypes in a mouse model of Rett syndrome.  Rett syndrome (RS) is a neurodevelopmental disorder that shares many symptomatic and pathological commonalities with idiopathic autism. Alterations in protein synthesis-dependent synaptic plasticity (PSDSP) are a hallmark of a number of syndromic forms of autism; in the present work, we explore the consequences of disruption and rescue of PSDSP in a mouse model of RS. We report that expression of a key regulator of synaptic protein synthesis, the metabotropic glutamate receptor 5 (mGlu5) protein, is significantly reduced in both the brains of RS model mice and in the motor cortex of human RS autopsy samples. Furthermore, we demonstrate that reduced mGlu5 expression correlates with attenuated DHPG-induced long-term depression in the hippocampus of RS model mice, and that administration of a novel mGlu5 positive allosteric modulator (PAM), termed VU0462807, can rescue synaptic plasticity defects. Additionally, treatment of Mecp2-deficient mice with VU0462807 improves motor performance (open-field behavior and gait dynamics), corrects repetitive clasping behavior, as well as normalizes cued fear-conditioning defects. Importantly, due to the rationale drug discovery approach used in its development, our novel mGlu5 PAM improves RS phenotypes and synaptic plasticity defects without evoking the overt adverse effects commonly associated with potentiation of mGlu5 signaling (i.e. seizures), or affecting cardiorespiratory defects in RS model mice. These findings provide strong support for the continued development of mGlu5 PAMs as potential therapeutic agents for use in RS, and, more broadly, for utility in idiopathic autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26929363  Title: Excitatory synapses are stronger in the hippocampus of Rett syndrome mice due to altered synaptic trafficking of AMPA-type glutamate receptors.  Deficits in long-term potentiation (LTP) at central excitatory synapses are thought to contribute to cognitive impairments in neurodevelopmental disorders associated with intellectual disability and autism. Using the methyl-CpG-binding protein 2 (Mecp2) knockout (KO) mouse model of Rett syndrome, we show that naïve excitatory synapses onto hippocampal pyramidal neurons of symptomatic mice have all of the hallmarks of potentiated synapses. Stronger Mecp2 KO synapses failed to undergo LTP after either theta-burst afferent stimulation or pairing afferent stimulation with postsynaptic depolarization. On the other hand, basal synaptic strength and LTP were not affected in slices from younger presymptomatic Mecp2 KO mice. Furthermore, spine synapses in pyramidal neurons from symptomatic Mecp2 KO are larger and do not grow in size or incorporate GluA1 subunits after electrical or chemical LTP. Our data suggest that LTP is occluded in Mecp2 KO mice by already potentiated synapses. The higher surface levels of GluA1-containing receptors are consistent with altered expression levels of proteins involved in AMPA receptor trafficking, suggesting previously unidentified targets for therapeutic intervention for Rett syndrome and other MECP2-related disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26918155  Title: Recent advances in understanding synaptic abnormalities in Rett syndrome.  Rett syndrome is an extremely disabling X-linked nervous system disorder that mainly affects girls in early childhood and causes autism-like behavior, severe intellectual disability, seizures, sleep disturbances, autonomic instability, and other disorders due to mutations in the MeCP2 (methyl CpG-binding protein 2) transcription factor. The disorder targets synapses and synaptic plasticity and has been shown to disrupt the balance between glutamate excitatory synapses and GABAergic inhibitory synapses. In fact, it can be argued that Rett syndrome is primarily a disorder of synaptic plasticity and that agents that can correct this imbalance may have beneficial effects on brain development. This review briefly summarizes the link between disrupted synaptic plasticity mechanisms and Rett syndrome and early clinical trials that aim to target these abnormalities to improve the outcome for these severely disabled children. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26908602  Title: Acute and crucial requirement for MeCP2 function upon transition from early to late adult stages of brain maturation.  Germline mutations in the X-linked gene, methyl-CpG-binding protein 2 (MECP2), underlie most cases of Rett syndrome (RTT), an autism spectrum disorder affecting approximately one in 10 000 female live births. The disease is characterized in affected girls by a latent appearance of symptoms between 12 and 18 months of age while boys usually die before the age of two. The nature of the latency is not known, but RTT-like phenotypes are recapitulated in mouse models, even when MeCP2 is removed at different postnatal stages, including juvenile and adolescent stages. Unexpectedly, here, we show that within a very brief developmental window, between 10 (adolescent) and 15 (adult) weeks after birth, symptom initiation and progression upon removal of MeCP2 in male mice transitions from 3 to 4 months to only several days, followed by lethality. We further show that this accelerated development of RTT phenotype and lethality occur at the transition to adult stage (15 weeks of age) and persists thereafter. Importantly, within this abbreviated time frame of days, the brain acquires dramatic anatomical, cellular and molecular abnormalities, typical of classical RTT. This study reveals a new postnatal developmental stage, which coincides with full-brain maturation, where the structure/function of the brain is extremely sensitive to levels of MeCP2 and loss of MeCP2 leads to precipitous collapse of the neuronal networks and incompatibility with life within days. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26808898  Title: Autism-like behaviours and germline transmission in transgenic monkeys overexpressing MeCP2.  Methyl-CpG binding protein 2 (MeCP2) has crucial roles in transcriptional regulation and microRNA processing. Mutations in the MECP2 gene are found in 90% of patients with Rett syndrome, a severe developmental disorder with autistic phenotypes. Duplications of MECP2-containing genomic segments cause the MECP2 duplication syndrome, which shares core symptoms with autism spectrum disorders. Although Mecp2-null mice recapitulate most developmental and behavioural defects seen in patients with Rett syndrome, it has been difficult to identify autism-like behaviours in the mouse model of MeCP2 overexpression. Here we report that lentivirus-based transgenic cynomolgus monkeys (Macaca fascicularis) expressing human MeCP2 in the brain exhibit autism-like behaviours and show germline transmission of the transgene. Expression of the MECP2 transgene was confirmed by western blotting and immunostaining of brain tissues of transgenic monkeys. Genomic integration sites of the transgenes were characterized by a deep-sequencing-based method. As compared to wild-type monkeys, MECP2 transgenic monkeys exhibited a higher frequency of repetitive circular locomotion and increased stress responses, as measured by the threat-related anxiety and defensive test. The transgenic monkeys showed less interaction with wild-type monkeys within the same group, and also a reduced interaction time when paired with other transgenic monkeys in social interaction tests. The cognitive functions of the transgenic monkeys were largely normal in the Wisconsin general test apparatus, although some showed signs of stereotypic cognitive behaviours. Notably, we succeeded in generating five F1 offspring of MECP2 transgenic monkeys by intracytoplasmic sperm injection with sperm from one F0 transgenic monkey, showing germline transmission and Mendelian segregation of several MECP2 transgenes in the F1 progeny. Moreover, F1 transgenic monkeys also showed reduced social interactions when tested in pairs, as compared to wild-type monkeys of similar age. Together, these results indicate the feasibility and reliability of using genetically engineered non-human primates to study brain disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26733678  Title: KCC2 rescues functional deficits in human neurons derived from patients with Rett syndrome.  Rett syndrome is a severe form of autism spectrum disorder, mainly caused by mutations of a single gene methyl CpG binding protein 2 (MeCP2) on the X chromosome. Patients with Rett syndrome exhibit a period of normal development followed by regression of brain function and the emergence of autistic behaviors. However, the mechanism behind the delayed onset of symptoms is largely unknown. Here we demonstrate that neuron-specific K(+)-Cl(-) cotransporter2 (KCC2) is a critical downstream gene target of MeCP2. We found that human neurons differentiated from induced pluripotent stem cells from patients with Rett syndrome showed a significant deficit in KCC2 expression and consequently a delayed GABA functional switch from excitation to inhibition. Interestingly, overexpression of KCC2 in MeCP2-deficient neurons rescued GABA functional deficits, suggesting an important role of KCC2 in Rett syndrome. We further identified that RE1-silencing transcriptional factor, REST, a neuronal gene repressor, mediates the MeCP2 regulation of KCC2. Because KCC2 is a slow onset molecule with expression level reaching maximum later in development, the functional deficit of KCC2 may offer an explanation for the delayed onset of Rett symptoms. Our studies suggest that restoring KCC2 function in Rett neurons may lead to a potential treatment for Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26733386  Title: Monogenic mouse models of autism spectrum disorders: Common mechanisms and missing links.  Autism spectrum disorders (ASDs) present unique challenges in the fields of genetics and neurobiology because of the clinical and molecular heterogeneity underlying these disorders. Genetic mutations found in ASD patients provide opportunities to dissect the molecular and circuit mechanisms underlying autistic behaviors using animal models. Ongoing studies of genetically modified models have offered critical insight into possible common mechanisms arising from different mutations, but links between molecular abnormalities and behavioral phenotypes remain elusive. The challenges encountered in modeling autism in mice demand a new analytic paradigm that integrates behavioral assessment with circuit-level analysis in genetically modified models with strong construct validity. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26606849  Title: High-Frequency Stimulation at the Subthalamic Nucleus Suppresses Excessive Self-Grooming in Autism-Like Mouse Models.  Approximately one quarter of individuals with an autism spectrum disorder (ASD) display self-injurious behavior (SIB) ranging from head banging to self-directed biting and punching. Sometimes, these behaviors are extreme and unresponsive to pharmacological and behavioral therapies. We have found electroconvulsive therapy (ECT) can produce life-changing results, with more than 90% suppression of SIB frequency. However, these patients typically require frequent maintenance ECT (mECT), as often as every 5 days, to sustain the improvement gained during the acute course. Long-term consequences of such frequent mECT started as early as childhood in some cases are unknown. Accordingly, there is a need for alternative forms of chronic stimulation for these patients. To explore the feasibility of deep brain stimulation (DBS) for intractable SIB seen in some patients with an ASD, we utilized two genetically distinct mouse models demonstrating excessive self-grooming, namely the Viaat-Mecp2(-/y) and Shank3B(-/-) lines, and administered high-frequency stimulation (HFS) via implanted electrodes at the subthalamic nucleus (STN-HFS). We found that STN-HFS significantly suppressed excessive self-grooming in both genetic lines. Suppression occurs both acutely when stimulation is switched on, and persists for several days after HFS is stopped. This effect was not explained by a change in locomotor activity, which was unaffected by STN-HFS. Likewise, social interaction deficits were not corrected by STN-HFS. Our data show STN-HFS suppresses excessive self-grooming in two autism-like mouse models, raising the possibility DBS might be used to treat intractable SIB associated with ASDs. Further studies are required to explore the circuitry engaged by STN-HFS, as well as other potential stimulation sites. Such studies might also yield clues about pathways, which could be modulated by non-invasive stimulatory techniques. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26605526  Title: Reversal of phenotypes in MECP2 duplication mice using genetic rescue or antisense oligonucleotides.  Copy number variations have been frequently associated with developmental delay, intellectual disability and autism spectrum disorders. MECP2 duplication syndrome is one of the most common genomic rearrangements in males and is characterized by autism, intellectual disability, motor dysfunction, anxiety, epilepsy, recurrent respiratory tract infections and early death. The broad range of deficits caused by methyl-CpG-binding protein 2 (MeCP2) overexpression poses a daunting challenge to traditional biochemical-pathway-based therapeutic approaches. Accordingly, we sought strategies that directly target MeCP2 and are amenable to translation into clinical therapy. The first question that we addressed was whether the neurological dysfunction is reversible after symptoms set in. Reversal of phenotypes in adult symptomatic mice has been demonstrated in some models of monogenic loss-of-function neurological disorders, including loss of MeCP2 in Rett syndrome, indicating that, at least in some cases, the neuroanatomy may remain sufficiently intact so that correction of the molecular dysfunction underlying these disorders can restore healthy physiology. Given the absence of neurodegeneration in MECP2 duplication syndrome, we propose that restoration of normal MeCP2 levels in MECP2 duplication adult mice would rescue their phenotype. By generating and characterizing a conditional Mecp2-overexpressing mouse model, here we show that correction of MeCP2 levels largely reverses the behavioural, molecular and electrophysiological deficits. We also reduced MeCP2 using an antisense oligonucleotide strategy, which has greater translational potential. Antisense oligonucleotides are small, modified nucleic acids that can selectively hybridize with messenger RNA transcribed from a target gene and silence it, and have been successfully used to correct deficits in different mouse models. We find that antisense oligonucleotide treatment induces a broad phenotypic rescue in adult symptomatic transgenic MECP2 duplication mice (MECP2-TG), and corrected MECP2 levels in lymphoblastoid cells from MECP2 duplication patients in a dose-dependent manner. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26590342  Title: Loss of MeCP2 in Parvalbumin-and Somatostatin-Expressing Neurons in Mice Leads to Distinct Rett Syndrome-like Phenotypes.  Inhibitory neurons are critical for proper brain function, and their dysfunction is implicated in several disorders, including autism, schizophrenia, and Rett syndrome. These neurons are heterogeneous, and it is unclear which subtypes contribute to specific neurological phenotypes. We deleted Mecp2, the mouse homolog of the gene that causes Rett syndrome, from the two most populous subtypes, parvalbumin-positive (PV+) and somatostatin-positive (SOM+) neurons. Loss of MeCP2 partially impairs the affected neuron, allowing us to assess the function of each subtype without profound disruption of neuronal circuitry. We found that mice lacking MeCP2 in either PV+ or SOM+ neurons have distinct, non-overlapping neurological features: mice lacking MeCP2 in PV+ neurons developed motor, sensory, memory, and social deficits, whereas those lacking MeCP2 in SOM+ neurons exhibited seizures and stereotypies. Our findings indicate that PV+ and SOM+ neurons contribute complementary aspects of the Rett phenotype and may have modular roles in regulating specific behaviors. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26575259  Title: Maternal immune activation induces GAD1 and GAD2 promoter remodeling in the offspring prefrontal cortex.  Maternal infection during pregnancy increases the risk of neurodevelopmental disorders in the offspring. In addition to its influence on other neuronal systems, this early-life environmental adversity has been shown to negatively affect cortical γ-aminobutyric acid (GABA) functions in adult life, including impaired prefrontal expression of enzymes required for GABA synthesis. The underlying molecular processes, however, remain largely unknown. In the present study, we explored whether epigenetic modifications represent a mechanism whereby maternal infection during pregnancy can induce such GABAergic impairments in the offspring. We used an established mouse model of prenatal immune challenge that is based on maternal treatment with the viral mimetic poly(I:C). We found that prenatal immune activation increased prefrontal levels of 5-methylated cytosines (5mC) and 5-hydroxymethylated cytosines (5hmC) in the promoter region of GAD1, which encodes the 67-kDa isoform of the GABA-synthesising enzyme glutamic acid decarboxylase (GAD67). The early-life challenge also increased 5mC levels at the promoter region of GAD2, which encodes the 65-kDa GAD isoform (GAD65). These effects were accompanied by elevated GAD1 and GAD2 promoter binding of methyl CpG-binding protein 2 (MeCP2) and by reduced GAD67 and GAD65 mRNA expression. Moreover, the epigenetic modifications at the GAD1 promoter correlated with prenatal infection-induced impairments in working memory and social interaction. Our study thus highlights that hypermethylation of GAD1 and GAD2 promoters may be an important molecular mechanism linking prenatal infection to presynaptic GABAergic impairments and associated behavioral and cognitive abnormalities in the offspring. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26237041  Title: MECP2 disorders: from the clinic to mice and back.  Two severe, progressive neurological disorders characterized by intellectual disability, autism, and developmental regression, Rett syndrome and MECP2 duplication syndrome, result from loss and gain of function, respectively, of the same critical gene, methyl-CpG-binding protein 2 (MECP2). Neurons acutely require the appropriate dose of MECP2 to function properly but do not die in its absence or overexpression. Instead, neuronal dysfunction can be reversed in a Rett syndrome mouse model if MeCP2 function is restored. Thus, MECP2 disorders provide a unique window into the delicate balance of neuronal health, the power of mouse models, and the importance of chromatin regulation in mature neurons. In this Review, we will discuss the clinical profiles of MECP2 disorders, the knowledge acquired from mouse models of the syndromes, and how that knowledge is informing current and future clinical studies. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26181491  Title: Mutations in JMJD1C are involved in Rett syndrome and intellectual disability.  Autism spectrum disorders are associated with defects in social response and communication that often occur in the context of intellectual disability. Rett syndrome is one example in which epilepsy, motor impairment, and motor disturbance may co-occur. Mutations in histone demethylases are known to occur in several of these syndromes. Herein, we aimed to identify whether mutations in the candidate histone demethylase JMJD1C (jumonji domain containing 1C) are implicated in these disorders. We performed the mutational and functional analysis of JMJD1C in 215 cases of autism spectrum disorders, intellectual disability, and Rett syndrome without a known genetic defect. We found seven JMJD1C variants that were not present in any control sample (~ 6,000) and caused an amino acid change involving a different functional group. From these, two de novo JMJD1C germline mutations were identified in a case of Rett syndrome and in a patient with intellectual disability. The functional study of the JMJD1C mutant Rett syndrome patient demonstrated that the altered protein had abnormal subcellular localization, diminished activity to demethylate the DNA damage-response protein MDC1, and reduced binding to MECP2. We confirmed that JMJD1C protein is widely expressed in brain regions and that its depletion compromises dendritic activity. Our findings indicate that mutations in JMJD1C contribute to the development of Rett syndrome and intellectual disability.Genet Med 18 1, 378-385. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26158416  Title: Cerebellar associative sensory learning defects in five mouse autism models.  Sensory integration difficulties have been reported in autism, but their underlying brain-circuit mechanisms are underexplored. Using five autism-related mouse models, Shank3+/ΔC, Mecp2(R308/Y), Cntnap2-/-, L7-Tsc1 (L7/Pcp2(Cre)::Tsc1(flox/+)), and patDp(15q11-13)/+, we report specific perturbations in delay eyeblink conditioning, a form of associative sensory learning requiring cerebellar plasticity. By distinguishing perturbations in the probability and characteristics of learned responses, we found that probability was reduced in Cntnap2-/-, patDp(15q11-13)/+, and L7/Pcp2(Cre)::Tsc1(flox/+), which are associated with Purkinje-cell/deep-nuclear gene expression, along with Shank3+/ΔC. Amplitudes were smaller in L7/Pcp2(Cre)::Tsc1(flox/+) as well as Shank3+/ΔC and Mecp2(R308/Y), which are associated with granule cell pathway expression. Shank3+/ΔC and Mecp2(R308/Y) also showed aberrant response timing and reduced Purkinje-cell dendritic spine density. Overall, our observations are potentially accounted for by defects in instructed learning in the olivocerebellar loop and response representation in the granule cell pathway. Our findings indicate that defects in associative temporal binding of sensory events are widespread in autism mouse models. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26098633  Title: MeCP2 Affects Skeletal Muscle Growth and Morphology through Non Cell-Autonomous Mechanisms.  Rett syndrome (RTT) is an autism spectrum disorder mainly caused by mutations in the X-linked MECP2 gene and affecting roughly 1 out of 10.000 born girls. Symptoms range in severity and include stereotypical movement, lack of spoken language, seizures, ataxia and severe intellectual disability. Notably, muscle tone is generally abnormal in RTT girls and women and the Mecp2-null mouse model constitutively reflects this disease feature. We hypothesized that MeCP2 in muscle might physiologically contribute to its development and/or homeostasis, and conversely its defects in RTT might alter the tissue integrity or function. We show here that a disorganized architecture, with hypotrophic fibres and tissue fibrosis, characterizes skeletal muscles retrieved from Mecp2-null mice. Alterations of the IGF-1/Akt/mTOR pathway accompany the muscle phenotype. A conditional mouse model selectively depleted of Mecp2 in skeletal muscles is characterized by healthy muscles that are morphologically and molecularly indistinguishable from those of wild-type mice raising the possibility that hypotonia in RTT is mainly, if not exclusively, mediated by non-cell autonomous effects. Our results suggest that defects in paracrine/endocrine signaling and, in particular, in the GH/IGF axis appear as the major cause of the observed muscular defects. Remarkably, this is the first study describing the selective deletion of Mecp2 outside the brain. Similar future studies will permit to unambiguously define the direct impact of MeCP2 on tissue dysfunctions. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26029044  Title: Potential primary roles of glial cells in the mechanisms of psychiatric disorders.  While neurons have long been considered the major player in multiple brain functions such as perception, emotion, and memory, glial cells have been relegated to a far lesser position, acting as merely a "glue" to support neurons. Multiple lines of recent evidence, however, have revealed that glial cells such as oligodendrocytes, astrocytes, and microglia, substantially impact on neuronal function and activities and are significantly involved in the underlying pathobiology of psychiatric disorders. Indeed, a growing body of evidence indicates that glial cells interact extensively with neurons both chemically (e.g., through neurotransmitters, neurotrophic factors, and cytokines) and physically (e.g., through gap junctions), supporting a role for these cells as likely significant modifiers not only of neural function in brain development but also disease pathobiology. Since questions have lingered as to whether glial dysfunction plays a primary role in the biology of neuropsychiatric disorders or a role related solely to their support of neuronal physiology in these diseases, informative and predictive animal models have been developed over the last decade. In this article, we review recent findings uncovered using glia-specific genetically modified mice with which we can evaluate both the causation of glia dysfunction and its potential role in neuropsychiatric disorders such as autism and schizophrenia. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25979331  Title: Methyl CpG Binding Protein 2 Gene Disruption Augments Tonic Currents of γ-Aminobutyric Acid Receptors in Locus Coeruleus Neurons: IMPACT ON NEURONAL EXCITABILITY AND BREATHING.  People with Rett syndrome and mouse models show autonomic dysfunction involving the brain stem locus coeruleus (LC). Neurons in the LC of Mecp2-null mice are overly excited, likely resulting from a defect in neuronal intrinsic membrane properties and a deficiency in GABA synaptic inhibition. In addition to the synaptic GABA receptors, there is a group of GABAA receptors (GABAARs) that is located extrasynaptically and mediates tonic inhibition. Here we show evidence for augmentation of the extrasynaptic GABAARs in Mecp2-null mice. In brain slices, exposure of LC neurons to GABAAR agonists increased tonic currents that were blocked by GABAAR antagonists. With 10 μm GABA, the bicuculline-sensitive tonic currents were ∼4-fold larger in Mecp2-null LC neurons than in the WT. Single-cell PCR analysis showed that the δ subunit, the principal subunit of extrasynaptic GABAARs, was present in LC neurons. Expression levels of the δ subunit were ∼50% higher in Mecp2-null neurons than in the WT. Also increased in expression in Mecp2-null mice was another extrasynaptic GABAAR subunit, α6, by ∼4-fold. The δ subunit-selective agonists 4,5,6,7-tetrahydroisoxazolo[5,4-c]pyridin-3-ol hydrochloride and 4-chloro-N-[2-(2-thienyl)imidazo[1,2-a]pyridin-3-yl]]benzamide activated the tonic GABAA currents in LC neurons and reduced neuronal excitability to a greater degree in Mecp2-null mice than in the WT. Consistent with these findings, in vivo application of 4,5,6,7-tetrahydroisoxazolo[5,4-c]pyridin-3-ol hydrochloride alleviated breathing abnormalities of conscious Mecp2-null mice. These results suggest that extrasynaptic GABAARs seem to be augmented with Mecp2 disruption, which may be a compensatory response to the deficiency in GABAergic synaptic inhibition and allows control of neuronal excitability and breathing abnormalities. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25875630  Title: Circadian cycle-dependent MeCP2 and brain chromatin changes.  Methyl CpG binding protein 2 (MeCP2) is a chromosomal protein of the brain, very abundant especially in neurons, where it plays an important role in the regulation of gene expression. Hence it has the potential to be affected by the mammalian circadian cycle. We performed expression analyses of mice brain frontal cortices obtained at different time points and we found that the levels of MeCP2 are altered circadianly, affecting overall organization of brain chromatin and resulting in a circadian-dependent regulation of well-stablished MeCP2 target genes. Furthermore, this data suggests that alterations of MeCP2 can be responsible for the sleeping disorders arising from pathological stages, such as in autism and Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25861995  Title: Rett Syndrome: Reaching for Clinical Trials.  Rett syndrome (RTT) is a syndromic autism spectrum disorder caused by loss-of-function mutations in MECP2. The methyl CpG binding protein 2 binds methylcytosine and 5-hydroxymethycytosine at CpG sites in promoter regions of target genes, controlling their transcription by recruiting co-repressors and co-activators. Several preclinical studies in mouse models have identified rational molecular targets for drug therapies aimed at correcting the underlying neural dysfunction. These targeted therapies are increasingly translating into human clinical trials. In this review, we present an overview of RTT and describe the current state of preclinical studies in methyl CpG binding protein 2-based mouse models, as well as current clinical trials in individuals with RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25762136  Title: Disruption of DNA-methylation-dependent long gene repression in Rett syndrome.  Disruption of the MECP2 gene leads to Rett syndrome (RTT), a severe neurological disorder with features of autism. MECP2 encodes a methyl-DNA-binding protein that has been proposed to function as a transcriptional repressor, but despite numerous mouse studies examining neuronal gene expression in Mecp2 mutants, no clear model has emerged for how MeCP2 protein regulates transcription. Here we identify a genome-wide length-dependent increase in gene expression in MeCP2 mutant mouse models and human RTT brains. We present evidence that MeCP2 represses gene expression by binding to methylated CA sites within long genes, and that in neurons lacking MeCP2, decreasing the expression of long genes attenuates RTT-associated cellular deficits. In addition, we find that long genes as a population are enriched for neuronal functions and selectively expressed in the brain. These findings suggest that mutations in MeCP2 may cause neurological dysfunction by specifically disrupting long gene expression in the brain. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25728234  Title: Gadd45b is an epigenetic regulator of juvenile social behavior and alters local pro-inflammatory cytokine production in the rodent amygdala.  Precise regulation of the epigenome during perinatal development is critical to the formation of species-typical behavior later in life. Recent data suggests that Gadd45b facilitates active DNA demethylation by recruiting proteins involved in base excision repair (BER), which will catalyze substitution of 5-methyl-cytosine (5mC) for an unmodified cytosine. While a role for Gadd45b has been implicated in both hippocampal and amygdalar learning tasks, to the best of our knowledge, no study has been done investigating the involvement of Gadd45b in neurodevelopmental programming of social behavior. To address this, we used a targeted siRNA delivery approach to transiently knock down Gadd45b expression in the neonatal rat amygdala. We chose to examine social behavior in the juvenile period, as social deficits associated with neurodevelopmental disorders tend to emerge in humans at an equivalent age. We find that neonatal Gadd45b knock-down results in altered juvenile social behavior and reduced expression of several genes implicated in psychiatric disorders, including methyl-CpG-binding protein 2 (MeCP2), Reelin, and brain derived neurotrophic factor (BDNF). We furthermore report a novel role for Gadd45b in the programmed expression of α2-adrenoceptor (Adra2a). Consistent with Gadd45b's role in the periphery, we also observed changes in the expression of pro-inflammatory cytokines interleukin-6 (Il-6) and interleukin-1beta (Il-1beta) in the amygdala, which could potentially mediate or exacerbate effects of Gadd45b knockdown on the organization of social behavior. These data suggest a prominent role for Gadd45b in the epigenetic programming of complex juvenile social interactions, and may provide insight into the etiology of juvenile behavioral disorders such as ADHD, autism, and/or schizophrenia. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25669997  Title: Dysregulated brain immunity and neurotrophin signaling in Rett syndrome and autism spectrum disorders.  Rett syndrome is a neurodevelopmental disorder, which occurs in about 1:15,000 females and presents with neurologic and communication defects. It is transmitted as an X-linked dominant linked to mutations of the methyl-CpG-binding protein (MeCP2), a gene transcription suppressor, but its definitive pathogenesis is unknown thus hindering development of effective treatments. Almost half of children with Rett syndrome also have behavioral symptoms consistent with those of autism spectrum disorders (ASDs). PubMed was searched (2005-2014) using the terms: allergy, atopy, brain, brain-derived neurotrophic factor (BDNF), corticotropin-releasing hormone (CRH), cytokines, gene mutations, inflammation, mast cells (MCs), microglia, mitochondria, neurotensin (NT), neurotrophins, seizures, stress, and treatment. There are a number of intriguing differences and similarities between Rett syndrome and ASDs. Rett syndrome occurs in females, while ASDs more often in males, and the former has neurologic disabilities unlike ASDs. There is evidence of dysregulated immune system early in life in both conditions. Lack of microglial phagocytosis and decreased levels of BDNF appear to distinguish Rett syndrome from ASDs, in which there is instead microglia activation and/or proliferation and possibly defective BDNF signaling. Moreover, brain mast cell (MC) activation and focal inflammation may be more prominent in ASDs than Rett syndrome. The flavonoid luteolin blocks microglia and MC activation, provides BDNF-like activity, reverses Rett phenotype in mouse models, and has a significant benefit in children with ASDs. Appropriate formulations of luteolin or other natural molecules may be useful in the treatment of Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25662825  Title: Mechanisms of Functional Hypoconnectivity in the Medial Prefrontal Cortex of Mecp2 Null Mice.  Frontal cortical dysfunction is thought to contribute to cognitive and behavioral features of autism spectrum disorders; however, underlying mechanisms are poorly understood. The present study sought to define how loss of Mecp2, the gene mutated in Rett syndrome (RTT), disrupts function in the murine medial prefrontal cortex (mPFC) using acute brain slices and behavioral testing. Compared with wildtype, pyramidal neurons in the Mecp2 null mPFC exhibit significant reductions in excitatory postsynaptic currents, the duration of excitatory UP-states, evoked population activity, and the ratio of NMDA:AMPA currents, as well as an increase in the relative fraction of NR2B currents. These functional changes are associated with reductions in the density of excitatory dendritic spines, the ratio of vesicular glutamate to GABA transporters and GluN1 expression. In contrast to recent reports on circuit defects in other brain regions, we observed no effect of Mecp2 loss on inhibitory synaptic currents or expression of the inhibitory marker parvalbumin. Consistent with mPFC hypofunction, Mecp2 nulls exhibit respiratory dysregulation in response to behavioral arousal. Our data highlight functional hypoconnectivity in the mPFC as a potential substrate for behavioral disruption in RTT and other disorders associated with reduced expression of Mecp2 in frontal cortical regions. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25578949  Title: Dendritic spine dysgenesis in autism related disorders.  The activity-dependent structural and functional plasticity of dendritic spines has led to the long-standing belief that these neuronal compartments are the subcellular sites of learning and memory. Of relevance to human health, central neurons in several neuropsychiatric illnesses, including autism related disorders, have atypical numbers and morphologies of dendritic spines. These so-called dendritic spine dysgeneses found in individuals with autism related disorders are consistently replicated in experimental mouse models. Dendritic spine dysgenesis reflects the underlying synaptopathology that drives clinically relevant behavioral deficits in experimental mouse models, providing a platform for testing new therapeutic approaches. By examining molecular signaling pathways, synaptic deficits, and spine dysgenesis in experimental mouse models of autism related disorders we find strong evidence for mTOR to be a critical point of convergence and promising therapeutic target. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25385366  Title: Modeling non-syndromic autism and the impact of TRPC6 disruption in human neurons.  An increasing number of genetic variants have been implicated in autism spectrum disorders (ASDs), and the functional study of such variants will be critical for the elucidation of autism pathophysiology. Here, we report a de novo balanced translocation disruption of TRPC6, a cation channel, in a non-syndromic autistic individual. Using multiple models, such as dental pulp cells, induced pluripotent stem cell (iPSC)-derived neuronal cells and mouse models, we demonstrate that TRPC6 reduction or haploinsufficiency leads to altered neuronal development, morphology and function. The observed neuronal phenotypes could then be rescued by TRPC6 complementation and by treatment with insulin-like growth factor-1 or hyperforin, a TRPC6-specific agonist, suggesting that ASD individuals with alterations in this pathway may benefit from these drugs. We also demonstrate that methyl CpG binding protein-2 (MeCP2) levels affect TRPC6 expression. Mutations in MeCP2 cause Rett syndrome, revealing common pathways among ASDs. Genetic sequencing of TRPC6 in 1041 ASD individuals and 2872 controls revealed significantly more nonsynonymous mutations in the ASD population, and identified loss-of-function mutations with incomplete penetrance in two patients. Taken together, these findings suggest that TRPC6 is a novel predisposing gene for ASD that may act in a multiple-hit model. This is the first study to use iPSC-derived human neurons to model non-syndromic ASD and illustrate the potential of modeling genetically complex sporadic diseases using such cells. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25309341  Title: Dendritic spine dysgenesis in Rett syndrome.  Spines are small cytoplasmic extensions of dendrites that form the postsynaptic compartment of the majority of excitatory synapses in the mammalian brain. Alterations in the numerical density, size, and shape of dendritic spines have been correlated with neuronal dysfunction in several neurological and neurodevelopmental disorders associated with intellectual disability, including Rett syndrome (RTT). RTT is a progressive neurodevelopmental disorder associated with intellectual disability that is caused by loss of function mutations in the transcriptional regulator methyl CpG-binding protein 2 (MECP2). Here, we review the evidence demonstrating that principal neurons in RTT individuals and Mecp2-based experimental models exhibit alterations in the number and morphology of dendritic spines. We also discuss the exciting possibility that signaling pathways downstream of brain-derived neurotrophic factor (BDNF), which is transcriptionally regulated by MeCP2, offer promising therapeutic options for modulating dendritic spine development and plasticity in RTT and other MECP2-associated neurodevelopmental disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25299635  Title: Anaplerotic triheptanoin diet enhances mitochondrial substrate use to remodel the metabolome and improve lifespan, motor function, and sociability in MeCP2-null mice.  Rett syndrome (RTT) is an autism spectrum disorder (ASD) caused by mutations in the X-linked MECP2 gene that encodes methyl-CpG binding protein 2 (MeCP2). Symptoms range in severity and include psychomotor disabilities, seizures, ataxia, and intellectual disability. Symptom onset is between 6-18 months of age, a critical period of brain development that is highly energy-dependent. Notably, patients with RTT have evidence of mitochondrial dysfunction, as well as abnormal levels of the adipokines leptin and adiponectin, suggesting overall metabolic imbalance. We hypothesized that one contributor to RTT symptoms is energy deficiency due to defective nutrient substrate utilization by the TCA cycle. This energy deficit would lead to a metabolic imbalance, but would be treatable by providing anaplerotic substrates to the TCA cycle to enhance energy production. We show that dietary therapy with triheptanoin significantly increased longevity and improved motor function and social interaction in male mice hemizygous for Mecp2 knockout. Anaplerotic therapy in Mecp2 knockout mice also improved indicators of impaired substrate utilization, decreased adiposity, increased glucose tolerance and insulin sensitivity, decreased serum leptin and insulin, and improved mitochondrial morphology in skeletal muscle. Untargeted metabolomics of liver and skeletal muscle revealed increases in levels of TCA cycle intermediates with triheptanoin diet, as well as normalizations of glucose and fatty acid biochemical pathways consistent with the improved metabolic phenotype in Mecp2 knockout mice on triheptanoin. These results suggest that an approach using dietary supplementation with anaplerotic substrate is effective in improving symptoms and metabolic health in RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25232122  Title: Cell-type-specific repression by methyl-CpG-binding protein 2 is biased toward long genes.  Mutations in methyl-CpG-binding protein 2 (MeCP2) cause Rett syndrome and related autism spectrum disorders (Amir et al., 1999). MeCP2 is believed to be required for proper regulation of brain gene expression, but prior microarray studies in Mecp2 knock-out mice using brain tissue homogenates have revealed only subtle changes in gene expression (Tudor et al., 2002; Nuber et al., 2005; Jordan et al., 2007; Chahrour et al., 2008). Here, by profiling discrete subtypes of neurons we uncovered more dramatic effects of MeCP2 on gene expression, overcoming the "dilution problem" associated with assaying homogenates of complex tissues. The results reveal misregulation of genes involved in neuronal connectivity and communication. Importantly, genes upregulated following loss of MeCP2 are biased toward longer genes but this is not true for downregulated genes, suggesting MeCP2 may selectively repress long genes. Because genes involved in neuronal connectivity and communication, such as cell adhesion and cell-cell signaling genes, are enriched among longer genes, their misregulation following loss of MeCP2 suggests a possible etiology for altered circuit function in Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25180573  Title: Prenatal maternal immune activation causes epigenetic differences in adolescent mouse brain.  Epigenetic processes such as DNA methylation have been implicated in the pathophysiology of neurodevelopmental disorders including schizophrenia and autism. Epigenetic changes can be induced by environmental exposures such as inflammation. Here we tested the hypothesis that prenatal inflammation, a recognized risk factor for schizophrenia and related neurodevelopmental conditions, alters DNA methylation in key brain regions linked to schizophrenia, namely the dopamine rich striatum and endocrine regulatory centre, the hypothalamus. DNA methylation across highly repetitive elements (long interspersed element 1 (LINE1) and intracisternal A-particles (IAPs)) were used to proxy global DNA methylation. We also investigated the Mecp2 gene because it regulates transcription of LINE1 and has a known association with neurodevelopmental disorders. Brain tissue was harvested from 6 week old offspring of mice exposed to the viral analog PolyI:C or saline on gestation day 9. We used Sequenom EpiTYPER assay to quantitatively analyze differences in DNA methylation at IAPs, LINE1 elements and the promoter region of Mecp2. In the hypothalamus, prenatal exposure to PolyI:C caused significant global DNA hypomethylation (t=2.44, P=0.019, PolyI:C mean 69.67%, saline mean 70.19%), especially in females, and significant hypomethylation of the promoter region of Mecp2, (t=3.32, P=0.002; PolyI:C mean 26.57%, saline mean 34.63%). IAP methylation was unaltered. DNA methylation in the striatum was not significantly altered. This study provides the first experimental evidence that exposure to inflammation during prenatal life is associated with epigenetic changes, including Mecp2 promoter hypomethylation. This suggests that environmental and genetic risk factors associated with neurodevelopmental disorders may act upon similar pathways. This is important because epigenetic changes are potentially modifiable and their investigation may open new avenues for treatment. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25131546  Title: Epigenetic mechanisms in autism spectrum disorder.  Autism spectrum disorder (ASD) is a neurodevelopmental condition characterized by impaired social interactions, language deficits, as well as restrictive or repetitive behaviors. ASD is clinically heterogeneous with a complex etiopathogenesis which may be conceptualized as a dynamic interplay between heterogeneous environmental cues and predisposing genetic factors involving complex epigenetic mechanisms. Inherited and de novo copy number variants provide novel information regarding genes contributing to ASD. Epigenetic marks are stable, yet potentially reversible, chromatin modifications that alter gene expression profiles by locally changing the degree of nucleosomal compaction, thereby opening or closing promoter access to the transcriptional machinery. Here, we review progress on studies designed to provide a better understanding of how epigenetic mechanisms impact transcriptional programs operative in the brain that contribute to ASD. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25088363  Title: Sensory integration in mouse insular cortex reflects GABA circuit maturation.  Insular cortex (IC) contributes to a variety of complex brain functions, such as communication, social behavior, and self-awareness through the integration of sensory, emotional, and cognitive content. How the IC acquires its integrative properties remains unexplored. We compared the emergence of multisensory integration (MSI) in the IC of behaviorally distinct mouse strains. While adult C57BL/6 mice exhibited robust MSI, this capacity was impaired in the inbred BTBR T+tf/J mouse model of idiopathic autism. The deficit reflected weakened γ-aminobutyric acid (GABA) circuits and compromised postnatal pruning of cross-modal input. Transient pharmacological enhancement by diazepam in BTBR mice during an early sensitive period rescued inhibition and integration in the adult IC. Moreover, impaired MSI was common across three other monogenic models (GAD65, Shank3, and Mecp2 knockout mice) displaying behavioral phenotypes and parvalbumin-circuit abnormalities. Our findings offer developmental insight into a key neural circuit relevant to neuropsychiatric conditions like schizophrenia and autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25082535  Title: MeCP2: multifaceted roles in gene regulation and neural development.  Methyl-CpG-binding protein 2 (MeCP2) is a classic methylated-DNA-binding protein, dysfunctions of which lead to various neurodevelopmental disorders such as Rett syndrome and autism spectrum disorder. Initially recognized as a transcriptional repressor, MeCP2 has been studied extensively and its functions have been expanded dramatically in the past two decades. Recently, it was found to be involved in gene regulation at the post-transcriptional level. MeCP2 represses nuclear microRNA processing by interacting directly with the Drosha/DGCR8 complex. In addition to its multifaceted functions, MeCP2 is remarkably modulated by posttranslational modifications such as phosphorylation, SUMOylation, and acetylation, providing more regulatory dimensions to its functions. The role of MeCP2 in the central nervous system has been studied extensively, from neurons to glia. Future investigations combining molecular, cellular, and physiological methods are necessary for defining the roles of MeCP2 in the brain and developing efficient treatments for MeCP2-related brain disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25009110  Title: Alterations in the cholinergic system of brain stem neurons in a mouse model of Rett syndrome.  Rett syndrome is an autism-spectrum disorder resulting from mutations to the X-linked gene, methyl-CpG binding protein 2 (MeCP2), which causes abnormalities in many systems. It is possible that the body may develop certain compensatory mechanisms to alleviate the abnormalities. The norepinephrine system originating mainly in the locus coeruleus (LC) is defective in Rett syndrome and Mecp2-null mice. LC neurons are subject to modulation by GABA, glutamate, and acetylcholine (ACh), providing an ideal system to test the compensatory hypothesis. Here we show evidence for potential compensatory modulation of LC neurons by post- and presynaptic ACh inputs. We found that the postsynaptic currents of nicotinic ACh receptors (nAChR) were smaller in amplitude and longer in decay time in the Mecp2-null mice than in the wild type. Single-cell PCR analysis showed a decrease in the expression of α3-, α4-, α7-, and β3-subunits and an increase in the α5- and α6-subunits in the mutant mice. The α5-subunit was present in many of the LC neurons with slow-decay nAChR currents. The nicotinic modulation of spontaneous GABAA-ergic inhibitory postsynaptic currents in LC neurons was enhanced in Mecp2-null mice. In contrast, the nAChR manipulation of glutamatergic input to LC neurons was unaffected in both groups of mice. Our current-clamp studies showed that the modulation of LC neurons by ACh input was reduced moderately in Mecp2-null mice, despite the major decrease in nAChR currents, suggesting possible compensatory processes may take place, thus reducing the defects to a lesser extent in LC neurons. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24918098  Title: Repeated insulin-like growth factor 1 treatment in a patient with rett syndrome: a single case study.  Rett syndrome (RTT) is a devastating neurodevelopmental disorder that has no cure. Patients show regression of acquired skills, motor, and speech impairment, cardio-respiratory distress, microcephaly, and stereotyped hand movements. The majority of RTT patients display mutations in the gene that codes for the Methyl-CpG binding protein 2 (MeCP2), which is involved in the development of the central nervous system, especially synaptic and circuit maturation. Thus, agents that promote brain development and synaptic function are good candidates for ameliorating the symptoms of RTT. In particular, insulin-like growth factor 1 (IGF1) and its active peptide (1-3) IGF1 cross the Blood Brain Barrier, and therefore are ideal treatments for RTT Indeed, both (1-3) IGF1 and IGF1 treatment significantly ameliorates RTT symptoms in a mouse model of the disease In a previous study, we established that IGF1 is safe and well tolerated on Rett patients. In this open label clinical case study, we assess the safety and tolerability of IGF1 administration in two cycles of the treatment. Before and after each cycle, we monitored the clinical and blood parameters, autonomic function, and social and cognitive abilities, and we found that IGF1 was well tolerated each time and did not induce any side effect, nor it interfered with the other treatments that the patient was undergoing. We noticed a moderate improvement in the cognitive, social, and autonomic abilities of the patient after each cycle but the benefits were not retained between the two cycles, consistent with the pre-clinical observation that treatments for RTT should be administered through life. We find that repeated IGF1 treatment is safe and well tolerated in Rett patients but observed effects are not retained between cycles. These results have applications to other pathologies considering that IGF1 has been shown to be effective in other disorders of the autism spectrum. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24917201  Title: Improvement of the Rett syndrome phenotype in a MeCP2 mouse model upon treatment with levodopa and a dopa-decarboxylase inhibitor.  Rett Syndrome is a neurodevelopmental autism spectrum disorder caused by mutations in the gene coding for methyl CpG-binding protein (MeCP2). The disease is characterized by abnormal motor, respiratory, cognitive impairment, and autistic-like behaviors. No effective treatment of the disorder is available. Mecp2 knockout mice have a range of physiological and neurological abnormalities that resemble the human syndrome and can be used as a model to interrogate new therapies. Herein, we show that the combined administration of Levodopa and a Dopa-decarboxylase inhibitor in RTT mouse models is well tolerated, diminishes RTT-associated symptoms, and increases life span. The amelioration of RTT symptomatology is particularly significant in those features controlled by the dopaminergic pathway in the nigrostratium, such as mobility, tremor, and breathing. Most important, the improvement of the RTT phenotype upon use of the combined treatment is reflected at the cellular level by the development of neuronal dendritic growth. However, much work is required to extend the duration of the benefit of the described preclinical treatment. |
| MECP2, SATB2, TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24906019  Title: Transcriptional consequences of 16p11.2 deletion and duplication in mouse cortex and multiplex autism families.  Reciprocal copy-number variation (CNV) of a 593 kb region of 16p11.2 is a common genetic cause of autism spectrum disorder (ASD), yet it is not completely penetrant and can manifest in a wide array of phenotypes. To explore its molecular consequences, we performed RNA sequencing of cerebral cortex from mouse models with CNV of the syntenic 7qF3 region and lymphoblast lines from 34 members of 7 multiplex ASD-affected families harboring the 16p11.2 CNV. Expression of all genes in the CNV region correlated well with their DNA copy number, with no evidence of dosage compensation. We observed effects on gene expression outside the CNV region, including apparent positional effects in cis and in trans at genomic segments with evidence of physical interaction in Hi-C chromosome conformation data. One of the most significant positional effects was telomeric to the 16p11.2 CNV and includes the previously described "distal" 16p11.2 microdeletion. Overall, 16p11.2 CNV was associated with altered expression of genes and networks that converge on multiple hypotheses of ASD pathogenesis, including synaptic function (e.g., NRXN1, NRXN3), chromatin modification (e.g., CHD8, EHMT1, MECP2), transcriptional regulation (e.g., TCF4, SATB2), and intellectual disability (e.g., FMR1, CEP290). However, there were differences between tissues and species, with the strongest effects being consistently within the CNV region itself. Our analyses suggest that through a combination of indirect regulatory effects and direct effects on nuclear architecture, alteration of 16p11.2 genes disrupts expression networks that involve other genes and pathways known to contribute to ASD, suggesting an overlap in mechanisms of pathogenesis. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24777420  Title: Cellular origins of auditory event-related potential deficits in Rett syndrome.  Dysfunction in sensory information processing is a hallmark of many neurological disorders, including autism spectrum disorders, schizophrenia and Rett syndrome (RTT). Using mouse models of RTT, a monogenic disorder caused by mutations in MECP2, we found that the large-scale loss of MeCP2 from forebrain GABAergic interneurons led to deficits in auditory event-related potentials and seizure manifestation, whereas the restoration of MeCP2 in specific classes of interneurons ameliorated these deficits. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24776741  Title: De novo mutations in schizophrenia implicate chromatin remodeling and support a genetic overlap with autism and intellectual disability.  Schizophrenia is a serious psychiatric disorder with a broadly undiscovered genetic etiology. Recent studies of de novo mutations (DNMs) in schizophrenia and autism have reinforced the hypothesis that rare genetic variation contributes to risk. We carried out exome sequencing on 57 trios with sporadic or familial schizophrenia. In sporadic trios, we observed a ~3.5-fold increase in the proportion of nonsense DNMs (0.101 vs 0.031, empirical P=0.01, Benjamini-Hochberg-corrected P=0.044). These mutations were significantly more likely to occur in genes with highly ranked probabilities of haploinsufficiency (P=0.0029, corrected P=0.006). DNMs of potential functional consequence were also found to occur in genes predicted to be less tolerant to rare variation (P=2.01 × 10(-)(5), corrected P=2.1 × 10(-)(3)). Genes with DNMs overlapped with genes implicated in autism (for example, AUTS2, CHD8 and MECP2) and intellectual disability (for example, HUWE1 and TRAPPC9), supporting a shared genetic etiology between these disorders. Functionally CHD8, MECP2 and HUWE1 converge on epigenetic regulation of transcription suggesting that this may be an important risk mechanism. Our results were consistent in an analysis of additional exome-based sequencing studies of other neurodevelopmental disorders. These findings suggest that perturbations in genes, which function in the epigenetic regulation of brain development and cognition, could have a central role in the susceptibility to, pathogenesis and treatment of mental disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24723848  Title: Reduced synaptic activity in neuronal networks derived from embryonic stem cells of murine Rett syndrome model.  Neurodevelopmental diseases such as the Rett syndrome (RTT) have received renewed attention, since the mechanisms involved may underlie a broad range of neuropsychiatric disorders such as schizophrenia and autism. In vertebrates early stages in the functional development of neurons and neuronal networks are difficult to study. Embryonic stem cell-derived neurons provide an easily accessible tool to investigate neuronal differentiation and early network formation. We used in vitro cultures of neurons derived from murine embryonic stem cells missing the methyl-CpG-binding protein 2 (MECP2) gene (MeCP2-/y) and from wild type cells of the corresponding background. Cultures were assessed using whole-cell patch-clamp electrophysiology and immunofluorescence. We studied the functional maturation of developing neurons and the activity of the synaptic connections they formed. Neurons exhibited minor differences in the developmental patterns for their intrinsic parameters, such as resting membrane potential and excitability; with the MeCP2-/y cells showing a slightly accelerated development, with shorter action potential half-widths at early stages. There was no difference in the early phase of synapse development, but as the cultures matured, significant deficits became apparent, particularly for inhibitory synaptic activity. MeCP2-/y embryonic stem cell-derived neuronal cultures show clear developmental deficits that match phenotypes observed in slice preparations and thus provide a compelling tool to further investigate the mechanisms behind RTT pathophysiology. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24615633  Title: Rett syndrome and MeCP2.  Rett syndrome (RTT) is a severe and progressive neurological disorder, which mainly affects young females. Mutations of the methyl-CpG binding protein 2 (MECP2) gene are the most prevalent cause of classical RTT cases. MECP2 mutations or altered expression are also associated with a spectrum of neurodevelopmental disorders such as autism spectrum disorders with recent links to fetal alcohol spectrum disorders. Collectively, MeCP2 relation to these neurodevelopmental disorders highlights the importance of understanding the molecular mechanisms by which MeCP2 impacts brain development, mental conditions, and compromised brain function. Since MECP2 mutations were discovered to be the primary cause of RTT, a significant progress has been made in the MeCP2 research, with respect to the expression, function and regulation of MeCP2 in the brain and its contribution in RTT pathogenesis. To date, there have been intensive efforts in designing effective therapeutic strategies for RTT benefiting from mouse models and cells collected from RTT patients. Despite significant progress in MeCP2 research over the last few decades, there is still a knowledge gap between the in vitro and in vivo research findings and translating these findings into effective therapeutic interventions in human RTT patients. In this review, we will provide a synopsis of Rett syndrome as a severe neurological disorder and will discuss the role of MeCP2 in RTT pathophysiology. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24594659  Title: Brain region-specific expression of MeCP2 isoforms correlates with DNA methylation within Mecp2 regulatory elements.  MeCP2 is a critical epigenetic regulator in brain and its abnormal expression or compromised function leads to a spectrum of neurological disorders including Rett Syndrome and autism. Altered expression of the two MeCP2 isoforms, MeCP2E1 and MeCP2E2 has been implicated in neurological complications. However, expression, regulation and functions of the two isoforms are largely uncharacterized. Previously, we showed the role of MeCP2E1 in neuronal maturation and reported MeCP2E1 as the major protein isoform in the adult mouse brain, embryonic neurons and astrocytes. Recently, we showed that DNA methylation at the regulatory elements (REs) within the Mecp2 promoter and intron 1 impact the expression of Mecp2 isoforms in differentiating neural stem cells. This current study is aimed for a comparative analysis of temporal, regional and cell type-specific expression of MeCP2 isoforms in the developing and adult mouse brain. MeCP2E2 displayed a later expression onset than MeCP2E1 during mouse brain development. In the adult female and male brain hippocampus, both MeCP2 isoforms were detected in neurons, astrocytes and oligodendrocytes. Furthermore, MeCP2E1 expression was relatively uniform in different brain regions (olfactory bulb, striatum, cortex, hippocampus, thalamus, brainstem and cerebellum), whereas MeCP2E2 showed differential enrichment in these brain regions. Both MeCP2 isoforms showed relatively similar distribution in these brain regions, except for cerebellum. Lastly, a preferential correlation was observed between DNA methylation at specific CpG dinucleotides within the REs and Mecp2 isoform-specific expression in these brain regions. Taken together, we show that MeCP2 isoforms display differential expression patterns during brain development and in adult mouse brain regions. DNA methylation patterns at the Mecp2 REs may impact this differential expression of Mecp2/MeCP2 isoforms in brain regions. Our results significantly contribute towards characterizing the expression profiles of Mecp2/MeCP2 isoforms and thereby provide insights on the potential role of MeCP2 isoforms in the developing and adult brain. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24472844  Title: MeCP2 is required for activity-dependent refinement of olfactory circuits.  Methyl CpG binding protein 2 (MeCP2) is a structural chromosomal protein involved in the regulation of gene expression. Alterations in the levels of MeCP2 have been related to neurodevelopmental disorders. Studies in mouse models of MeCP2 deficiency have demonstrated that this protein is important for neuronal maturation, neurite complexity, synaptogenesis, and synaptic plasticity. However, the mechanisms by which MeCP2 dysfunction leads to neurodevelopmental defects, and the role of activity, remain unclear, as most studies examine the adult nervous system, which may obfuscate the primary consequences of MeCP2 mutation. We hypothesize that MeCP2 plays a role during the formation and activity-driven maturation of neural circuits at early postnatal stages. To test this hypothesis, we use the olfactory system as a neurodevelopmental model. This system undergoes postnatal neurogenesis; axons from olfactory neurons form highly stereotyped projections to higher-order neurons, facilitating the detection of possible defects in the establishment of connectivity. In vivo olfactory stimulation paradigms were used to produce physiological synaptic activity in gene-targeted mice in which specific olfactory circuits are visualized. Our results reveal defective postnatal refinement of olfactory circuits in Mecp2 knock out (KO) mice after sensory (odorant) stimulation. This failure in refinement was associated with deficits in the normal responses to odorants, including brain-derived neurotrophic factor (BDNF) production, as well as changes in adhesion molecules known to regulate axonal convergence. The defective refinement observed in Mecp2 KO mice was prevented by daily treatment with ampakine beginning after the first postnatal week. These observations indicate that increasing synaptic activity at early postnatal stage might circumvent the detrimental effect of MeCP2 deficiency on circuitry maturation. The present results provide in vivo evidence in real time for the role of MeCP2 in activity-dependent maturation of olfactory circuitry, with implications for understanding the mechanism of MeCP2 mutations in the development of neural connectivity. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24336718  Title: Dendritic arborization and spine dynamics are abnormal in the mouse model of MECP2 duplication syndrome.  MECP2 duplication syndrome is a childhood neurological disorder characterized by intellectual disability, autism, motor abnormalities, and epilepsy. The disorder is caused by duplications spanning the gene encoding methyl-CpG-binding protein-2 (MeCP2), a protein involved in the modulation of chromatin and gene expression. MeCP2 is thought to play a role in maintaining the structural integrity of neuronal circuits. Loss of MeCP2 function causes Rett syndrome and results in abnormal dendritic spine morphology and decreased pyramidal dendritic arbor complexity and spine density. The consequences of MeCP2 overexpression on dendritic pathophysiology remain unclear. We used in vivo two-photon microscopy to characterize layer 5 pyramidal neuron spine turnover and dendritic arborization as a function of age in transgenic mice expressing the human MECP2 gene at twice the normal levels of MeCP2 (Tg1; Collins et al., 2004). We found that spine density in terminal dendritic branches is initially higher in young Tg1 mice but falls below control levels after postnatal week 12, approximately correlating with the onset of behavioral symptoms. Spontaneous spine turnover rates remain high in older Tg1 animals compared with controls, reflecting the persistence of an immature state. Both spine gain and loss rates are higher, with a net bias in favor of spine elimination. Apical dendritic arbors in both simple- and complex-tufted layer 5 Tg1 pyramidal neurons have more branches of higher order, indicating that MeCP2 overexpression induces dendritic overgrowth. P70S6K was hyperphosphorylated in Tg1 somatosensory cortex, suggesting that elevated mTOR signaling may underlie the observed increase in spine turnover and dendritic growth. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24300809  Title: [Subchromosomal microdeletion identified by molecular karyotyping using DNA microarrays (array CGH) in Rett syndrome girls negative for MECP2 gene mutations].  Molecular karyotyping using DNA microarrays (array CGH) was applied for identification of subchromosomal microdeletions in a cohort of 12 girls with clinical features of RETT syndrome, but negative for MECP2 gene mutations. Recurrent microdeletions of MECP2 gene in chromosome X (locus Xq28) were identified in 5 girls of 12 studied. Probably RTT girls with subchromosomic microdeletions in Xq28 could represent a special subtype of the disease, which appears as clinically milder than the classic form of disease. In one case, an atypical form of RTT was associated with genomic abnormalities affecting CDKL5 gene and region critical for microdeletion Prader-Willi and Angelman syndromes (15q11.2). In addition, data are presented for the first time that genetic variation in regions 3p13, 3q27.1, and 1q21.1-1q21.2 could associate with RTT-like clinical manifestations. Without application of molecular karyotyping technology and bioinformatic method of assessing the pathogenic significance of genomic rearrangements these RTT-like girls negative for MECP2 gene mutations were considered as cases of idiopathic mental retardation associated with autism. It should be noted that absence of intragenic mutations in MECP2 gene is not sufficient criteria to reject the clinical diagnosis of RTT. To avoid errors in the genetic diagnosis of this genetically heterogeneous brain disease molecular cytogenetic studies using high resolution oligonucleotide array CGH (molecular karyotyping) are needed. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24238559  Title: Decitabine alters the expression of Mecp2 isoforms via dynamic DNA methylation at the Mecp2 regulatory elements in neural stem cells.  Aberrant MeCP2 expression in brain is associated with neurodevelopmental disorders including autism. In the brain of stressed mouse and autistic human patients, reduced MeCP2 expression is correlated with Mecp2/MECP2 promoter hypermethylation. Altered expression of MeCP2 isoforms (MeCP2E1 and MeCP2E2) is associated with neurological disorders, highlighting the importance of proper regulation of both isoforms. While known regulatory elements (REs) within the MECP2/Mecp2 promoter and intron 1 are involved in MECP2/Mecp2 regulation, Mecp2 isoform-specific regulatory mechanisms are unknown. We hypothesized that DNA methylation at these REs may impact the expression of Mecp2 isoforms. We used a previously characterized in vitro differentiating neural stem cell (NSC) system to investigate the interplay between Mecp2 isoform-specific expression and DNA methylation at the Mecp2 REs. We studied altered expression of Mecp2 isoforms, affected by global DNA demethylation and remethylation, induced by exposure and withdrawal of decitabine (5-Aza-2'-deoxycytidine). Further, we performed correlation analysis between DNA methylation at the Mecp2 REs and the expression of Mecp2 isoforms after decitabine exposure and withdrawal. At different stages of NSC differentiation, Mecp2 isoforms showed reciprocal expression patterns associated with minor, but significant changes in DNA methylation at the Mecp2 REs. Decitabine treatment induced Mecp2e1/MeCP2E1 (but not Mecp2e2) expression at day (D) 2, associated with DNA demethylation at the Mecp2 REs. In contrast, decitabine withdrawal downregulated both Mecp2 isoforms to different extents at D8, without affecting DNA methylation at the Mecp2 REs. NSC cell fate commitment was minimally affected by decitabine under tested conditions. Expression of both isoforms negatively correlated with methylation at specific regions of the Mecp2 promoter, both at D2 and D8. The correlation between intron 1 methylation and Mecp2e1 (but not Mecp2e2) varied depending on the stage of NSC differentiation (D2: negative; D8: positive). Our results show the correlation between the expression of Mecp2 isoforms and DNA methylation in differentiating NSC, providing insights on the potential role of DNA methylation at the Mecp2 REs in Mecp2 isoform-specific expression. The ability of decitabine to induce Mecp2e1/MeCP2E1, but not Mecp2e2 suggests differential sensitivity of Mecp2 isoforms to decitabine and is important for future drug therapies for autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24238429  Title: Transcriptomic analysis of genetically defined autism candidate genes reveals common mechanisms of action.  Austism spectrum disorder (ASD) is a heterogeneous behavioral disorder or condition characterized by severe impairment of social engagement and the presence of repetitive activities. The molecular etiology of ASD is still largely unknown despite a strong genetic component. Part of the difficulty in turning genetics into disease mechanisms and potentially new therapeutics is the sheer number and diversity of the genes that have been associated with ASD and ASD symptoms. The goal of this work is to use shRNA-generated models of genetic defects proposed as causative for ASD to identify the common pathways that might explain how they produce a core clinical disability. Transcript levels of Mecp2, Mef2a, Mef2d, Fmr1, Nlgn1, Nlgn3, Pten, and Shank3 were knocked-down in mouse primary neuron cultures using shRNA constructs. Whole genome expression analysis was conducted for each of the knockdown cultures as well as a mock-transduced culture and a culture exposed to a lentivirus expressing an anti-luciferase shRNA. Gene set enrichment and a causal reasoning engine was employed to identify pathway level perturbations generated by the transcript knockdown. Quantification of the shRNA targets confirmed the successful knockdown at the transcript and protein levels of at least 75% for each of the genes. After subtracting out potential artifacts caused by viral infection, gene set enrichment and causal reasoning engine analysis showed that a significant number of gene expression changes mapped to pathways associated with neurogenesis, long-term potentiation, and synaptic activity. This work demonstrates that despite the complex genetic nature of ASD, there are common molecular mechanisms that connect many of the best established autism candidate genes. By identifying the key regulatory checkpoints in the interlinking transcriptional networks underlying autism, we are better able to discover the ideal points of intervention that provide the broadest efficacy across the diverse population of autism patients. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24164323  Title: Identification of two novel Shank3 transcripts in the developing mouse neocortex.  SHANK3 is a synaptic scaffolding protein enriched in the post-synaptic density of excitatory synapses. Since several SHANK3 mutations have been identified in a particular phenotypic group of patients with autism spectrum disorder (ASD), SHANK3 is strongly suspected of being involved in the pathogenesis and neuropathology of ASD. Several SHANK3 isoforms are known to be produced in the developing brain, but they have not been fully investigated. Here, we identified two different amino-terminus truncated Shank3 transcripts. One transcript, designated as Shank3c-3, produces an isoform that contains the entire carboxyl-terminus, but the other transcript, designated as Shank3c-4, produces a carboxyl-terminus truncated isoform. During development, expression of the novel Shank3 transcripts increased after birth, transiently decreased at P14 and then gradually increased again thereafter. We also determined that methyl CpG-binding protein 2 (MeCP2) is involved in regulating expression of the novel Shank3 transcripts. MeCP2 is a transcriptional regulator that has been identified as the causative molecule of Rett syndrome, a neurodevelopmental disorder that includes autistic behavior. We demonstrated a difference between the expression of the novel Shank3 transcripts in wild-type mice and Mecp2-deficient mice. These findings suggest that the SHANK3 isoforms may be implicated in the synaptic abnormality in Rett syndrome. SHANK3 is a synaptic scaffolding protein and is suspected of being implicated in the pathogenesis and neuropathology of ASD. We here identified two different amino-terminus truncated Shank3 transcripts, Shank3c-3 and Shank3c-4, expressed from the intron 10 of the Shank3 gene, and also suggested the epigenetic regulation of their expression via methyl CpG-binding protein 2 (MeCP2) that has been identified as the causative molecule of Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24151012  Title: Genetic effects on cerebellar structure across mouse models of autism using a magnetic resonance imaging atlas.  Magnetic resonance imaging (MRI) of autism populations is confounded by the inherent heterogeneity in the individuals' genetics and environment, two factors difficult to control for. Imaging genetic animal models that recapitulate a mutation associated with autism quantify the impact of genetics on brain morphology and mitigate the confounding factors in human studies. Here, we used MRI to image three genetic mouse models with single mutations implicated in autism: Neuroligin-3 R451C knock-in, Methyl-CpG binding protein-2 (MECP2) 308-truncation and integrin β3 homozygous knockout. This study identified the morphological differences specific to the cerebellum, a structure repeatedly linked to autism in human neuroimaging and postmortem studies. To accomplish a comparative analysis, a segmented cerebellum template was created and used to segment each study image. This template delineated 39 different cerebellar structures. For Neuroligin-3 R451C male mutants, the gray (effect size (ES) = 1.94, FDR q = 0.03) and white (ES = 1.84, q = 0.037) matter of crus II lobule and the gray matter of the paraflocculus (ES = 1.45, q = 0.045) were larger in volume. The MECP2 mutant mice had cerebellar volume changes that increased in scope depending on the genotype: hemizygous males to homozygous females. The integrin β3 mutant mouse had a drastically smaller cerebellum than controls with 28 out of 39 cerebellar structures smaller. These imaging results are discussed in relation to repetitive behaviors, sociability, and learning in the context of autism. This work further illuminates the cerebellum's role in autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24040966  Title: Role of conserved cis-regulatory elements in the post-transcriptional regulation of the human MECP2 gene involved in autism.  The MECP2 gene codes for methyl CpG binding protein 2 which regulates activities of other genes in the early development of the brain. Mutations in this gene have been associated with Rett syndrome, a form of autism. The purpose of this study was to investigate the role of evolutionarily conserved cis-elements in regulating the post-transcriptional expression of the MECP2 gene and to explore their possible correlations with a mutation that is known to cause mental retardation. A bioinformatics approach was used to map evolutionarily conserved cis-regulatory elements in the transcribed regions of the human MECP2 gene and its mammalian orthologs. Cis-regulatory motifs including G-quadruplexes, microRNA target sites, and AU-rich elements have gained significant importance because of their role in key biological processes and as therapeutic targets. We discovered in the 5'-UTR (untranslated region) of MECP2 mRNA a highly conserved G-quadruplex which overlapped a known deletion in Rett syndrome patients with decreased levels of MeCP2 protein. We believe that this 5'-UTR G-quadruplex could be involved in regulating MECP2 translation. We mapped additional evolutionarily conserved G-quadruplexes, microRNA target sites, and AU-rich elements in the key sections of both untranslated regions. Our studies suggest the regulation of translation, mRNA turnover, and development-related alternative MECP2 polyadenylation, putatively involving interactions of conserved cis-regulatory elements with their respective trans factors and complex interactions among the trans factors themselves. We discovered highly conserved G-quadruplex motifs that were more prevalent near alternative splice sites as compared to the constitutive sites of the MECP2 gene. We also identified a pair of overlapping G-quadruplexes at an alternative 5' splice site that could potentially regulate alternative splicing in a negative as well as a positive way in the MECP2 pre-mRNAs. A Rett syndrome mutation with decreased protein expression was found to be associated with a conserved G-quadruplex. Our studies suggest that MECP2 post-transcriptional gene expression could be regulated by several evolutionarily conserved cis-elements like G-quadruplex motifs, microRNA target sites, and AU-rich elements. This phylogenetic analysis has provided some interesting and valuable insights into the regulation of the MECP2 gene involved in autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23770587  Title: Activity-dependent phosphorylation of MeCP2 threonine 308 regulates interaction with NCoR.  Rett syndrome (RTT) is an X-linked human neurodevelopmental disorder with features of autism and severe neurological dysfunction in females. RTT is caused by mutations in methyl-CpG-binding protein 2 (MeCP2), a nuclear protein that, in neurons, regulates transcription, is expressed at high levels similar to that of histones, and binds to methylated cytosines broadly across the genome. By phosphotryptic mapping, we identify three sites (S86, S274 and T308) of activity-dependent MeCP2 phosphorylation. Phosphorylation of these sites is differentially induced by neuronal activity, brain-derived neurotrophic factor, or agents that elevate the intracellular level of 3',5'-cyclic AMP (cAMP), indicating that MeCP2 may function as an epigenetic regulator of gene expression that integrates diverse signals from the environment. Here we show that the phosphorylation of T308 blocks the interaction of the repressor domain of MeCP2 with the nuclear receptor co-repressor (NCoR) complex and suppresses the ability of MeCP2 to repress transcription. In knock-in mice bearing the common human RTT missense mutation R306C, neuronal activity fails to induce MeCP2 T308 phosphorylation, suggesting that the loss of T308 phosphorylation might contribute to RTT. Consistent with this possibility, the mutation of MeCP2 T308A in mice leads to a decrease in the induction of a subset of activity-regulated genes and to RTT-like symptoms. These findings indicate that the activity-dependent phosphorylation of MeCP2 at T308 regulates the interaction of MeCP2 with the NCoR complex, and that RTT in humans may be due, in part, to the loss of activity-dependent MeCP2 T308 phosphorylation and a disruption of the phosphorylation-regulated interaction of MeCP2 with the NCoR complex. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23759142  Title: Comparative DNA methylation among females with neurodevelopmental disorders and seizures identifies TAC1 as a MeCP2 target gene.  Several proteins involved in epigenetic regulation cause syndromic neurodevelopmental disorders when human genes are mutated. More general involvement of epigenetic mechanisms in neurodevelopmental phenotypes is unclear. In an attempt to determine whether DNA methylation differentiates clinical subgroups, profiling was performed on bisulfite converted DNA from lymphoblastoid cell lines (LCLs) in discovery (n = 20) and replication (n = 40) cohorts of females with Rett syndrome (RTT; n = 18), autism (AUT; n = 17), seizure disorder (SEZ; n = 6), and controls (CTL; n = 19) using Illumina HumanMethylation27 arrays. TAC1 CpGs were validated using a Sequenom EpiTYPER assay and expression was measured in LCLs and postmortem brain. Chromatin immunoprecipitation was performed in HEK cells. Cells were treated with valproic acid and MeCP2 binding was assessed. Two female-only cohorts were analyzed. DNA methylation profiling in a discovery cohort identified 40 CpGs that exhibited statistically significant differential methylation (≥15%) between clinical groups (P <0.01). Hierarchical clustering and principal components analysis suggested neurodevelopmental groups were distinct from CTL, but not from each other. In a larger and more heterogeneous replication cohort, these 40 CpG sites suggested no clear difference between clinical groups. Pooled analysis of DNA methylation across all 60 samples suggested only four differentially methylated CpG sites (P <0.0005), including TAC1. TAC1 promoter CpG hypermethylation was validated in AUT and SEZ (P <0.005). Analyzed for the first time in postmortem brain, TAC1 expression was reduced in cingulate cortex in RTT and AUT+SEZ (P = 0.003). However, no significant difference in TAC1 promoter CpG methylation was detected in RTT and AUT+SEZ brains. Additional molecular analyses revealed that MeCP2 binds directly to the TAC1 promoter and is sensitive to antiepileptic drug treatment. These data suggest that DNA methylation is not widely altered in RTT, consistent with subtle changes in gene expression previously observed. However, TAC1 may be an important target for further functional analyses in RTT. Studies of larger sample cohorts using primary cells that also consider shared clinical features and drug treatments may be required to address apparent subtle disruptions of DNA methylation in neurodevelopmental disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23750231  Title: MeCP2 regulates the synaptic expression of a Dysbindin-BLOC-1 network component in mouse brain and human induced pluripotent stem cell-derived neurons.  Clinical, epidemiological, and genetic evidence suggest overlapping pathogenic mechanisms between autism spectrum disorder (ASD) and schizophrenia. We tested this hypothesis by asking if mutations in the ASD gene MECP2 which cause Rett syndrome affect the expression of genes encoding the schizophrenia risk factor dysbindin, a subunit of the biogenesis of lysosome-related organelles complex-1 (BLOC-1), and associated interacting proteins. We measured mRNA and protein levels of key components of a dysbindin interaction network by, quantitative real time PCR and quantitative immunohistochemistry in hippocampal samples of wild-type and Mecp2 mutant mice. In addition, we confirmed results by performing immunohistochemistry of normal human hippocampus and quantitative qRT-PCR of human inducible pluripotent stem cells (iPSCs)-derived human neurons from Rett syndrome patients. We defined the distribution of the BLOC-1 subunit pallidin in human and mouse hippocampus and contrasted this distribution with that of symptomatic Mecp2 mutant mice. Neurons from mutant mice and Rett syndrome patients displayed selectively reduced levels of pallidin transcript. Pallidin immunoreactivity decreased in the hippocampus of symptomatic Mecp2 mutant mice, a feature most prominent at asymmetric synapses as determined by immunoelectron microcopy. Pallidin immunoreactivity decreased concomitantly with reduced BDNF content in the hippocampus of Mecp2 mice. Similarly, BDNF content was reduced in the hippocampus of BLOC-1 deficient mice suggesting that genetic defects in BLOC-1 are upstream of the BDNF phenotype in Mecp2 deficient mice. Our results demonstrate that the ASD-related gene Mecp2 regulates the expression of components belonging to the dysbindin interactome and these molecular differences may contribute to synaptic phenotypes that characterize Mecp2 deficiencies and ASD. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23449173  Title: Mechanisms and therapeutic challenges in autism spectrum disorders: insights from Rett syndrome.  A major challenge for understanding neurodevelopmental disorders, including autism spectrum disorders (ASDs), is to advance the findings from gene discovery to an exposition of neurobiological mechanisms that underlie these disorders and subsequently translate this knowledge into mechanism-based therapeutics. A promising way to proceed is revealed by the recent studies of rare subsets of ASDs. In this review, we summarize the latest advances in the mechanisms and emerging therapeutics for a rare single-gene ASD, Rett syndrome. Rett syndrome is caused by mutations in the gene coding for methyl CpG-binding protein 2 (MeCP2). Although MeCP2 has diverse functions, examination of MeCP2 mutant mice suggests the hypothesis that MeCP2 deficiency leads to aberrant maturation and maintenance of synapses and circuits in multiple brain systems. Some of the deficits arise from alterations in specific intracellular pathways such as the PI3K/Akt signaling pathway. These abnormalities can be at least partially rescued in MeCP2 mutant mice by treatment with therapeutic agents. Mechanism-based therapeutics are emerging for single-gene neurodevelopmental disorders such as Rett syndrome. Given the complexity of MeCP2 function, future directions include combination therapeutics that target multiple molecules and pathways. Such approaches will likely be applicable to other ASDs as well. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23392116  Title: GABAergic synaptic inputs of locus coeruleus neurons in wild-type and Mecp2-null mice.  Rett syndrome is an autism spectrum disorder resulting from defects in the gene encoding the methyl-CpG-binding protein 2 (MeCP2). Deficiency of the Mecp2 gene causes abnormalities in several systems in the brain, especially the norepinephrinergic and GABAergic systems. The norepinephrinergic neurons in the locus coeruleus (LC) modulate a variety of neurons and play an important role in multiple functions in the central nervous system. In Mecp2(-/Y) mice, defects in the intrinsic membrane properties of LC neurons have been identified, while how their synaptic inputs are affected remains unclear. Therefore, we performed these brain slice studies to demonstrate how LC neurons are regulated by GABAergic inputs and how such synaptic inputs are affected by Mecp2 knockout. In whole cell current clamp, the firing activity of LC neurons was strongly inhibited by the GABAA receptor agonist muscimol, accompanied by hyperpolarization and a decrease in input resistance. Such a postsynaptic inhibition was significantly reduced (by ~30%) in Mecp2(-/Y) mice. Post- and presynaptic GABABergic inputs were found in LC neurons, which were likely mediated by the G protein-coupled, Ba(2+)-sensitive K(+) channels. The postsynaptic GABABergic inhibition was deficient by ~50% in Mecp2 knockout mice. Although the presynaptic GABABergic modulation appeared normal, both frequency and amplitude of the GABAAergic mIPSCs were drastically decreased (by 30-40%) in Mecp2-null mice. These results suggest that the Mecp2 disruption causes defects in both post- and presynaptic GABAergic systems in LC neurons, impairing GABAAergic and GABABergic postsynaptic inhibition and decreasing the GABA release from presynaptic terminals. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25003017  Title: A role for metabolism in Rett syndrome pathogenesis: New clinical findings and potential treatment targets.  Rett syndrome (RTT), an X-linked neurological disorder caused by mutations in MECP2, may have a metabolic component. We reported a genetic suppressor screen in a Mecp2-null mouse model to identify pathways for therapeutic improvement of RTT symptoms. Of note, one suppressor mutation implied that cholesterol homeostasis was perturbed in Mecp2 null mice; indeed, cholesterol synthesis was elevated in the brain and body system. Remarkably, the genetic effect of downregulating the cholesterol pathway could be mimicked chemically by statin drugs, improving motor symptoms, and increasing longevity in the mouse. Our work linked cholesterol metabolism to RTT pathology for the first time. Both neurological and systemic effects of perturbed cholesterol homeostasis overlap with many RTT symptoms. Here we show in patients that peripheral cholesterol, triglycerides, and/or LDLs may be elevated early in RTT disease onset, providing a biomarker for patients that could be aided by therapeutic interventions that modulate lipid metabolism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23226951  Title: The relationship of Rett syndrome and MECP2 disorders to autism.  Rett syndrome (RTT, MIM#312750) is a neurodevelopmental disorder that is classified as an autism spectrum disorder. Clinically, RTT is characterized by psychomotor regression with loss of volitional hand use and spoken language, the development of repetitive hand stereotypies, and gait impairment. The majority of people with RTT have mutations in Methyl-CpG-binding Protein 2 (MECP2), a transcriptional regulator. Interestingly, alterations in the function of the protein product produced by MECP2, MeCP2, have been identified in a number of other clinical conditions. The many clinical features found in RTT and the various clinical problems that result from alteration in MeCP2 function have led to the belief that understanding RTT will provide insight into a number of other neurological disorders. Excitingly, RTT is reversible in a mouse model, providing inspiration and hope that such a goal may be achieved for RTT and potentially for many neurodevelopmental disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23035095  Title: Brain activity mapping in Mecp2 mutant mice reveals functional deficits in forebrain circuits, including key nodes in the default mode network, that are reversed with ketamine treatment.  Excitatory-inhibitory imbalance has been identified within specific brain microcircuits in models of Rett syndrome (RTT) and other autism spectrum disorders (ASDs). However, macrocircuit dysfunction across the RTT brain as a whole has not been defined. To approach this issue, we mapped expression of the activity-dependent, immediate-early gene product Fos in the brains of wild-type (Wt) and methyl-CpG-binding protein 2 (Mecp2)-null (Null) mice, a model of RTT, before and after the appearance of overt symptoms (3 and 6 weeks of age, respectively). At 6 weeks, Null mice exhibit significantly less Fos labeling than Wt in limbic cortices and subcortical structures, including key nodes in the default mode network. In contrast, Null mice exhibit significantly more Fos labeling than Wt in the hindbrain, most notably in cardiorespiratory regions of the nucleus tractus solitarius (nTS). Using nTS as a model, whole-cell recordings demonstrated that increased Fos expression in Nulls at 6 weeks of age is associated with synaptic hyperexcitability, including increased frequency of spontaneous and miniature EPSCs and increased amplitude of evoked EPSCs in Nulls. No such effect of genotype on Fos or synaptic function was seen at 3 weeks. In the mutant forebrain, reduced Fos expression, as well as abnormal sensorimotor function, were reversed by the NMDA receptor antagonist ketamine. In light of recent findings that the default mode network is hypoactive in autism, our data raise the possibility that hypofunction within this meta-circuit is a shared feature of RTT and other ASDs and is reversible. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24999353  Title: The neural circuit basis of Rett syndrome.  Rett syndrome is an Autism Spectrum Disorder caused by mutations in the gene encoding methyl-CpG binding protein (MeCP2). Following a period of normal development, patients lose learned communication and motor skills, and develop a number of symptoms including motor disturbances, cognitive impairments and often seizures. In this review, we discuss the role of MeCP2 in regulating synaptic function and how synaptic dysfunctions lead to neuronal network impairments and alterations in sensory information processing. We propose that Rett syndrome is a disorder of neural circuits as a result of non-linear accumulated dysfunction of synapses at the level of individual cell populations across multiple neurotransmitter systems and brain regions. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22973007  Title: MeCP2 phosphorylation is required for modulating synaptic scaling through mGluR5.  MeCP2 (methyl CpG binding protein 2) is a key player in recognizing methylated DNA and interpreting the epigenetic information encoded in different DNA methylation patterns. The functional significance of MeCP2 to the mammalian nervous system is highlighted by the discovery that mutations in the MECP2 gene cause Rett syndrome (RTT), a devastating neurological disease that shares many features with autism. Synaptic scaling is a form of non-Hebbian homeostatic plasticity that allows neurons to regulate overall excitability in response to changes in network neuronal activity levels. While it is known that neuronal activity can induce phosphorylation of MeCP2 and that MeCP2 can regulate synaptic scaling, the molecular link between MeCP2 phosphorylation and synaptic scaling remains undefined. We show here that MeCP2 phosphorylation is specifically required for bicuculline-induced synaptic scaling down in mouse hippocampal neurons and this phenotype is mediated by mGluR5 (metabotropic glutamate receptor 5). Our results reveal an important function of MeCP2 in regulating neuronal homeostasis and may eventually help us understand how MECP2 mutations cause RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22865604  Title: Disease modeling using embryonic stem cells: MeCP2 regulates nuclear size and RNA synthesis in neurons.  Mutations in the gene encoding the methyl-CpG-binding protein MECP2 are the major cause of Rett syndrome, an autism spectrum disorder mainly affecting young females. MeCP2 is an abundant chromatin-associated protein, but how and when its absence begins to alter brain function is still far from clear. Using a stem cell-based system allowing the synchronous differentiation of neuronal progenitors, we found that in the absence of MeCP2, the size of neuronal nuclei fails to increase at normal rates during differentiation. This is accompanied by a marked decrease in the rate of ribonucleotide incorporation, indicating an early role of MeCP2 in regulating total gene transcription, not restricted to selected mRNAs. We also found that the levels of brain-derived neurotrophic factor (BDNF) were decreased in mutant neurons, while those of the presynaptic protein synaptophysin increased at similar rates in wild-type and mutant neurons. By contrast, nuclear size, transcription rates, and BDNF levels remained unchanged in astrocytes lacking MeCP2. Re-expressing MeCP2 in mutant neurons rescued the nuclear size phenotype as well as BDNF levels. These results reveal a new role of MeCP2 in regulating overall RNA synthesis in neurons during the course of their maturation, in line with recent findings indicating a reduced nucleolar size in neurons of the developing brain of mice lacking Mecp2. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22815516  Title: MeCP2 is critical for maintaining mature neuronal networks and global brain anatomy during late stages of postnatal brain development and in the mature adult brain.  Mutations in the X-linked gene, methyl-CpG binding protein 2 (Mecp2), underlie a wide range of neuropsychiatric disorders, most commonly, Rett Syndrome (RTT), a severe autism spectrum disorder that affects approximately one in 10,000 female live births. Because mutations in the Mecp2 gene occur in the germ cells with onset of neurological symptoms occurring in early childhood, the role of MeCP2 has been ascribed to brain maturation at a specific developmental window. Here, we show similar kinetics of onset and progression of RTT-like symptoms in mice, including lethality, if MeCP2 is removed postnatally during the developmental stage that coincides with RTT onset, or adult stage. For the first time, we show that brains that lose MeCP2 at these two different stages are actively shrinking, resulting in higher than normal neuronal cell density. Furthermore, we show that mature dendritic arbors of pyramidal neurons are severely retracted and dendritic spine density is dramatically reduced. In addition, hippocampal astrocytes have significantly less complex ramified processes. These changes accompany a striking reduction in the levels of several synaptic proteins, including CaMKII α/β, AMPA, and NMDA receptors, and the synaptic vesicle proteins Vglut and Synapsin, which represent critical modifiers of synaptic function and dendritic arbor structure. Importantly, the mRNA levels of these synaptic proteins remains unchanged, suggesting that MeCP2 likely regulates these synaptic proteins post-transcriptionally, directly or indirectly. Our data suggest a crucial role for MeCP2 in post-transcriptional regulation of critical synaptic proteins involved in maintaining mature neuronal networks during late stages of postnatal brain development. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22586411  Title: Rett syndrome: genes, synapses, circuits, and therapeutics.  Development of the nervous system proceeds through a set of complex checkpoints which arise from a combination of sequential gene expression and early neural activity sculpted by the environment. Genetic and environmental insults lead to neurodevelopmental disorders which encompass a large group of diseases that result from anatomical and physiological abnormalities during maturation and development of brain circuits. Rett syndrome (RTT) is a neurological disorder of genetic origin, caused by mutations in the X-linked gene methyl-CpG binding protein 2 (MeCP2). It features a range of neuropsychiatric abnormalities including motor dysfunctions and mild to severe cognitive impairment. Here, we discuss key questions and recent studies describing animal models, cell-type specific functions of methyl-CpG binding protein 2 (MeCP2), defects in neural circuit plasticity, and attempts to evaluate possible therapeutic strategies for RTT. We also discuss how genes, proteins, and overlapping signaling pathways affect the molecular etiology of apparently unrelated neuropsychiatric disorders, an understanding of which can offer novel therapeutic strategies for a range of autism spectrum disorders (ASDs). |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22522469  Title: Dendritic spine pathology in epilepsy: cause or consequence?  Abnormalities in dendritic spines have commonly been observed in brain specimens from epilepsy patients and animal models of epilepsy. However, the functional implications and clinical consequences of this dendritic pathology for epilepsy are uncertain. Dendritic spine abnormalities may promote hyperexcitable circuits and seizures in some types of epilepsy, especially in specific genetic syndromes with documented dendritic pathology, but in these cases it is difficult to differentiate their effects on seizures versus other comorbidities, such as cognitive deficits and autism. In other situations, seizures themselves may cause damage to dendrites and dendritic spines and this seizure-induced brain injury may then contribute to progressive epileptogenesis, memory problems and other neurological deficits in epilepsy patients. The mechanistic basis of dendritic spine abnormalities in epilepsy has begun to be elucidated and suggests novel therapeutic strategies for treating epilepsy and its complications. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22425995  Title: Wild-type microglia arrest pathology in a mouse model of Rett syndrome.  Rett syndrome is an X-linked autism spectrum disorder. The disease is characterized in most cases by mutation of the MECP2 gene, which encodes a methyl-CpG-binding protein. Although MECP2 is expressed in many tissues, the disease is generally attributed to a primary neuronal dysfunction. However, as shown recently, glia, specifically astrocytes, also contribute to Rett pathophysiology. Here we examine the role of another form of glia, microglia, in a murine model of Rett syndrome. Transplantation of wild-type bone marrow into irradiation-conditioned Mecp2-null hosts resulted in engraftment of brain parenchyma by bone-marrow-derived myeloid cells of microglial phenotype, and arrest of disease development. However, when cranial irradiation was blocked by lead shield, and microglial engraftment was prevented, disease was not arrested. Similarly, targeted expression of MECP2 in myeloid cells, driven by Lysm(cre) on an Mecp2-null background, markedly attenuated disease symptoms. Thus, through multiple approaches, wild-type Mecp2-expressing microglia within the context of an Mecp2-null male mouse arrested numerous facets of disease pathology: lifespan was increased, breathing patterns were normalized, apnoeas were reduced, body weight was increased to near that of wild type, and locomotor activity was improved. Mecp2(+/-) females also showed significant improvements as a result of wild-type microglial engraftment. These benefits mediated by wild-type microglia, however, were diminished when phagocytic activity was inhibited pharmacologically by using annexin V to block phosphatydilserine residues on apoptotic targets, thus preventing recognition and engulfment by tissue-resident phagocytes. These results suggest the importance of microglial phagocytic activity in Rett syndrome. Our data implicate microglia as major players in the pathophysiology of this devastating disorder, and suggest that bone marrow transplantation might offer a feasible therapeutic approach for it. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22406388  Title: Oxytocin receptor and Mecp2 308/Y knockout mice exhibit altered expression of autism-related social behaviors.  The development of tasks measuring behaviors specific to the three major symptom categories for autism makes it possible to differentiate mouse models of autism spectrum disorders (ASD) in terms of changes in these specific categories. Prior studies indicate that BTBR T+tf/J mice, the strain that has been evaluated most extensively, show autism-relevant changes in all three symptom categories; reciprocal social interactions; communication; and repetitive, ritualized behaviors. This report reviews the behaviors of oxytocin receptor (Oxtr) and Mecp2(308/Y) wild-type (WT) and knockout (KO) mice, in a number of tests specifically designed to provide information on behaviors that may show functional parallels to the core symptoms of ASD. Oxtr KO mice show robust decreases in reciprocal social interactions, and reduced levels of communication, but no changes in repetitive, ritualized behaviors; whereas Mecp2(308/Y) KO mice show a slight but consistent enhancement of social behavior and communication, and no changes in repetitive, ritualized behaviors. This data base, although small, strongly indicates that mouse models can sort the diagnostic symptoms of autism, and suggests that biological and physiological analyses of these strains may be capable of providing differential information on the brain systems involved in particular symptoms of this disorder. Profiles of behavioral changes in other mouse models of ASD should provide additional specificity in the search for biomarkers associated with particular ASD symptoms and symptom clusters. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22249109  Title: MeCP2+/- mouse model of RTT reproduces auditory phenotypes associated with Rett syndrome and replicate select EEG endophenotypes of autism spectrum disorder.  Impairments in cortical sensory processing have been demonstrated in Rett syndrome (RTT) and Autism Spectrum Disorders (ASD) and are thought to contribute to high-order phenotypic deficits. However, underlying pathophysiological mechanisms for these abnormalities are unknown. This study investigated auditory sensory processing in a mouse model of RTT with a heterozygous loss of MeCP2 function. Cortical abnormalities in a number of neuropsychiatric disorders, including ASD are reflected in auditory evoked potentials and fields measured by EEG and MEG. One of these abnormalities, increased latency of cortically sourced components, is associated with language and developmental delay in autism. Additionally, gamma-band abnormalities have recently been identified as an endophenotype of idiopathic autism. Both of these cortical abnormalities are potential clinical endpoints for assessing treatment. While ascribing similar mechanisms of idiopathic ASD to Rett syndrome (RTT) has been controversial, we sought to determine if mouse models of RTT replicate these intermediate phenotypes. Mice heterozygous for the null mutations of the gene MeCP2, were implanted for EEG. In response to auditory stimulation, these mice recapitulated specific latency differences as well as select gamma and beta band abnormalities associated with ASD. MeCP2 disruption is the predominant cause of RTT, and reductions in MeCP2 expression predominate in ASD. This work further suggests a common cortical pathophysiology for RTT and ASD, and indicates that the MeCP2+/- model may be useful for preclinical development targeting specific cortical processing abnormalities in RTT with potential relevance to ASD. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22231481  Title: Crh and Oprm1 mediate anxiety-related behavior and social approach in a mouse model of MECP2 duplication syndrome.  Genomic duplications spanning Xq28 are associated with a spectrum of phenotypes, including anxiety and autism. The minimal region shared among affected individuals includes MECP2 and IRAK1, although it is unclear which gene when overexpressed causes anxiety and social behavior deficits. We report that doubling MECP2 levels causes heightened anxiety and autism-like features in mice and alters the expression of genes that influence anxiety and social behavior, such as Crh and Oprm1. To test the hypothesis that alterations in these two genes contribute to heightened anxiety and social behavior deficits, we analyzed MECP2 duplication mice (MECP2-TG1) that have reduced Crh and Oprm1 expression. In MECP2-TG1 animals, reducing the levels of Crh or its receptor, Crhr1, suppressed anxiety-like behavior; in contrast, reducing Oprm1 expression improved abnormal social behavior. These data indicate that increased MeCP2 levels affect molecular pathways underlying anxiety and social behavior and provide new insight into potential therapies for MECP2-related disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22230884  Title: Rett syndrome induced pluripotent stem cell-derived neurons reveal novel neurophysiological alterations.  Rett syndrome (RTT) is a neurodevelopmental autism spectrum disorder caused by mutations in the methyl-CpG-binding protein 2 (MECP2) gene. Here, we describe the first characterization and neuronal differentiation of induced pluripotent stem (iPS) cells derived from Mecp2-deficient mice. Fully reprogrammed wild-type (WT) and heterozygous female iPS cells express endogenous pluripotency markers, reactivate the X-chromosome and differentiate into the three germ layers. We directed iPS cells to produce glutamatergic neurons, which generated action potentials and formed functional excitatory synapses. iPS cell-derived neurons from heterozygous Mecp2(308) mice showed defects in the generation of evoked action potentials and glutamatergic synaptic transmission, as previously reported in brain slices. Further, we examined electrophysiology features not yet studied with the RTT iPS cell system and discovered that MeCP2-deficient neurons fired fewer action potentials, and displayed decreased action potential amplitude, diminished peak inward currents and higher input resistance relative to WT iPS-derived neurons. Deficiencies in action potential firing and inward currents suggest that disturbed Na(+) channel function may contribute to the dysfunctional RTT neuronal network. These phenotypes were additionally confirmed in neurons derived from independent WT and hemizygous mutant iPS cell lines, indicating that these reproducible deficits are attributable to MeCP2 deficiency. Taken together, these results demonstrate that neuronally differentiated MeCP2-deficient iPS cells recapitulate deficits observed previously in primary neurons, and these identified phenotypes further illustrate the requirement of MeCP2 in neuronal development and/or in the maintenance of normal function. By validating the use of iPS cells to delineate mechanisms underlying RTT pathogenesis, we identify deficiencies that can be targeted for in vitro translational screens. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23946910  Title: Beyond Widespread Mecp2 Deletions to Model Rett Syndrome: Conditional Spatio-Temporal Knockout, Single-Point Mutations and Transgenic Rescue Mice.  Rett syndrome (RTT) is one of the leading causes of intellectual disabilities in women. In addition to a few autistic features, characteristic symptoms that distinguish from classical autism include stereotypic hand movements, motor coordination deficits, breathing abnormalities, seizures and loss of acquired speech as well as purposeful hand use. RTT is highly associated with MECP2, the gene encoding for the transcription factor that binds methylated Cytosine in C-p-G islands in DNA, controlling gene expression and chromatin remodeling. In this review, we will briefly discuss current perspectives on MeCP2 function, and then will describe in detail novel mouse models of RTT based on loss-of-function of Mecp2 and their use for establishing rescue models, wherein we pay close attention to behavioral and morphological phenotypes. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22194327  Title: 7,8-dihydroxyflavone exhibits therapeutic efficacy in a mouse model of Rett syndrome.  Rett syndrome (RTT), caused by mutations in the methyl-CpG binding protein 2 gene (MECP2), is a debilitating autism spectrum developmental disorder predominantly affecting females. Mecp2 mutant mice have reduced levels of brain-derived neurotrophic factor (BDNF) in the brain; conditional deletion and overexpression of BDNF in the brain accelerates and slows, respectively, disease progression in Mecp2 mutant mice. Thus we tested the hypothesis that 7,8-dihydroxyflavone (7,8-DHF), a small molecule reported to activate the high affinity BDNF receptor (TrkB) in the CNS, would attenuate disease progression in Mecp2 mutant mice. Following weaning, 7,8-DHF was administered in drinking water throughout life. Treated mutant mice lived significantly longer compared with untreated mutant littermates (80 ± 4 and 66 ± 2 days, respectively). 7,8-DHF delayed body weight loss, increased neuronal nuclei size and enhanced voluntary locomotor (running wheel) distance in Mecp2 mutant mice. In addition, administration of 7,8-DHF partially improved breathing pattern irregularities and returned tidal volumes to near wild-type levels. Thus although the specific mechanisms are not completely known, 7,8-DHF appears to reduce disease symptoms in Mecp2 mutant mice and may have potential as a therapeutic treatment for RTT patients. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22138506  Title: Normal mitral cell dendritic development in the setting of Mecp2 mutation.  Rett syndrome (RTT) is an autism spectrum disorder caused by mutation in the gene encoding methyl CpG binding protein 2 (MECP2). Evidence to date suggests that these disorders display defects in synaptic organization and plasticity. A hallmark of the pathology in RTT has been identified as decreased dendritic arborization, which has been interpreted to represent abnormal dendritic formation and pruning during development. Our previous studies revealed that olfactory axons display defective pathfinding and targeting in the setting of Mecp2 mutation. In the present work, we use Mecp2 mutant mouse models and the olfactory system to investigate dendritic development. Here, we demonstrate that mitral cell dendritic development proceeds normally in mutant mice, resulting in typical dendritic morphology at early postnatal ages. We also failed to detect abnormalities in dendritic inputs at symptomatic stages when glomeruli from mutant mice appear smaller in area than the wild type (WT) (6 weeks postnatally). Collectively, these findings suggest that the initial defects in glomeruli impairment seen with Mecp2 mutation do not result from abnormal dendritic development. Our results using the olfactory system indicate that dendritic abnormalities are not an early feature in the abnormalities incurred by Mecp2 mutation. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22030357  Title: Elevated transcription factor specificity protein 1 in autistic brains alters the expression of autism candidate genes.  Profound changes in gene expression can result from abnormalities in the concentrations of sequence-specific transcription factors like specificity protein 1 (Sp1). Specificity protein 1 binding sites have been reported in the promoter regions of several genes implicated in autism. We hypothesize that dysfunction of Sp1 could affect the expression of multiple autism candidate genes, contributing to the heterogeneity of autism. We assessed any alterations in the expression of Sp1 and that of autism candidate genes in the postmortem brain (anterior cingulate gyrus [ACG], motor cortex, and thalamus) of autism patients (n = 8) compared with healthy control subjects (n = 13). Alterations in the expression of candidate genes upon Sp1/DNA binding inhibition with mithramycin and Sp1 silencing by RNAi were studied in SK-N-SH neuronal cells. We observed elevated expression of Sp1 in ACG of autism patients (p = .010). We also observed altered expression of several autism candidate genes. GABRB3, RELN, and HTR2A showed reduced expression, whereas CD38, ITGB3, MAOA, MECP2, OXTR, and PTEN showed elevated expression in autism. In SK-N-SH cells, OXTR, PTEN, and RELN showed reduced expression upon Sp1/DNA binding inhibition and Sp1 silencing. The RNA integrity number was not available for any of the samples. Transcription factor Sp1 is dysfunctional in the ACG of autistic brain. Consequently, the expression of potential autism candidate genes regulated by Sp1, especially OXTR and PTEN, could be affected. The diverse downstream pathways mediated by the Sp1-regulated genes, along with the environmental and intracellular signal-related regulation of Sp1, could explain the complex phenotypes associated with autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21982370  Title: Genome-wide activity-dependent MeCP2 phosphorylation regulates nervous system development and function.  Autism spectrum disorders such as Rett syndrome (RTT) have been hypothesized to arise from defects in experience-dependent synapse maturation. RTT is caused by mutations in MECP2, a nuclear protein that becomes phosphorylated at S421 in response to neuronal activation. We show here that disruption of MeCP2 S421 phosphorylation in vivo results in defects in synapse development and behavior, implicating activity-dependent regulation of MeCP2 in brain development and RTT. We investigated the mechanism by which S421 phosphorylation regulates MeCP2 function and show by chromatin immunoprecipitation-sequencing that this modification occurs on MeCP2 bound across the genome. The phosphorylation of MeCP2 S421 appears not to regulate the expression of specific genes; rather, MeCP2 functions as a histone-like factor whose phosphorylation may facilitate a genome-wide response of chromatin to neuronal activity during nervous system development. We propose that RTT results in part from a loss of this experience-dependent chromatin remodeling. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21966470  Title: Isogenic pairs of wild type and mutant induced pluripotent stem cell (iPSC) lines from Rett syndrome patients as in vitro disease model.  Rett syndrome (RTT) is an autism spectrum developmental disorder caused by mutations in the X-linked methyl-CpG binding protein 2 (MECP2) gene. Excellent RTT mouse models have been created to study the disease mechanisms, leading to many important findings with potential therapeutic implications. These include the identification of many MeCP2 target genes, better understanding of the neurobiological consequences of the loss- or mis-function of MeCP2, and drug testing in RTT mice and clinical trials in human RTT patients. However, because of potential differences in the underlying biology between humans and common research animals, there is a need to establish cell culture-based human models for studying disease mechanisms to validate and expand the knowledge acquired in animal models. Taking advantage of the nonrandom pattern of X chromosome inactivation in female induced pluripotent stem cells (iPSC), we have generated isogenic pairs of wild type and mutant iPSC lines from several female RTT patients with common and rare RTT mutations. R294X (arginine 294 to stop codon) is a common mutation carried by 5-6% of RTT patients. iPSCs carrying the R294X mutation has not been studied. We differentiated three R294X iPSC lines and their isogenic wild type control iPSC into neurons with high efficiency and consistency, and observed characteristic RTT pathology in R294X neurons. These isogenic iPSC lines provide unique resources to the RTT research community for studying disease pathology, screening for novel drugs, and testing toxicology. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21909962  Title: Mecp2 truncation in male mice promotes affiliative social behavior.  Mouse models of Rett syndrome, with targeted mutations in the Mecp2 gene, show a high degree of phenotypic consistency with the clinical syndrome. In addition to severe and age-specific regression in motor and cognitive abilities, a variety of studies have demonstrated that Mecp2 mutant mice display impaired social behavior. Conversely, other studies indicate complex enhancements of social behavior in Mecp2 mutant mice. Since social behavior is a complicated accumulation of constructs, we performed a series of classic and refined social behavior tasks and revealed a relatively consistent pattern of enhanced pro-social behavior in hypomorphic Mecp2 (308/Y) mutant mice. Analyses of repetitive motor acts, and cognitive stereotypy did not reveal any profound differences due to genotype. Taken together, these results suggest that the mutations associated with Rett syndrome are not necessarily associated with autism-relevant social impairment in mice. However, this gene may be a valuable candidate for revealing basic mechanisms of affiliative behavior. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21840925  Title: 15q11.2-13.3 chromatin analysis reveals epigenetic regulation of CHRNA7 with deficiencies in Rett and autism brain.  Copy number variations (CNVs) within human 15q11.2-13.3 show reduced penetrance and variable expressivity in a range of neurologic disorders. Therefore, characterizing 15q11.2-13.3 chromatin structure is important for understanding the regulation of this locus during normal neuronal development. Deletion of the Prader-Willi imprinting center (PWS-IC) within 15q11.2-13.3 disrupts long-range imprinted gene expression resulting in Prader-Willi syndrome. Previous results establish that MeCP2 binds to the PWS-IC and is required for optimal expression of distal GABRB3 and UBE3A. To examine the hypothesis that MeCP2 facilitates 15q11.2-13.3 transcription by linking the PWS-IC with distant elements, chromosome capture conformation on chip (4C) analysis was performed in human SH-SY5Y neuroblastoma cells. SH-SY5Y neurons had 2.84-fold fewer 15q11.2-13.3 PWS-IC chromatin interactions than undifferentiated SH-SY5Y neuroblasts, revealing developmental chromatin de-condensation of the locus. Out of 68 PWS-IC interactions with15q11.2-13.3 identified by 4C analysis and 62 15q11.2-13.3 MeCP2-binding sites identified by previous ChIP-chip studies, only five sites showed overlap. Remarkably, two of these overlapping PWS-IC- and MeCP2-bound sites mapped to sites flanking CHRNA7 (cholinergic receptor nicotinic alpha 7) encoding the cholinergic receptor, nicotinic, alpha 7. PWS-IC interaction with CHRNA7 in neurons was independently confirmed by fluorescent in situ hybridization analysis. Subsequent quantitative transcriptional analyses of frontal cortex from Rett syndrome and autism patients revealed significantly reduced CHRNA7 expression compared with controls. Together, these results suggest that transcription of CHRNA7 is modulated by chromatin interactions with the PWS-IC. Thus, loss of long-range chromatin interactions within 15q11.2-13.3 may contribute to multiple human neurodevelopmental disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21733672  Title: Synaptic microcircuit dysfunction in genetic models of neurodevelopmental disorders: focus on Mecp2 and Met.  Recent findings in the genetics of neurodevelopmental syndromes have ushered in an exciting era of discovery in which substrates of neurologic dysfunction are being identified at the synaptic and microcircuit levels in mouse models of these disorders. We review recent progress in this area, focusing on two examples of mouse models of autism spectrum disorders (ASDs): Mecp2 models of Rett syndrome, and a Met-knockout model of non-syndromic forms of autism. In both cases, a dominant theme is changes in synaptic strength, associated with hyper-connectivity or hypo-connectivity in specific microcircuits. Alterations in intrinsic neuronal excitability are also found, but do not appear to be as common. The microcircuit-specific nature of synaptic changes observed in these ASD models indicates that it will be necessary to define mechanisms of circuit dysfunction on a case-by-case basis, not only in neocortex but also in brainstem and other sub-cortical areas. Thus, functional microcircuit analysis is emerging as an important line of investigation, highly complementary to neurogenetic and molecular strategies, and holds promise for generating models of the underlying pathophysiology and for guiding development of novel therapeutic strategies. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21721946  Title: The role of MeCP2 in the brain.  Methyl-CpG binding protein 2 (MeCP2) was first identified in 1992 as a protein that binds specifically to methylated DNA. Mutations in the MECP2 gene were later found to be the cause of an autism spectrum disorder, Rett syndrome. Despite almost 20 years of research into the molecular mechanisms of MeCP2 function, many questions are yet to be answered conclusively. This review considers several key questions and attempts to evaluate the current state of evidence. For example, is MeCP2 just a methyl-CpG binding protein? Is it a multifunctional protein or primarily a transcriptional repressor? We also consider whether MeCP2, as a chromosome-binding protein, acts at specific sites within the genome or more globally, and in which cell types it is functionally important. Finally, we consider two alternative views of MeCP2 in the brain: as a regulator of brain development or as a factor that helps maintain neuronal/glial function. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21629824  Title: Habituation without NMDA Receptor-Dependent Desensitization of Hering-Breuer Apnea Reflex in a Mecp2 Mutant Mouse Model of Rett Syndrome.  Non-associative learning is a basic neuroadaptive behavior exhibited in almost all animal species and sensory modalities but its functions and mechanisms in the mammalian brain are poorly understood. Previous studies have identified two distinct forms of non-associative learning in the classic Hering-Breuer inflation reflex (HBIR) induced apnea in rats: NMDA receptor (NMDAR)-independent habituation in a primary vagal pathway and NMDAR-dependent desensitization in a secondary pontine pathway. Here, we show that abnormal non-associative learning of the HBIR may underlie the endophenotypic tachypnea in an animal model of Rett syndrome (RTT), an autism-spectrum disorder caused by mutations in the X-linked gene encoding methyl-CpG-binding protein 2 (MECP2). Mecp2(+/-) symptomatic mice on a mixed-strain background demonstrated significantly increased resting respiratory frequency with shortened expiration and normal inspiratory duration compared with asymptomatic mutants and wild-type controls, a phenotype that is characteristic of girls with RTT. Low-intensity electrical stimulation of the vagus nerve elicited fictive HBIR with time-dependent habituation in both Mecp2(+/-) and wild-type mice. However, time-dependent desensitization of the HBIR was evidenced only in wild-type controls and asymptomatic mutant mice but was absent or suppressed in Mecp2(+/-) symptomatic mice or in wild-type mice after blockade of NMDAR with dizocilpine. Remarkably, ∼50% of the Mecp2(+/-) mice developed these X-linked phenotypes despite somatic mosaicism. Such RTT-like respiratory endophenotypes in mixed-strain Mecp2(+/-) mice differed from those previously reported in Mecp2(-/y) mice on pure C57BL/6J background. These findings provide the first evidence indicating that impaired NMDAR-dependent desensitization of the HBIR may contribute to the endophenotypic tachypnea in RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21624480  Title: Cognitive deficits in Rett syndrome: what we know and what we need to know to treat them.  Rett syndrome is an autism spectrum disorder and a leading cause of severe mental retardation in girls. The nature of the cognitive abnormalities in Rett, as described in humans and other animal models, and its potential reversibility and treatment are the subject of this review. Rett syndrome is associated with severe mental retardation and a host of impairments that include social and motor deficits, and respiratory and bone abnormalities. More than 80% of Rett girls have mutations in the gene that encodes MeCP2, which is a protein with a complex set of functions that include transcriptional repression and activation. The complex phenotype associated with Rett and the knowledge of the causal genetic mutation provide a unique opportunity within the autism spectrum to explore the relationship between transcriptional control, brain abnormalities and specific behavioral functions, importantly the elusive cognitive dysfunctions associated with mental retardation. The nature of the cognitive abnormalities related to Rett and the potential reversibility and treatment of these abnormalities have not been studied as extensively as some of the other aspects of the Rett phenotype. The cognitive phenotype associated with Rett is also less well studied relative to that in other well known developmental disorders, such as Down syndrome and Fragile X. Nevertheless, some recent studies provide hope that the cognitive impairments, as well as other symptoms of Rett, can be rescued. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21531908  Title: Analysis of MECP2 gene copy number in boys with autism.  Autism is a severe neurodevelopmental disorder with a strong genetic basis.The methyl-CpG binding protein 2 gene (MECP2) is a dosage-sensitive gene in brain development and has been implicated as a candidate gene for autism. Duplication of the MECP2 gene has been reported in a few boys with autistic features. To further investigate the association of MECP2 duplication with autism, the authors performed real-time quantitative polymerase chain reaction (PCR) to detect copy number variations of the MECP2 gene in 82 autistic boys. No copy number variation was found in these patients, indicating that duplication of the MECP2 gene is not frequent in autistic patients. The authors consider that duplication of the MECP2 gene has no major effect on the susceptibility to autism. Replication of studies in a large-sized sample and a well-characterized subgroup of autism are warranted to further identify the association of MECP2 gene duplication with autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21442598  Title: [Genetic analyses for identifying molecular mechanisms in autism spectrum disorders].  Autism spectrum disorders (ASD) are severe neurodevelopmental disorders with marked deficits in social communication, verbal development, and behaviour. The broad phenotype and the clinical heterogeneity point to a polygenic disorder - despite high heritability among siblings. According to recent findings not only do single-rare mutations but also copy number variations and single nucleotide polymorphisms impact the ASD phenotype. Because of the scope of national and international consortia, many linkage and genome-wide association studies have evolved which elucidate candidate and susceptibility genomic regions and genes relevant for ASD. In contrast to polygenic or genetic complex models for autism, a few monogenetic forms of ASD are known to be caused by single gene defects, e.g., fragile-X syndrome. Knock-out animal models of monogenetic autism (e.g. FMRP(-/-)) or neurodegenerative disorders (e.g. MeCP2(-/-)) are often used to analyze the molecular mechanisms underlying ASD. In this review we describe the state of the art of genome analyses in ASD, the most widely used mouse models for polygenic or monogenetic forms of autism and discuss new insights gained from these analyses. The susceptibility genes so far identified seem to be involved in the proper establishment of the synaptic cleft, the secretion of surface proteins, or the overall cellular translation processes. Theses findings suggest that impacting translation-dependent processes like synaptic plasticity or cell-to-cell connectivity may lead to an ASD phenotype. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21372149  Title: Isolation of MECP2-null Rett Syndrome patient hiPS cells and isogenic controls through X-chromosome inactivation.  Rett syndrome (RTT) is a neurodevelopmental autism spectrum disorder that affects girls due primarily to mutations in the gene encoding methyl-CpG binding protein 2 (MECP2). The majority of RTT patients carry missense and nonsense mutations leading to a hypomorphic MECP2, while null mutations leading to the complete absence of a functional protein are rare. MECP2 is an X-linked gene subject to random X-chromosome inactivation resulting in mosaic expression of mutant MECP2. The lack of human brain tissue motivates the need for alternative human cellular models to study RTT. Here we report the characterization of a MECP2 mutation in a classic female RTT patient involving rearrangements that remove exons 3 and 4 creating a functionally null mutation. To generate human neuron models of RTT, we isolated human induced pluripotent stem (hiPS) cells from RTT patient fibroblasts. RTT-hiPS cells retained the MECP2 mutation, are pluripotent and fully reprogrammed, and retained an inactive X-chromosome in a nonrandom pattern. Taking advantage of the latter characteristic, we obtained a pair of isogenic wild-type and mutant MECP2 expressing RTT-hiPS cell lines that retained this MECP2 expression pattern upon differentiation into neurons. Phenotypic analysis of mutant RTT-hiPS cell-derived neurons demonstrated a reduction in soma size compared with the isogenic control RTT-hiPS cell-derived neurons from the same RTT patient. Analysis of isogenic control and mutant hiPS cell-derived neurons represents a promising source for understanding the pathogenesis of RTT and the role of MECP2 in human neurons. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21328568  Title: Behavioral profiles of mouse models for autism spectrum disorders.  Autism spectrum disorders (ASD) are characterized by impairments in reciprocal social communication, and stereotyped verbal and nonverbal behaviors. In approximately 10-25% of the affected individuals, a genetic mutation associated with the condition can be identified. Recently, mutations altering synapse formation, cellular/synaptic growth rate and regulation of excitatory and inhibitory currents were identified in patients with intellectual disability, typical autism, Asperger syndrome or neurological syndromes associated with autistic traits. Following these genetic findings, mouse models carrying mutations similar to those identified in patients have been generated. These models offer the opportunity to investigate in vivo the physiological and behavioral consequences of the mutations. Here, we review the existing data on the phenotypes of mice carrying mutations in genes associated with ASD including neuroligin, neurexin and Shank mutant mice as well as the Fmr1, Mecp2, Ube3a, Nf1, Pten and Tsc1/Tsc2 mutant mice. The diversity and complexity of the phenotype of these mouse models reflect the broad range of phenotypes observed in patients with ASD. Remarkably, results from therapeutic approaches (e.g., modulation of gene expression, administration of pharmacological and nonpharmacological substances, enriched environment) are encouraging since some behavioral alterations could be reversed even when treatment was performed on adult mice. These ongoing studies should therefore increase our understanding of the biological alterations associated with ASD as well as the development of knowledge-based treatments. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21316312  Title: Autonomic dysfunction with mutations in the gene that encodes methyl-CpG-binding protein 2: insights into Rett syndrome.  Rett syndrome (RTT) is an autism spectrum disorder with an incidence of ~1:10,000 females (reviewed in Bird, 2008; Chahrour et al., 2007; Francke, 2006). Affected individuals are apparently normal at birth. Between 6-18 months of age, however, RTT patients begin to exhibit deceleration of head growth, replacement of purposeful hand movements with stereotypic hand wringing, loss of speech, social withdrawal and other autistic features. RTT is caused by loss of function mutations in the gene that encodes methyl-CpG-binding protein 2 (Mecp2) (Amir et al., 1999), a transcriptional repressor that targets genes essential for neuronal survival, dendritic growth, synaptogenesis, and activity dependent plasticity. MECP2 is X-linked, and males die soon after birth. Included in the RTT phenotype are cardiorespiratory disorders involving the autonomic nervous system. The respiratory disorders, including the roles of bioaminergic and brain derived neurotrophic factor (BDNF) signaling in the respiratory pathophysiology of RTT have been recently reviewed (Bissonnette et al., 2007a; Ogier et al., 2008; Katz et al., 2009). Here we will cover the work on RTT regarding respiration that has appeared since 2009 as well as cardiovascular abnormalities. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21239731  Title: Experimental models of Rett syndrome based on Mecp2 dysfunction.  Rett syndrome (RTT) is a neurodevelopmental disorder predominantly occurring in females with an incidence of 1:10,000 births and caused by sporadic mutations in the MECP2 gene, which encodes methyl-CpG-binding protein-2, an epigenetic transcription factor that binds methylated DNA. The clinical hallmarks include a period of apparently normal early development followed by a plateau and then subsequent frank regression. Impaired visual and aural contact often lead to an initial diagnosis of autism. The characterization of experimental models based on the loss-of-function of the mouse Mecp2 gene revealed that subtle changes in the morphology and function of brain cells and synapses have profound consequences on network activities that underlie critical brain functions. Furthermore, these experimental models have been used for successful reversals of RTT-like symptoms by genetic, pharmacological and environmental manipulations, raising hope for novel therapeutic strategies to improve the quality of life of RTT individuals. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20956852  Title: The role of calcium-dependent gene expression in autism spectrum disorders: lessons from MeCP2, Ube3a and beyond.  During the last decade, autism spectrum disorders (ASD) have become the center of attention where several branches of modern biology unexpectedly meet, such as neural development, molecular biology, epigenetics, neurophysiology and psychiatry. This review will focus on the molecular mechanism by which calcium-dependent gene expression regulates brain development and how ASD may occur if this process is compromised. Specifically, the studies of the calcium-dependent transcriptional repressor MeCP2 gave us much insight about how abnormal development may lead to ASD. Most recently, studies about Ube3a, a critical component of the ubiquitination system enzyme, shed light on how neural activity regulates synapse function through the protein degradation pathway. Taken together, these studies suggest that ASD may be caused by the incapability of neurons to generate adaptive responses via regulating gene expression upon incoming activity. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20921395  Title: Correction of respiratory disorders in a mouse model of Rett syndrome.  Rett syndrome (RTT) is an autism spectrum disorder caused by mutations in the X-linked gene that encodes the transcription factor methyl-CpG-binding protein 2 (MeCP2). A major debilitating phenotype in affected females is frequent apneas, and heterozygous Mecp2-deficient female mice mimic the human respiratory disorder. GABA defects have been demonstrated in the brainstem of Mecp2-deficient mice. Here, using an intact respiratory network, we show that apnea in RTT mice is characterized by excessive excitatory activity in expiratory cranial and spinal nerves. Augmenting GABA markedly improves the respiratory phenotype. In addition, a serotonin 1a receptor agonist that depresses expiratory neuron activity also reduces apnea, corrects the irregular breathing pattern, and prolongs survival in MeCP2 null males. Combining a GABA reuptake blocker with a serotonin 1a agonist in heterozygous females completely corrects their respiratory defects. The results indicate that GABA and serotonin 1a receptor activity are candidates for treatment of the respiratory disorders in Rett syndrome. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20425298  Title: The role of MeCP2 in brain development and neurodevelopmental disorders.  Methyl CpG binding protein-2 (MeCP2) is an essential epigenetic regulator in human brain development. Rett syndrome, the primary disorder caused by mutations in the X-linked MECP2 gene, is characterized by a period of cognitive decline and development of hand stereotypies and seizures following an apparently normal early infancy. In addition, MECP2 mutations and duplications are observed in a spectrum of neurodevelopmental disorders, including severe neonatal encephalopathy, X-linked mental retardation, and autism, implicating MeCP2 as an essential regulator of postnatal brain development. In this review, we compare the mutation types and inheritance patterns of the human disorders associated with MECP2. In addition, we summarize the current understanding of MeCP2 as a central epigenetic regulator of activity-dependent synaptic maturation. As MeCP2 occupies a central role in the pathogenesis of multiple neurodevelopmental disorders, continued investigation into MeCP2 function and regulatory pathways may show promise for developing broad-spectrum therapies. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20392956  Title: Rett syndrome microglia damage dendrites and synapses by the elevated release of glutamate.  MECP2, an X-linked gene encoding the epigenetic factor methyl-CpG-binding protein-2, is mutated in Rett syndrome (RTT) and aberrantly expressed in autism. Most children affected by RTT are heterozygous Mecp2(-/+) females whose brain function is impaired postnatally due to MeCP2 deficiency. Recent studies suggest a role of glia in causing neuronal dysfunction via a non-cell-autonomous effect in RTT. Here we report a potent neurotoxic activity in the conditioned medium (CM) obtained from Mecp2-null microglia. Hippocampal neurons treated with CM from Mecp2-null microglia showed an abnormal stunted and beaded dendritic morphology, and signs of microtubule disruption and damage of postsynaptic glutamatergic components within 24 h. We identified that the toxic factor in the CM is glutamate, because (1) Mecp2-null microglia released a fivefold higher level of glutamate, (2) blockage of microglial glutamate synthesis by a glutaminase inhibitor abolished the neurotoxic activity, (3) blockage of microglial glutamate release by gap junction hemichannel blockers abolished the neurotoxic activity, and (4) glutamate receptor antagonists blocked the neurotoxicity of the Mecp2-null microglia CM. We further identified that increased levels of glutaminase and connexin 32 in Mecp2-null microglia are responsible for increased glutamate production and release, respectively. In contrast, the CM from highly pure Mecp2-null astrocyte cultures showed no toxic effect. Our results suggest that microglia may influence the onset and progression of RTT and that microglia glutamate synthesis or release could be a therapeutic target for RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20172507  Title: Early environmental enrichment moderates the behavioral and synaptic phenotype of MeCP2 null mice.  Rett syndrome (RTT) is an X-linked progressive neurodevelopmental disorder characterized by a variety of symptoms including motor abnormalities, mental retardation, anxiety, and autism. Most of RTT cases are caused by mutations of MeCP2. In mice, impaired MeCP2 function results in synaptic deficits associated with motor, cognitive, and emotional alterations. Environmental enrichment (EE) is a rearing condition that enhances synapse formation and plasticity. Previous studies analyzing the effects of postweaning EE found limited effects on motor performance of male MeCP2 mutants. However, EE during early postnatal development produces powerful effects on neural development and plasticity. Thus, we tested whether early EE could ameliorate several phenotypes of male homozygous and female heterozygous MeCP2 mutants. We investigated the effects of early EE on motor coordination, structural and functional synaptic plasticity, and brain-derived neurotrophic factor expression in male MeCP2 null mice. Anxiety-related behavior and spatial learning was analyzed in heterozygous MeCP2 female mice. In male mutants, EE modified excitatory and to a lesser extent inhibitory synaptic density in cerebellum and cortex, reversed the cortical long-term potentiation deficit and augmented cortical brain-derived neurotrophic factor levels. Environmental enrichment also ameliorated motor coordination and motor learning. In female heterozygous mice, a model closely mimicking some aspects of RTT symptoms, EE rescued memory deficits in the Morris water maze and decreased anxiety-related behavior. Early EE dramatically improves several phenotypes of MeCP2 mutants. Thus, environmental factors should be taken into account when analyzing phenotypes of MeCP2 knockout mice, an accepted model of RTT. Early EE might be beneficial in RTT patients. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20093853  Title: MeCP2 deficiency downregulates specific nuclear proteins that could be partially recovered by valproic acid in vitro.  MeCP2, the major causative factor of Rett syndrome and related phenotypes including autism, is a two-face nuclear modulator acting via transcriptional and chromatin remodeling mechanisms. This study investigated the expression of several nuclear proteins and their dependence on MeCP2 dose and presence of the Rett causative R306C mutation. To this end, we developed in vitro models representing MeCP2 deficiency induced by siRNAs, and cells expressing the R306C mutation. Using an extended antibody microarray validated by specific assays, revealed that MeCP2 dose was correlated with specific nuclear proteins profiles including the BRM/SNF2 component of SWI/SNF complex, PRMT1 methyl transferase and HDAC2. Furthermore, while exposing the MeCP2 knock-down system to therapeutic concentrations of valproic acid (VPA), a known HDACs inhibitor, we observed a partial restoration of MeCP2 expression levels. Exposure to VPA also increased the levels of BRM, as well as of BDNF, an important co-factor in MeCP2-mediated pathway. Our findings provide additional evidence of diverse mechanisms of MeCP2 function as transcriptional repressor and activator of specific genes. As it has been recently demonstrated that post-natal restoration of MeCP2 deficiency may reverse neurological defects in a mouse model of Rett syndrome, we suggest to study the restorative effect of HDAC inhibitors in MeCP2-deficient mouse model. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/20045424  Title: Cognitive and social functions and growth factors in a mouse model of Rett syndrome.  Rett syndrome (RTT) is an autism-spectrum disorder caused by mutations in the X-linked gene encoding methyl-CpG-binding protein 2 (MeCP2). Abnormalities in social behavior, stereotyped movements, and restricted interests are common features in both RTT and classic autism. While mouse models of both RTT and autism exist, social behaviors have not been explored extensively in mouse models of RTT. Here, we report cognitive and social abnormalities in Mecp2(1lox) null mice, an animal model of RTT. The null mice show severe deficits in short- and long-term object recognition memories, reminiscent of the severe cognitive deficits seen in RTT girls. Social behavior, however, is abnormal in that the null mice spend more time in contact with stranger mice than do wildtype controls. These findings are consistent with reports of increased reciprocal social interaction in RTT girls relative to classic autism. We also report here that the levels of the neurotrophins brain-derived neurotrophic factor (BDNF), insulin-like growth factor-1 (IGF-1), and nerve growth factor (NGF) are decreased in the hippocampus of the null mice, and discuss how this may provide an underlying mechanism for both the cognitive deficits and the increased motivation for social contact observed in the Mecp2(1lox) null mice. These studies support a differential etiology between RTT and autism, particularly with respect to sociability deficits. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/21423514  Title: Synaptic determinants of rett syndrome.  There is mounting evidence showing that the structural and molecular organization of synaptic connections is affected both in human patients and in animal models of neurological and psychiatric diseases. As a consequence of these experimental observations, it has been introduced the concept of synapsopathies, a notion describing brain disorders of synaptic function and plasticity. A close correlation between neurological diseases and synaptic abnormalities is especially relevant for those syndromes including also mental retardation in their symptomatology, such as Rett syndrome (RS). RS (MIM312750) is an X-linked dominant neurological disorder that is caused in the majority of cases by mutations in methyl-CpG-binding protein 2 (MeCP2). This review will focus on the current knowledge of the synaptic alterations produced by mutations of the gene MeCP2 in mouse models of RS and will highlight prospects experimental therapies currently in use. Different experimental approaches have revealed that RS could be the consequence of an impairment in the homeostasis of synaptic transmission in specific brain regions. Indeed, several forms of experience-induced neuronal plasticity are impaired in the absence of MeCP2. Based on the results presented in this review, it is reasonable to propose that understanding how the brain is affected by diseases such as RS is at reach. This effort will bring us closer to identify the neurobiological bases of human cognition. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19812320  Title: Synaptic circuit abnormalities of motor-frontal layer 2/3 pyramidal neurons in an RNA interference model of methyl-CpG-binding protein 2 deficiency.  Rett syndrome, an autism spectrum disorder with prominent motor and cognitive features, results from mutations in the gene for methyl-CpG-binding protein 2 (MeCP2). Here, to identify cortical circuit abnormalities that are specifically associated with MeCP2 deficiency, we used glutamate uncaging and laser scanning photostimulation to survey intracortical networks in mouse brain slices containing motor-frontal cortex. We used in utero transfection of short hairpin RNA constructs to knock down MeCP2 expression in a sparsely distributed subset of layer (L) 2/3 pyramidal neurons in wild-type mice, and compared input maps recorded from transfected-untransfected pairs of neighboring neurons. The effect of MeCP2 deficiency on local excitatory input pathways was severe, with an average reduction in excitatory synaptic input from middle cortical layers (L3/5A) of >30% compared with MeCP2-replete controls. MeCP2 deficiency primarily affected the strength, rather than the topography, of excitatory intracortical pathways. Inhibitory synaptic inputs and intrinsic eletrophysiological properties were unaffected in the MeCP2-knockdown neurons. These studies indicate that MeCP2 deficiency in individual postsynaptic cortical pyramidal neurons is sufficient to induce a pathological synaptic defect in excitatory intracortical circuits. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19741133  Title: Intact long-term potentiation but reduced connectivity between neocortical layer 5 pyramidal neurons in a mouse model of Rett syndrome.  Mutations in MECP2 cause Rett syndrome and some related forms of mental retardation and autism. Mecp2-null mice exhibit symptoms reminiscent of Rett syndrome including deficits in learning. Previous reports demonstrated impaired long-term potentiation (LTP) in slices of symptomatic Mecp2-null mice, and decreased excitatory neurotransmission, but the causal relationship between these phenomena is unclear. Reduced plasticity could lead to altered transmission, or reduced excitatory transmission could alter the ability to induce LTP. To help distinguish these possibilities, we compared LTP induction and baseline synaptic transmission at synapses between layer 5 cortical pyramidal neurons in slices of wild-type and Mecp2-null mice. Paired recordings reveal that LTP induction mechanisms are intact in Mecp2-null connections, even after the onset of symptoms. However, fewer connections were found in Mecp2-null mice and individual connections were weaker. These data suggest that loss of MeCP2 function reduces excitatory synaptic connectivity and that this precedes deficits in plasticity. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19710912  Title: MECP2 isoform-specific vectors with regulated expression for Rett syndrome gene therapy.  Rett Syndrome (RTT) is an Autism Spectrum Disorder and the leading cause of mental retardation in females. RTT is caused by mutations in the Methyl CpG-Binding Protein-2 (MECP2) gene and has no treatment. Our objective is to develop viral vectors for MECP2 gene transfer into Neural Stem Cells (NSC) and neurons suitable for gene therapy of Rett Syndrome. We generated self-inactivating (SIN) retroviral vectors with the ubiquitous EF1alpha promoter avoiding known silencer elements to escape stem-cell-specific viral silencing. High efficiency NSC infection resulted in long-term EGFP expression in transduced NSC and after differentiation into neurons. Infection with Myc-tagged MECP2-isoform-specific (E1 and E2) vectors directed MeCP2 to heterochromatin of transduced NSC and neurons. In contrast, vectors with an internal mouse Mecp2 promoter (MeP) directed restricted expression only in neurons and glia and not NSC, recapitulating the endogenous expression pattern required to avoid detrimental consequences of MECP2 ectopic expression. In differentiated NSC from adult heterozygous Mecp2(tm1.1Bird)+/- female mice, 48% of neurons expressed endogenous MeCP2 due to random inactivation of the X-linked Mecp2 gene. Retroviral MECP2 transduction with EF1alpha and MeP vectors rescued expression in 95-100% of neurons resulting in increased dendrite branching function in vitro. Insulated MECP2 isoform-specific lentiviral vectors show long-term expression in NSC and their differentiated neuronal progeny, and directly infect dissociated murine cortical neurons with high efficiency. MeP vectors recapitulate the endogenous expression pattern of MeCP2 in neurons and glia. They have utility to study MeCP2 isoform-specific functions in vitro, and are effective gene therapy vectors for rescuing dendritic maturation of neurons in an ex vivo model of RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19386901  Title: Rett syndrome astrocytes are abnormal and spread MeCP2 deficiency through gap junctions.  MECP2, an X-linked gene encoding the epigenetic factor methyl-CpG-binding protein-2, is mutated in Rett syndrome (RTT) and aberrantly expressed in autism. Most children affected by RTT are heterozygous Mecp2-/+ females whose brain function is impaired postnatally due to MeCP2 deficiency. While prior functional investigations of MeCP2 have focused exclusively on neurons and have concluded the absence of MeCP2 in astrocytes, here we report that astrocytes express MeCP2, and MeCP2 deficiency in astrocytes causes significant abnormalities in BDNF regulation, cytokine production, and neuronal dendritic induction, effects that may contribute to abnormal neurodevelopment. In addition, we show that the MeCP2 deficiency state can progressively spread at least in part via gap junction communications between mosaic Mecp2-/+ astrocytes in a novel non-cell-autonomous mechanism. This mechanism may lead to the pronounced loss of MeCP2 observed selectively in astrocytes in mouse Mecp2-/+ brain, which is coincident with phenotypic regression characteristic of RTT. Our results suggest that astrocytes are viable therapeutic targets for RTT and perhaps regressive forms of autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19138667  Title: Altered expression of Autism-associated genes in the brain of Fragile X mouse model.  Autism is a severe neurodevelopmental disorder, which typically emerges in early childhood. Most cases of autism have not been linked to mutations in a specific gene, and the etioloty of the disorder remains to be established [S.S. Moy, J.J. Nadler, T.R. Magnuson, J.N. Crawley, Mouse models of autism spectrum disorders: the challenge for behavioral genetics, Am. J. Med. Genet. 142 (2006) 40-51]. Fragile X syndrome is caused by mutation in the FMR1 gene and is characterized by mental retardation, physical abnormalities, and, in most case, autistic-like behavior [R.J. Hagerman, A.W. Jackson, A. Levitas, B. Rimland, M. Braden, An analysis of autism in fifty males with the Fragile X syndrome, Am. J. Med. Genet. 23 (1986) 359-374, C.E. Bakker, C. Verheij, R. Willemsen, R. van der Helm, F. Oerlemans, M. Vermeij, A. Bygrave, A.T. Hoogeveen, B.A. Oostra, E. Reyniers, K. De Boulle, R. D'Hooge, P. Cras, D. van Velzen, G. Nagels, J.J. Marti, P. De Deyn, J.K. Darby, P.J. Willems, Fmr1 knockout mice: a model to study Fragile X mental retardation, Cell 78 (1994) 23-33]. The FMR1 knockout (KO) mouse is one of the best characterized animal models for human disorders associated with autism [S.S. Moy, J.J. Nadler, T.R. Magnuson, J.N. Crawley, Mouse models of autism spectrum disorders: the challenge for behavioral genetics, Am. J. Med. Genet. 142 (2006) 40-51]. We have used real-time PCR to investigate changes in expression levels of three genes: WNT2, MECP2, and FMR1 in different brain regions of Fagile X mice and litter mate controls. We found major changes in the expression pattern for the three genes examined. FMR1, MECP2, and WNT2 expression were drastically down regulated in the Fragile X mouse brain. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18817733  Title: Deletion of Mecp2 in Sim1-expressing neurons reveals a critical role for MeCP2 in feeding behavior, aggression, and the response to stress.  Rett Syndrome (RTT) is an autism spectrum disorder caused by mutations in the X-linked gene encoding methyl-CpG binding protein 2 (MeCP2). In order to map the neuroanatomic origins of the complex neuropsychiatric behaviors observed in patients with RTT and to uncover endogenous functions of MeCP2 in the hypothalamus, we removed Mecp2 from Sim1-expressing neurons in the hypothalamus using Cre-loxP technology. Loss of MeCP2 in Sim1-expressing neurons resulted in mice that recapitulated the abnormal physiological stress response that is seen upon MeCP2 dysfunction in the entire brain. Surprisingly, we also uncovered a role for MeCP2 in the regulation of social and feeding behaviors since the Mecp2 conditional knockout (CKO) mice were aggressive, hyperphagic, and obese. This study demonstrates that deleting Mecp2 in a defined brain region is an excellent approach to map the neuronal origins of complex behaviors and provides new insight about the function of MeCP2 in specific neurons. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18631120  Title: The methyl-CpG-binding protein MeCP2 and neurological disease.  The methyl-CpG-binding protein MeCP2 was discovered over 15 years ago as part of a search for proteins that selectively bind methylated DNA. It is a nuclear protein that is largely chromatin-bound and has a strong preference for binding to methylated DNA sequences in vivo. Evidence from model systems shows that MeCP2 can recruit the Sin3a co-repressor complex to promoters leading to transcriptional repression, therefore suggesting that MeCP2 can interpret the DNA methylation signal to bring about gene silencing. Mutations in the human MECP2 gene cause the autism spectrum disorder Rett Syndrome. MeCP2 is most highly expressed in neurons, and mice lacking this protein show symptoms that strikingly parallel those of Rett patients. Surprisingly, these symptoms are efficiently reversed by delayed activation of a 'stopped' Mecp2 gene, raising hopes that human Rett syndrome may also be reversible. Future studies of MeCP2 promise to shed light upon brain function, neurological disease and the biology of DNA methylation. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18321864  Title: A partial loss of function allele of methyl-CpG-binding protein 2 predicts a human neurodevelopmental syndrome.  Rett Syndrome, an X-linked dominant neurodevelopmental disorder characterized by regression of language and hand use, is primarily caused by mutations in methyl-CpG-binding protein 2 (MECP2). Loss of function mutations in MECP2 are also found in other neurodevelopmental disorders such as autism, Angelman-like syndrome and non-specific mental retardation. Furthermore, duplication of the MECP2 genomic region results in mental retardation with speech and social problems. The common features of human neurodevelopmental disorders caused by the loss or increase of MeCP2 function suggest that even modest alterations of MeCP2 protein levels result in neurodevelopmental problems. To determine whether a small reduction in MeCP2 level has phenotypic consequences, we characterized a conditional mouse allele of Mecp2 that expresses 50% of the wild-type level of MeCP2. Upon careful behavioral analysis, mice that harbor this allele display a spectrum of abnormalities such as learning and motor deficits, decreased anxiety, altered social behavior and nest building, decreased pain recognition and disrupted breathing patterns. These results indicate that precise control of MeCP2 is critical for normal behavior and predict that human neurodevelopmental disorders will result from a subtle reduction in MeCP2 expression. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18207372  Title: Rett syndrome in adults with severe intellectual disability: exploration of behavioral characteristics.  Rett syndrome is a genetically linked form of autism spectrum disorder (ASD) accompanied by intellectual disability (ID). The disorder is also characterized by cardiorespiratory dysregulation, disturbance in muscle tone, reduced brain growth and scoliosis. Over 300 studies have been published on the disorder, most of which has focused on identification of causative factors, which appears to be the result of mutations of gene MECP2. Rarely have adults with Rett syndrome been studied, and behavioral characteristics in these individuals are largely unknown. The present study aimed to extend what little is known about behavioral characteristics of Rett syndrome in adults, with particular emphasis on social, communicative, and adaptive behavior. Rett syndrome adults with severe ID were matched to autistic adults with ID and ID only controls. The implications of these data for more fully describing and diagnosing the condition in adults are discussed. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18203756  Title: MeCP2-dependent repression of an imprinted miR-184 released by depolarization.  Both fragile X syndrome and Rett syndrome are commonly associated with autism spectrum disorders and involve defects in synaptic plasticity. MicroRNA is implicated in synaptic plasticity because fragile X mental retardation protein was recently linked to the microRNA pathway. DNA methylation is also involved in synaptic plasticity since methyl CpG-binding protein 2 (MeCP2) is mutated in patients with Rett syndrome. Here we report that expression of miR-184, a brain-specific microRNA repressed by the binding of MeCP2 to its promoter, is upregulated by the release of MeCP2 after depolarization. The restricted release of MeCP2 from the paternal allele results in paternal allele-specific expression of miR-184. Our finding provides a clue to the link between the microRNA and DNA methylation pathways. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18058824  Title: The MeCP2-null mouse hippocampus displays altered basal inhibitory rhythms and is prone to hyperexcitability.  Rett syndrome is an autism-spectrum disorder caused by loss of function mutations within the gene encoding methyl CpG-binding protein 2 (MeCP2). While subtle decreases in synaptic plasticity have been detected within cortical and hippocampal neurons of Mecp2-null mice, only minimal information exists regarding how the loss of MeCP2 affects network activity in the brain. To address this issue, we compared the intrinsic network activities of Mecp2-null hippocampal slices derived from symptomatic mice to wild-type slices. Extracellular and whole-cell patch recordings revealed that although spontaneous, IPSP-based rhythmic activity is present in Mecp2-null slices; its frequency is significantly reduced from wild-type. This reduction was not associated with alterations in the gross electrophysiological properties of hippocampal neurons, but was associated with a decreased level of spontaneous glutamate receptor-mediated synaptic currents in hippocampal CA3 neurons. Paradoxically, however, repetitive sharp wave-like discharges were readily induced in the Mecp2-null hippocampal slices by a brief train of high-frequency stimulation commonly used to establish long-term potentiation at wild-type slices. Taken together, our data indicate that the Mecp2-null hippocampal CA3 circuit has diminished basal inhibitory rhythmic activity, which in turn renders the circuitry prone to hyperexcitability. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17980307  Title: The overlapping spectrum of rett and angelman syndromes: a clinical review.  Rett and Angelman syndromes comprise part of the spectrum of neurologic disorders associated with autism. Their clinical presentations overlap, with both presenting in later infancy with global developmental delays, severe speech and communication impairments, progressive microcephaly, seizures, autistic behaviors, and characteristic albeit different movement disorders and stereotypic hand movements. Although other features can help differentiate these disorders, significant phenotypic overlap and variation in severity sometimes cloud the underlying diagnosis. Rett syndrome is caused by a mutation in the MECP2 gene located on Xq28, whereas Angelman syndrome results from the loss of UBE3A function on chromosomal region 15q11-q13 related to a variety of molecular genetic mechanisms. Recent advances have uncovered interactions between these and other genes that affect the function and structure of neurons in the brain. The reversal of symptoms of Rett syndrome in a mature mouse model suggests the possibility for treatment of these and perhaps other autism-related disorders in the future. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17965627  Title: Epigenetics and Neural developmental disorders: Washington DC, September 18 and 19, 2006.  Neural developmental disorders, such as autism, Rett Syndrome, Fragile X syndrome, and Angelman syndrome manifest during early postnatal neural development. Although the genes responsible for some of these disorders have been identified, how the mutations of these genes affect neural development is currently unclear. Emerging evidence suggest that these disorders share common underlying defects in neuronal morphology, synaptic connectivity and brain plasticity. In particular, alterations in dendritic branching and spine morphology play a central role in the pathophysiology of most mental retardation disorders, suggesting that common pathways regulating neuronal function may be affected. Epigenetic modulations, mediated by DNA methylation, RNA-associated silencing, and histone modification, can serve as an intermediate process that imprints dynamic environmental experiences on the "fixed" genome, resulting in stable alterations in phenotypes. Disturbance in epigenetic regulations can lead to inappropriate expression or silencing of genes, causing an array of multi-system disorders and neoplasias. Rett syndrome, the most common form of mental retardation in young girls, is due to l mutation of MECP2, encoding a methylated DNA binding protein that translates DNA methylation into gene repression. Angelman syndrome is due to faulty genomic imprinting or maternal mutations in UBE3A. Fragile X Syndrome, in most cases, results from the hypermethylation of FMR1 promoter, hence the loss of expression of functional FMRP protein. Autism, with its complex etiology, may have strong epigenetic link. Together, these observations strongly suggest that epigenetic mechanisms may play a critical role in brain development and etiology of related disorders. This report summarizes the scientific discussions and major conclusions from a recent conference that aimed to gain insight into the common molecular pathways affected among these disorders and discover potential therapeutic targets that have been missed by looking at one disorder at a time. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17965589  Title: Sex difference in mecp2 expression during a critical period of rat brain development.  Pervasive developmental disorder is a classification covering five related conditions including the neurodevelopmental disorder Rett syndrome (RTT) and autism. Of these five conditions, only RTT has a known genetic cause with mutations in Methyl-CpG-binding protein 2 (MeCP2), a global repressor of gene expression, responsible for the majority of RTT cases. However, recent evidence indicates that reduced MeCP2 expression or activity is also found in autism and other disorders with overlapping phenotypes. Considering the sex difference in autism diagnosis, with males diagnosed four times more often than females, we questioned if a sex difference existed in the expression of MeCP2, in particular within the amygdala, a region that develops atypically in autism. We found that male rats express significantly less mecp2 mRNA and protein than females within the amygdala, as well as the ventromedial hypothalamus (VMH), but not within the preoptic area (POA) on post-natal day 1 (PN1). At PN10 these differences were gone; however, on this day males had more mecp2 mRNA than females within the POA. The transient sex difference of mecp2 expression during the steroid-sensitive period of brain development suggests that mecp2 may participate in normal sexual differentiation of the rat brain. Considering the strong link between MeCP2 and neurodevelopmental disorders, the lower levels of mecp2 expression in males may also underlie a biological risk for mecp2-related neural disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17562589  Title: Rett syndrome.  Rett syndrome (RS) is an X-linked neurodevelopmental disorder and the second most common cause of genetic mental retardation in females. Different mutations in MECP2 are found in up to 95% of typical cases of RS. This mainly neuronal expressed gene functions as a major transcription repressor. Extensive studies on girls who have RS and mouse models are aimed at finding main gene targets for MeCP2 protein and defining neuropathologic changes caused by its defects. Studies comparing autistic features in RS with idiopathic autism and mentally retarded patients are presented. Decreased dendritic arborization is common to RS and autism, leading to further research on similarities in pathogenesis, including MeCP2 protein levels in autistic brains and MeCP2 effects on genes connected to autism, like DLX5 and genes on 15q11-13 region. This area also is involved in Angelman syndrome, which has many similarities to RS. Despite these connections, MECP2 mutations in nonspecific autistic and mentally retarded populations are rare. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17486179  Title: Reduced MeCP2 expression is frequent in autism frontal cortex and correlates with aberrant MECP2 promoter methylation.  Mutations in MECP2, encoding methyl CpG binding protein 2 (MeCP2), cause most cases of Rett syndrome (RTT), an X-linked neurodevelopmental disorder. Both RTT and autism are "pervasive developmental disorders" and share a loss of social, cognitive and language skills and a gain in repetitive stereotyped behavior, following apparently normal perinatal development. Although MECP2 coding mutations are a rare cause of autism, MeCP2 expression defects were previously found in autism brain. To further study the role of MeCP2 in autism spectrum disorders (ASDs), we determined the frequency of MeCP2 expression defects in brain samples from autism and other ASDs. We also tested the hypotheses that MECP2 promoter mutations or aberrant promoter methylation correlate with reduced expression in cases of idiopathic autism. MeCP2 immunofluorescence in autism and other neurodevelopmental disorders was quantified by laser scanning cytometry and compared with control postmortem cerebral cortex samples on a large tissue microarray. A significant reduction in MeCP2 expression compared to age-matched controls was found in 11/14 autism (79%), 9/9 RTT (100%), 4/4 Angelman syndrome (100%), 3/4 Prader-Willi syndrome (75%), 3/5 Down syndrome (60%), and 2/2 attention deficit hyperactivity disorder (100%) frontal cortex samples. One autism female was heterozygous for a rare MECP2 promoter variant that correlated with reduced MeCP2 expression. A more frequent occurrence was significantly increased MECP2 promoter methylation in autism male frontal cortex compared to controls. Furthermore, percent promoter methylation of MECP2 significantly correlated with reduced MeCP2 protein expression. These results suggest that both genetic and epigenetic defects lead to reduced MeCP2 expression and may be important in the complex etiology of autism. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17464364  Title: Dynamic changes in Histone H3 lysine 9 acetylation localization patterns during neuronal maturation require MeCP2.  Mutations within the gene encoding methyl CpG binding protein 2 (MECP2) cause the autism-spectrum neurodevelopmental disorder Rett Syndrome (RTT). MECP2 recruits histone deacetylase to methylated DNA and acts as a long-range regulator of methylated genes. Despite ubiquitous MECP2 expression, the phenotype of RTT and the Mecp2-deficient mouse is largely restricted to the postnatal brain. Since Mecp2-deficient mice have a defect in neuronal maturation, we sought to understand how MECP2/Mecp2 mutations globally affect histone modifications during postnatal brain development by an immunofluorescence approach. Using an antibody specific to acetylated histone H3 lysine 9 (H3K9ac), a bright punctate nuclear staining pattern was observed as MECP2 expression increased in early postnatal neuronal nuclei. As neurons matured in juvenile and adult brain samples, the intensity of H3K9ac staining was reduced. Mecp2-deficient mouse and RTT cerebral neurons lacked this developmental reduction in H3K9ac staining compared to age-matched controls, resulting in a significant increase in neuronal nuclei with bright H3K9ac punctate staining. In contrast, trimethylated histone H3 lysine 9 (H3K9me3) localized to heterochromatin independent of MeCP2, but showed significantly reduced levels in Mecp2 deficient mouse and RTT brain. Autism brain with reduced MECP2 expression displayed similar histone H3 alterations as RTT brain. These observations suggest that MeCP2 regulates global histone modifications during a critical postnatal stage of neuronal maturation. These results have implications for understanding the molecular pathogenesis of RTT and autism in which MECP2 mutation or deficiency corresponds with arrested neurodevelopment. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17339270  Title: 15q11-13 GABAA receptor genes are normally biallelically expressed in brain yet are subject to epigenetic dysregulation in autism-spectrum disorders.  Human chromosome 15q11-13 is a complex locus containing imprinted genes as well as a cluster of three GABA(A) receptor subunit (GABR) genes-GABRB3, GABRA5 and GABRG3. Deletion or duplication of 15q11-13 GABR genes occurs in multiple human neurodevelopmental disorders including Prader-Willi syndrome (PWS), Angelman syndrome (AS) and autism. GABRB3 protein expression is also reduced in Rett syndrome (RTT), caused by mutations in MECP2 on Xq28. Although Gabrb3 is biallelically expressed in mouse brain, conflicting data exist regarding the imprinting status of the 15q11-13 GABR genes in humans. Using coding single nucleotide polymorphisms we show that all three GABR genes are biallelically expressed in 21 control brain samples, demonstrating that these genes are not imprinted in normal human cortex. Interestingly, four of eight autism and one of five RTT brain samples showed monoallelic or highly skewed allelic expression of one or more GABR gene, suggesting that epigenetic dysregulation of these genes is common to both disorders. Quantitative real-time RT-PCR analysis of PWS and AS samples with paternal and maternal 15q11-13 deletions revealed a paternal expression bias of GABRB3, while RTT brain samples showed a significant reduction in GABRB3 and UBE3A. Chromatin immunoprecipitation and bisulfite sequencing in SH-SY5Y neuroblastoma cells demonstrated that MeCP2 binds to methylated CpG sites within GABRB3. Our previous studies demonstrated that homologous 15q11-13 pairing in neurons was dependent on MeCP2 and was disrupted in RTT and autism cortex. Combined, these results suggest that MeCP2 acts as a chromatin organizer for optimal expression of both alleles of GABRB3 in neurons. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/17289941  Title: Reversal of neurological defects in a mouse model of Rett syndrome.  Rett syndrome is an autism spectrum disorder caused by mosaic expression of mutant copies of the X-linked MECP2 gene in neurons. However, neurons do not die, which suggests that this is not a neurodegenerative disorder. An important question for future therapeutic approaches to this and related disorders concerns phenotypic reversibility. Can viable but defective neurons be repaired, or is the damage done during development without normal MeCP2 irrevocable? Using a mouse model, we demonstrate robust phenotypic reversal, as activation of MeCP2 expression leads to striking loss of advanced neurological symptoms in both immature and mature adult animals. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/16647848  Title: MeCP2 dysfunction in Rett syndrome and related disorders.  Rett syndrome, a neurodevelopmental disorder caused by mutations in the X-linked gene encoding methyl-CpG-binding protein 2 (MeCP2), is a leading cause of mental retardation with autistic features in females. MECP2 mutations have also been identified in individuals with a variety of clinical syndromes, including mild learning-disability in females, neonatal encephalopathy in males, and psychiatric disorders, autism and X-linked mental retardation in both males and females. Furthermore, MECP2 duplications have been shown to cause a progressive postnatal neurological disorder. MeCP2 is a transcriptional repressor that binds to methylated CpG dinucleotides flanked by AT-rich segments and recruits a co-repressor complex, thereby altering chromatin structure. Subtle gene expression changes have been identified in Rett patients and mouse models; however, MeCP2 dysfunction has also been shown to cause abnormalities of RNA splicing, suggesting a complex molecular pathogenesis. Discovering which genes are misregulated in the absence of functional MeCP2 and demonstrating their role in causing neuronal dysfunction and disease manifestations are challenging but important steps for understanding the pathogenesis of Rett syndrome and related disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/16613900  Title: Identification of cis-regulatory elements for MECP2 expression.  Rett syndrome (RTT) is an X-linked dominant disabling neurodevelopmental disorder caused by loss of function mutations in the MECP2 gene, located at Xq28, which encodes a multifunctional protein. MECP2 expression is regulated in a developmental stage and cell-type-specific manner. The need for tightly controlled MeCP2 levels in brain is strongly suggested by neurologically abnormal phenotypes of mouse models with mild overexpression and by mental retardation in human males with MECP2 duplication. We set out to identify long-range cis-regulatory sequences that differentially regulate MECP2 transcription and, when mutated, may contribute to the pathogenesis of RTT, autism or X-linked mental retardation. By inter-species sequence comparisons, we detected 27 highly conserved non-coding DNA sequences within a 210 kb region covering MECP2. We functionally confirmed four enhancer and two silencer elements by performing luciferase reporter assays in four different human cell lines. The transcription factor binding capability of the identified regulatory elements was tested by gel shift assays. To locate the human MECP2 core promoter, we dissected the promoter region by reporter assays with deletion constructs. We then used chromosome conformation capture methods to document long-range interactions of three enhancers and two silencers with the MECP2 promoter. Acting over distances of up to 130 kb, these elements may influence chromatin configurations and regulate MECP2 transcription. Our study has defined the "MECP2 functional expression module" and identified enhancer and silencer elements that are likely to be responsible for the tissue-specific, developmental stage-specific or splice-variant-specific control of MeCP2 protein expression. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/16399702  Title: Learning and memory and synaptic plasticity are impaired in a mouse model of Rett syndrome.  Loss-of-function mutations or abnormal expression of the X-linked gene encoding methyl CpG binding protein 2 (MeCP2) cause a spectrum of postnatal neurodevelopmental disorders including Rett syndrome (RTT), nonsyndromic mental retardation, learning disability, and autism. Mice expressing a truncated allele of Mecp2 (Mecp2(308)) reproduce the motor and social behavior abnormalities of RTT; however, it is not known whether learning deficits are present in these animals. We investigated learning and memory, neuronal morphology, and synaptic function in Mecp2(308) mice. Hippocampus-dependent spatial memory, contextual fear memory, and social memory were significantly impaired in Mecp2(308) mutant males (Mecp2(308/Y)). The morphology of dendritic arborizations, the biochemical composition of synaptosomes and postsynaptic densities, and brain-derived neurotrophic factor expression were not altered in these mice. However, reduced postsynaptic density cross-sectional length was identified in asymmetric synapses of area CA1 of the hippocampus. In the hippocampus of symptomatic Mecp2(308/Y) mice, Schaffer-collateral synapses exhibited enhanced basal synaptic transmission and decreased paired-pulse facilitation, suggesting that neurotransmitter release was enhanced. Schaffer-collateral long-term potentiation (LTP) was impaired. LTP was also reduced in the motor and sensory regions of the neocortex. Finally, very early symptomatic Mecp2(308/Y) mice had increased basal synaptic transmission and deficits in the induction of long-term depression. These data demonstrate a requirement for MeCP2 in learning and memory and suggest that functional and ultrastructural synaptic dysfunction is an early event in the pathogenesis of RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/16251272  Title: Regulation of RNA splicing by the methylation-dependent transcriptional repressor methyl-CpG binding protein 2.  Rett syndrome (RTT) is a postnatal neurodevelopmental disorder characterized by the loss of acquired motor and language skills, autistic features, and unusual stereotyped movements. RTT is caused by mutations in the X-linked gene encoding methyl-CpG binding protein 2 (MeCP2). Mutations in MECP2 cause a variety of neurodevelopmental disorders including X-linked mental retardation, psychiatric disorders, and some cases of autism. Although MeCP2 was identified as a methylation-dependent transcriptional repressor, transcriptional profiling of RNAs from mice lacking MeCP2 did not reveal significant gene expression changes, suggesting that MeCP2 does not simply function as a global repressor. Changes in expression of a few genes have been observed, but these alterations do not explain the full spectrum of Rett-like phenotypes, raising the possibility that additional MeCP2 functions play a role in pathogenesis. In this study, we show that MeCP2 interacts with the RNA-binding protein Y box-binding protein 1 and regulates splicing of reporter minigenes. Importantly, we found aberrant alternative splicing patterns in a mouse model of RTT. Thus, we uncovered a previously uncharacterized function of MeCP2 that involves regulation of splicing, in addition to its role as a transcriptional repressor. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/15689352  Title: Homologous pairing of 15q11-13 imprinted domains in brain is developmentally regulated but deficient in Rett and autism samples.  Rett syndrome (RTT), caused by mutations in MECP2 (encoding methyl CpG binding protein 2), and Angelman syndrome (AS), caused by maternal deficiency of chromosome 15q11-13, are autism-spectrum neurodevelopmental disorders. MeCP2 is a transcriptional repressor of methylated genes, but MECP2 mutation does not directly affect the imprinted expression of genes within 15q11-13. We tested a potential role for MeCP2 in the homologous pairing of imprinted 15q11-13 alleles in human brain tissue and differentiated neurons by fluorescence in situ hybridization (FISH). FISH analysis of control cerebral samples demonstrated a significant increase in homologous pairing specific to chromosome 15 from infant to juvenile brain samples. Significant and specific deficiencies in the percentage of paired chromosome 15 alleles were observed in RTT, AS and autism brain samples when compared with normal controls. SH-SY5Y neuroblastoma cells also showed a significant and specific increase in the percentage of chromosome 15q11-13 paired alleles following induced differentiation in vitro. Transfection with a methylated oligonucleotide decoy specifically blocked binding of MeCP2 to the SNURF/SNRPN promoter within 15q11-13 and significantly lowered the percentage of paired 15q11-13 alleles in SH-SY5Y cells. These combined results suggest a role for MeCP2 in chromosome organization in the developing brain and provide a potential mechanistic association between several related neurodevelopmental disorders. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/15615769  Title: Epigenetic overlap in autism-spectrum neurodevelopmental disorders: MECP2 deficiency causes reduced expression of UBE3A and GABRB3.  Autism is a common neurodevelopmental disorder of complex genetic etiology. Rett syndrome, an X-linked dominant disorder caused by MECP2 mutations, and Angelman syndrome, an imprinted disorder caused by maternal 15q11-q13 or UBE3A deficiency, have phenotypic and genetic overlap with autism. MECP2 encodes methyl-CpG-binding protein 2 that acts as a transcriptional repressor for methylated gene constructs but is surprisingly not required for maintaining imprinted gene expression. Here, we test the hypothesis that MECP2 deficiency may affect the level of expression of UBE3A and neighboring autism candidate gene GABRB3 without necessarily affecting imprinted expression. Multiple quantitative methods were used including automated quantitation of immunofluorescence and in situ hybridization by laser scanning cytometry on tissue microarrays, immunoblot and TaqMan PCR. The results demonstrated significant defects in UBE3A/E6AP expression in two different Mecp2 deficient mouse strains and human Rett, Angelman and autism brains compared with controls. Although no difference was observed in the allelic expression of several imprinted transcripts in Mecp2-null brain, Ube3a sense expression was significantly reduced, consistent with the decrease in protein. A non-imprinted gene from 15q11-q13, GABRB3, encoding the beta3 subunit of the GABAA receptor, also showed significantly reduced expression in multiple Rett, Angelman and autism brain samples, and Mecp2 deficient mice by quantitative immunoblot. These results suggest an overlapping pathway of gene dysregulation within 15q11-q13 in Rett, Angelman and autism and implicate MeCP2 in the regulation of UBE3A and GABRB3 expressions in the postnatal mammalian brain. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/15362175  Title: Rett syndrome: of girls and mice--lessons for regression in autism.  Rett syndrome (RTT) is a neurodevelopmental disorder occurring almost exclusively in females. Regression is a defining feature of RTT. During the regression stage, RTT girls display many autistic features, such as loss of communication and social skills, poor eye contact, and lack of interest, and initially may be given the diagnosis of autism. The discovery of the genetic cause of RTT, mutations in the MECP2 gene, a transcriptional repressor, has promoted the early diagnosis of RTT and development of mouse models. The phenotype of one mouse model includes features such as regression and abnormal behavioral and social interactions. The timing of the period of regression in RTT--during ages 1 to 2 years--parallels the period of intense synaptic development. The effects of the MECP2 mutation also increases concomitantly with peak synaptogenesis. Neuropathological findings in Rett include the selective reduction of dendritric spines in the pyramidal cells of RTT brains; this feature has also been reported in autism. Studies have observed that MECP influences the expression of brain-derived neurotrophic factor and thus may influence synaptic plasticity. Abnormalities in synapse maintenance and modulation may contribute to regression in RTT and autism. Studies of the clinical aspects of the regression period and of the mouse model may be useful in understanding the pathophysiology of RTT and other neurodevelopmental disorders such as autism. A recent study observed abnormal expression of MeCP2 in RTT and other neurodevelopmental disorders such as autism. Although the genetic background and certain clinical features differ in RTT and autism, a similar mechanism involving MeCP2 regulation and expression may contribute to regression. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/14734626  Title: Multiple pathways regulate MeCP2 expression in normal brain development and exhibit defects in autism-spectrum disorders.  Rett syndrome (RTT) is a neurodevelopmental disorder caused by mutations in MECP2, encoding methyl-CpG-binding protein 2 (MeCP2). Although MECP2 is ubiquitously transcribed, MeCP2 expression is developmentally regulated and heterogeneous in neuronal subpopulations, defined as MeCP2(lo) and MeCP2(hi). To test the hypothesis that pathways affecting MeCP2 expression changes may be defective in RTT, autism and other neurodevelopmental disorders without MECP2 mutations, a high-throughput quantitation of MeCP2 expression was performed on a tissue microarray containing frontal cortex samples from 28 different patients with neurodevelopmental disorders and age-matched controls. Combined quantitative analyses of MeCP2 protein and alternatively polyadenylated transcript levels were performed by laser scanning cytometry and tested for significant differences from age-matched controls. Normal cerebral samples showed an increase in total MeCP2 expression and the percentage of MeCP2(hi) cells with age that could be explained by increased MECP2 transcription within the MeCP2(hi) population. A significant decrease in the relative usage of the long transcript in the MeCP2(lo) population was observed in postnatal compared to fetal brain, but alternate polyadenylation did not correlate with MeCP2 expression changes at the single cell level. Brain samples from several related neurodevelopmental disorders, including autism, pervasive developmental disorder, Prader-Willi and Angelman syndromes showed significant differences in MeCP2 expression from age-matched controls by apparently different transcriptional and post-transcriptional mechanisms. These results suggest that multiple pathways regulate the complex developmental expression of MeCP2 and are defective in autism-spectrum disorders in addition to RTT. |
| MECP2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/12027529  Title: Minor form of trigonocephaly is an autistic skull shape? A suggestion based on homeobox gene variants and MECP2 mutations.  A possible role for Hoxa1 genotype in susceptibility to autism spectrum disorders was recently proposed. Furthermore, it has been demonstrated that Rett syndrome, which is categorized into pervasive developmental disorders the same as the autism spectrum disorders are, is associated with mutations in MECP2 gene. These findings suggest that the genetic backgrounds of these behavioral conditions may involve genes which also have an important role in the development of skull, because Hoxa1 is a key gene for skull development as well as for brain development and one of the clinical characteristics of Rett syndrome is deceleration in head growth. Together with this evolving knowledge, a series of ethical arguments concerning the indication of surgical treatment in patients with minor forms of trigonocephaly with autistic behaviors and/or hyperactivity leads us to hypothesize the presence of an autism subtype which may frequently be accompanied by specific morphological skull characteristics (autistic skull shape). |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/36180228  Title: Peripheral Auditory Nerve Impairment in a Mouse Model of Syndromic Autism.  Dysfunction of the peripheral auditory nerve (AN) contributes to dynamic changes throughout the central auditory system, resulting in abnormal auditory processing, including hypersensitivity. Altered sound sensitivity is frequently observed in autism spectrum disorder (ASD), suggesting that AN deficits and changes in auditory information processing may contribute to ASD-associated symptoms, including social communication deficits and hyperacusis. The MEF2C transcription factor is associated with risk for several neurodevelopmental disorders, and mutations or deletions of MEF2C produce a haploinsufficiency syndrome characterized by ASD, language, and cognitive deficits. A mouse model of this syndromic ASD (Mef2c-Het) recapitulates many of the MEF2C haploinsufficiency syndrome-linked behaviors, including communication deficits. We show here that Mef2c-Het mice of both sexes exhibit functional impairment of the peripheral AN and a modest reduction in hearing sensitivity. We find that MEF2C is expressed during development in multiple AN and cochlear cell types; and in Mef2c-Het mice, we observe multiple cellular and molecular alterations associated with the AN, including abnormal myelination, neuronal degeneration, neuronal mitochondria dysfunction, and increased macrophage activation and cochlear inflammation. These results reveal the importance of MEF2C function in inner ear development and function and the engagement of immune cells and other non-neuronal cells, which suggests that microglia/macrophages and other non-neuronal cells might contribute, directly or indirectly, to AN dysfunction and ASD-related phenotypes. Finally, our study establishes a comprehensive approach for characterizing AN function at the physiological, cellular, and molecular levels in mice, which can be applied to animal models with a wide range of human auditory processing impairments.SIGNIFICANCE STATEMENT This is the first report of peripheral auditory nerve (AN) impairment in a mouse model of human MEF2C haploinsufficiency syndrome that has well-characterized ASD-related behaviors, including communication deficits, hyperactivity, repetitive behavior, and social deficits. We identify multiple underlying cellular, subcellular, and molecular abnormalities that may contribute to peripheral AN impairment. Our findings also highlight the important roles of immune cells (e.g., cochlear macrophages) and other non-neuronal elements (e.g., glial cells and cells in the stria vascularis) in auditory impairment in ASD. The methodological significance of the study is the establishment of a comprehensive approach for evaluating peripheral AN function and impact of peripheral AN deficits with minimal hearing loss. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/34991657  Title: Progress on the roles of MEF2C in neuropsychiatric diseases.  Myocyte Enhancer Factor 2 C (MEF2C), one of the transcription factors of the MADS-BOX family, is involved in embryonic brain development, neuronal formation and differentiation, as well as in the growth and pruning of axons and dendrites. MEF2C is also involved in the development of various neuropsychiatric disorders, such as autism spectrum disorders (ASD), epilepsy, schizophrenia and Alzheimer's disease (AD). Here, we review the relationship between MEF2C and neuropsychiatric disorders, and provide further insights into the mechanism of these diseases. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/33972691  Title: Cadherin-13 is a critical regulator of GABAergic modulation in human stem-cell-derived neuronal networks.  Activity in the healthy brain relies on a concerted interplay of excitation (E) and inhibition (I) via balanced synaptic communication between glutamatergic and GABAergic neurons. A growing number of studies imply that disruption of this E/I balance is a commonality in many brain disorders; however, obtaining mechanistic insight into these disruptions, with translational value for the patient, has typically been hampered by methodological limitations. Cadherin-13 (CDH13) has been associated with autism and attention-deficit/hyperactivity disorder. CDH13 localizes at inhibitory presynapses, specifically of parvalbumin (PV) and somatostatin (SST) expressing GABAergic neurons. However, the mechanism by which CDH13 regulates the function of inhibitory synapses in human neurons remains unknown. Starting from human-induced pluripotent stem cells, we established a robust method to generate a homogenous population of SST and MEF2C (PV-precursor marker protein) expressing GABAergic neurons (iGABA) in vitro, and co-cultured these with glutamatergic neurons at defined E/I ratios on micro-electrode arrays. We identified functional network parameters that are most reliably affected by GABAergic modulation as such, and through alterations of E/I balance by reduced expression of CDH13 in iGABAs. We found that CDH13 deficiency in iGABAs decreased E/I balance by means of increased inhibition. Moreover, CDH13 interacts with Integrin-β1 and Integrin-β3, which play opposite roles in the regulation of inhibitory synaptic strength via this interaction. Taken together, this model allows for standardized investigation of the E/I balance in a human neuronal background and can be deployed to dissect the cell-type-specific contribution of disease genes to the E/I balance. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/32975584  Title: Genes influenced by MEF2C contribute to neurodevelopmental disease via gene expression changes that affect multiple types of cortical excitatory neurons.  Myocyte enhancer factor 2 C (MEF2C) is an important transcription factor during neurodevelopment. Mutation or deletion of MEF2C causes intellectual disability (ID), and common variants within MEF2C are associated with cognitive function and schizophrenia risk. We investigated if genes influenced by MEF2C during neurodevelopment are enriched for genes associated with neurodevelopmental phenotypes and if this can be leveraged to identify biological mechanisms and individual brain cell types affected. We used a set of 1055 genes that were differentially expressed in the adult mouse brain following early embryonic deletion of Mef2c in excitatory cortical neurons. Using genome-wide association studies data, we found these differentially expressed genes (DEGs) to be enriched for genes associated with schizophrenia, intelligence and educational attainment but not autism spectrum disorder (ASD). For this gene set, genes that overlap with target genes of the Fragile X mental retardation protein (FMRP) are a major driver of these enrichments. Using trios data, we found these DEGs to be enriched for genes containing de novo mutations reported in ASD and ID, but not schizophrenia. Using single-cell RNA sequencing data, we identified that a number of different excitatory glutamatergic neurons in the cortex were enriched for these DEGs including deep layer pyramidal cells and cells in the retrosplenial cortex, entorhinal cortex and subiculum, and these cell types are also enriched for FMRP target genes. The involvement of MEF2C and FMRP in synapse elimination suggests that disruption of this process in these cell types during neurodevelopment contributes to cognitive function and risk of neurodevelopmental disorders. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/32418612  Title: MEF2C Hypofunction in Neuronal and Neuroimmune Populations Produces MEF2C Haploinsufficiency Syndrome-like Behaviors in Mice.  Microdeletions of the MEF2C gene are linked to a syndromic form of autism termed MEF2C haploinsufficiency syndrome (MCHS). MEF2C hypofunction in neurons is presumed to underlie most of the symptoms of MCHS. However, it is unclear in which cell populations MEF2C functions to regulate neurotypical development. Multiple biochemical, molecular, electrophysiological, behavioral, and transgenic mouse approaches were used to characterize MCHS-relevant synaptic, behavioral, and gene expression changes in mouse models of MCHS. We showed that MCHS-associated missense mutations cluster in the conserved DNA binding domain and disrupt MEF2C DNA binding. DNA binding-deficient global Mef2c heterozygous mice (Mef2c-Het) displayed numerous MCHS-related behaviors, including autism-related behaviors, changes in cortical gene expression, and deficits in cortical excitatory synaptic transmission. We detected hundreds of dysregulated genes in Mef2c-Het cortex, including significant enrichments of autism risk and excitatory neuron genes. In addition, we observed an enrichment of upregulated microglial genes, but this was not due to neuroinflammation in the Mef2c-Het cortex. Importantly, conditional Mef2c heterozygosity in forebrain excitatory neurons reproduced a subset of the Mef2c-Het phenotypes, while conditional Mef2c heterozygosity in microglia reproduced social deficits and repetitive behavior. Taken together, our findings show that mutations found in individuals with MCHS disrupt the DNA-binding function of MEF2C, and DNA binding-deficient Mef2c global heterozygous mice display numerous MCHS-related phenotypes, including excitatory neuron and microglia gene expression changes. Our findings suggest that MEF2C regulates typical brain development and function through multiple cell types, including excitatory neuronal and neuroimmune populations. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/29133852  Title: NitroSynapsin therapy for a mouse MEF2C haploinsufficiency model of human autism.  Transcription factor MEF2C regulates multiple genes linked to autism spectrum disorder (ASD), and human MEF2C haploinsufficiency results in ASD, intellectual disability, and epilepsy. However, molecular mechanisms underlying MEF2C haploinsufficiency syndrome remain poorly understood. Here we report that Mef2c +/-(Mef2c-het) mice exhibit behavioral deficits resembling those of human patients. Gene expression analyses on brains from these mice show changes in genes associated with neurogenesis, synapse formation, and neuronal cell death. Accordingly, Mef2c-het mice exhibit decreased neurogenesis, enhanced neuronal apoptosis, and an increased ratio of excitatory to inhibitory (E/I) neurotransmission. Importantly, neurobehavioral deficits, E/I imbalance, and histological damage are all ameliorated by treatment with NitroSynapsin, a new dual-action compound related to the FDA-approved drug memantine, representing an uncompetitive/fast off-rate antagonist of NMDA-type glutamate receptors. These results suggest that MEF2C haploinsufficiency leads to abnormal brain development, E/I imbalance, and neurobehavioral dysfunction, which may be mitigated by pharmacological intervention. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/28993242  Title: A distinct microRNA expression profile is associated with α[11C]-methyl-L-tryptophan (AMT) PET uptake in epileptogenic cortical tubers resected from patients with tuberous sclerosis complex.  Tuberous sclerosis complex (TSC) is characterized by hamartomatous lesions in various organs and arises due to mutations in the TSC1 or TSC2 genes. TSC mutations lead to a range of neurological manifestations including epilepsy, cognitive impairment, autism spectrum disorders (ASD), and brain lesions that include cortical tubers. There is evidence that seizures arise at or near cortical tubers, but it is unknown why some tubers are epileptogenic while others are not. We have previously reported increased tryptophan metabolism measured with α[11C]-methyl-l-tryptophan (AMT) positron emission tomography (PET) in epileptogenic tubers in approximately two-thirds of patients with tuberous sclerosis and intractable epilepsy. However, the underlying mechanisms leading to seizure onset in TSC remain poorly characterized. MicroRNAs are enriched in the brain and play important roles in neurodevelopment and brain function. Recent reports have shown aberrant microRNA expression in epilepsy and TSC. In this study, we performed microRNA expression profiling in brain specimens obtained from TSC patients undergoing epilepsy surgery for intractable epilepsy. Typically, in these resections several non-seizure onset tubers are resected together with the seizure-onset tubers because of their proximity. We directly compared seizure onset tubers, with and without increased tryptophan metabolism measured with PET, and non-onset tubers to assess the role of microRNAs in epileptogenesis associated with these lesions. Whether a particular tuber was epileptogenic or non-epileptogenic was determined with intracranial electrocorticography, and tryptophan metabolism was measured with AMT PET. We identified a set of five microRNAs (miR-142-3p, 142-5p, 223-3p, 200b-3p and 32-5p) that collectively distinguish among the three primary groups of tubers: non-onset/AMT-cold (NC), onset/AMT-cold (OC), and onset/AMT-hot (OH). These microRNAs were significantly upregulated in OH tubers compared to the other two groups, and microRNA expression was most significantly associated with AMT-PET uptake. The microRNAs target a group of genes enriched for synaptic signaling and epilepsy risk, including SLC12A5, SYT1, GRIN2A, GRIN2B, KCNB1, SCN2A, TSC1, and MEF2C. We confirmed the interaction between miR-32-5p and SLC12A5 using a luciferase reporter assay. Our findings provide a new avenue for subsequent mechanistic studies of tuber epileptogenesis in TSC. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/27779093  Title: MEF2C regulates cortical inhibitory and excitatory synapses and behaviors relevant to neurodevelopmental disorders.  Numerous genetic variants associated with MEF2C are linked to autism, intellectual disability (ID) and schizophrenia (SCZ) - a heterogeneous collection of neurodevelopmental disorders with unclear pathophysiology. MEF2C is highly expressed in developing cortical excitatory neurons, but its role in their development remains unclear. We show here that conditional embryonic deletion of Mef2c in cortical and hippocampal excitatory neurons (Emx1-lineage) produces a dramatic reduction in cortical network activity in vivo, due in part to a dramatic increase in inhibitory and a decrease in excitatory synaptic transmission. In addition, we find that MEF2C regulates E/I synapse density predominantly as a cell-autonomous, transcriptional repressor. Analysis of differential gene expression in Mef2c mutant cortex identified a significant overlap with numerous synapse- and autism-linked genes, and the Mef2c mutant mice displayed numerous behaviors reminiscent of autism, ID and SCZ, suggesting that perturbing MEF2C function in neocortex can produce autistic- and ID-like behaviors in mice. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/26642739  Title: Postnatal Loss of Mef2c Results in Dissociation of Effects on Synapse Number and Learning and Memory.  Myocyte enhancer factor 2 (MEF2) transcription factors play critical roles in diverse cellular processes during central nervous system development. Studies attempting to address the role of MEF2 in brain have largely relied on overexpression of a constitutive MEF2 construct that impairs memory formation or knockdown of MEF2 function that increases spine numbers and enhances memory formation. Genetic deletion of individual MEF2 isoforms in brain during embryogenesis demonstrated that Mef2c loss negatively regulates spine numbers resulting in learning and memory deficits, possibly as a result of its essential role in development. To investigate MEF2C function in brain further, we genetically deleted Mef2c during postnatal development in mice. We characterized these conditional Mef2c knockout mice in an array of behavioral paradigms and examined the impact of postnatal loss of Mef2c on long-term potentiation. We observed increased spine numbers in hippocampus of the conditional Mef2c knockout mice. However, the postnatal loss of Mef2c did not impact learning and memory, long-term potentiation, or social and repetitive behaviors. Our findings demonstrate a critical role for MEF2C in the regulation of spine numbers with a dissociation of learning and memory, synaptic plasticity, and measures of autism-related behaviors in postnatal brain. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/20412115  Title: Refining the phenotype associated with MEF2C haploinsufficiency.  Recently, submicroscopic deletions of the 5q14.3 region have been described in patients with severe mental retardation (MR), stereotypic movements, epilepsy and cerebral malformations. Further delineation of a critical region of overlap in these patients pointed to MEF2C as the responsible gene. This finding was further reinforced by the identification of a nonsense mutation in a patient with a similar phenotype. In brain, MEF2C is essential for early neurogenesis, neuronal migration and differentiation. Here we present two additional patients with severe MR, autism spectrum disorder and epilepsy, carrying a very small deletion encompassing the MEF2C gene. This finding strengthens the role of this gene in severe MR, and enables further delineation of the clinical phenotype. |
| MEF2C |
| Url: https://pubmed.ncbi.nlm.nih.gov/18599437  Title: Transcription factor MEF2C influences neural stem/progenitor cell differentiation and maturation in vivo.  Emerging evidence suggests that myocyte enhancer factor 2 (MEF2) transcription factors act as effectors of neurogenesis in the brain, with MEF2C the predominant isoform in developing cerebrocortex. Here, we show that conditional knockout of Mef2c in nestin-expressing neural stem/progenitor cells (NSCs) impaired neuronal differentiation in vivo, resulting in aberrant compaction and smaller somal size. NSC proliferation and survival were not affected. Conditional null mice surviving to adulthood manifested more immature electrophysiological network properties and severe behavioral deficits reminiscent of Rett syndrome, an autism-related disorder. Our data support a crucial role for MEF2C in programming early neuronal differentiation and proper distribution within the layers of the neocortex. |
| MEIS2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36418415  Title: The autism-associated Meis2 gene is necessary for cardiac baroreflex regulation in mice.  Recent understanding of Autism Spectrum Disorder (ASD) showed that peripheral primary mechanosensitive neurons involved in touch sensation and central neurons affected in ASD share transcriptional regulators. Mutant mice for ASD-associated transcription factors exhibit impaired primary tactile perception and restoring those genes specifically in primary sensory neurons rescues some of the anxiety-like behavior and social interaction defects. Interestingly, peripheral mechanosensitive sensory neurons also project to internal organs including the cardiovascular system, and an imbalance of the cardio-vascular sympathovagal regulation is evidenced in ASD and intellectual disability. ASD patients have decreased vagal tone, suggesting dysfunction of sensory neurons involved in cardio-vascular sensing. In light of our previous finding that the ASD-associated Meis2 gene is necessary for normal touch neuron development and function, we investigated here if its inactivation in mouse peripheral sensory neurons also affects cardio-vascular sympathovagal regulation and baroreflex. Combining echocardiography, pharmacological challenge, blood pressure monitoring, and heart rate variability analysis, we found that Meis2 mutant mice exhibited a blunted vagal response independently of any apparent cardiac malformation. These results suggest that defects in primary sensory neurons with mechanosensitive identity could participate in the imbalanced cardio-vascular sympathovagal tone found in ASD patients, reinforcing current hypotheses on the role of primary sensory neurons in the etiology of ASD. |
| MEIS2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32393163  Title: Transcriptome analysis of neural progenitor cells derived from Lowe syndrome induced pluripotent stem cells: identification of candidate genes for the neurodevelopmental and eye manifestations.  Lowe syndrome (LS) is caused by loss-of-function mutations in the X-linked gene OCRL, which codes for an inositol polyphosphate 5-phosphatase that plays a key role in endosome recycling, clathrin-coated pit formation, and actin polymerization. It is characterized by congenital cataracts, intellectual and developmental disability, and renal proximal tubular dysfunction. Patients are also at high risk for developing glaucoma and seizures. We recently developed induced pluripotent stem cell (iPSC) lines from three patients with LS who have hypomorphic variants affecting the 3' end of the gene, and their neurotypical brothers to serve as controls. In this study, we used RNA sequencing (RNA-seq) to obtain transcriptome profiles in LS and control neural progenitor cells (NPCs). In a comparison of the patient and control NPCs (n = 3), we found 16 differentially expressed genes (DEGs) at the multiple test adjusted p value (padj) < 0.1, with nine at padj < 0.05. Using nominal p value < 0.05, 319 DEGs were detected. The relatively small number of DEGs could be due to the fact that OCRL is not a transcription factor per se, although it could have secondary effects on gene expression through several different mechanisms. Although the number of DEGs passing multiple test correction was small, those that were found are quite consistent with some of the known molecular effects of OCRL protein, and the clinical manifestations of LS. Furthermore, using gene set enrichment analysis (GSEA), we found that genes increased expression in the patient NPCs showed enrichments of several gene ontology (GO) terms (false discovery rate < 0.25): telencephalon development, pallium development, NPC proliferation, and cortex development, which are consistent with a condition characterized by intellectual disabilities and psychiatric manifestations. In addition, a significant enrichment among the nominal DEGs for genes implicated in autism spectrum disorder (ASD) was found (e.g., AFF2, DNER, DPP6, DPP10, RELN, CACNA1C), as well as several that are strong candidate genes for the development of eye problems found in LS, including glaucoma. The most notable example is EFEMP1, a well-known candidate gene for glaucoma and other eye pathologies. Overall, the RNA-seq findings present several candidate genes that could help explain the underlying basis for the neurodevelopmental and eye problems seen in boys with LS. |
| MNT |
| Url: https://pubmed.ncbi.nlm.nih.gov/31088591  Title: Surface-based shared and distinct resting functional connectivity in attention-deficit hyperactivity disorder and autism spectrum disorder.  Both attention-deficit hyperactivity disorder (ADHD) and autism spectrum disorder (ASD) are neurodevelopmental disorders with a high prevalence. They are often comorbid and both exhibit abnormalities in sustained attention, yet common and distinct neural patterns of ASD and ADHD remain unidentified.AimsTo investigate shared and distinct functional connectivity patterns in a relatively large sample of boys (7- to 15-year-olds) with ADHD, ASD and typical development matched by age, gender and IQ. We applied machine learning techniques to investigate patterns of surface-based brain resting-state connectivity in 86 boys with ASD, 83 boys with ADHD and 125 boys with typical development. We observed increased functional connectivity within the limbic and somatomotor networks in boys with ASD compared with boys with typical development. We also observed increased functional connectivity within the limbic, visual, default mode, somatomotor, dorsal attention, frontoparietal and ventral attention networks in boys with ADHD compared with boys with ASD. In addition, using a machine learning approach, we were able to discriminate typical development from ASD, typical development from ADHD and ASD from ADHD with accuracy rates of 76.3%, 84.1%, and 79.3%, respectively. Our results may shed new light on the underlying mechanisms of ASD and ADHD and facilitate the development of new diagnostic methods for these disorders.Declaration of interestJ.K. holds equity in a startup company, MNT. |
| MNT |
| Url: https://pubmed.ncbi.nlm.nih.gov/29985556  Title: X-ray Structures and Feasibility Assessment of CLK2 Inhibitors for Phelan-McDermid Syndrome.  CLK2 inhibition has been proposed as a potential mechanism to improve autism and neuronal functions in Phelan-McDermid syndrome (PMDS). Herein, the discovery of a very potent indazole CLK inhibitor series and the CLK2 X-ray structure of the most potent analogue are reported. This new indazole series was identified through a biochemical CLK2 Caliper assay screen with 30k compounds selected by an in silico approach. Novel high-resolution X-ray structures of all CLKs, including the first CLK4 X-ray structure, bound to known CLK2 inhibitor tool compounds (e.g., TG003, CX-4945), are also shown and yield insight into inhibitor selectivity in the CLK family. The efficacy of the new CLK2 inhibitors from the indazole series was demonstrated in the mouse brain slice assay, and potential safety concerns were investigated. Genotoxicity findings in the human lymphocyte micronucleus test (MNT) assay are shown by using two structurally different CLK inhibitors to reveal a major concern for pan-CLK inhibition in PMDS. |
| MYT1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/35538503  Title: Myt1l haploinsufficiency leads to obesity and multifaceted behavioral alterations in mice.  The zinc finger domain containing transcription factor Myt1l is tightly associated with neuronal identity and is the only transcription factor known that is both neuron-specific and expressed in all neuronal subtypes. We identified Myt1l as a powerful reprogramming factor that, in combination with the proneural bHLH factor Ascl1, could induce neuronal fate in fibroblasts. Molecularly, we found it to repress many non-neuronal gene programs, explaining its supportive role to induce and safeguard neuronal identity in combination with proneural bHLH transcriptional activators. Moreover, human genetics studies found MYT1L mutations to cause intellectual disability and autism spectrum disorder often coupled with obesity. Here, we generated and characterized Myt1l-deficient mice. A comprehensive, longitudinal behavioral phenotyping approach was applied. Myt1l was necessary for survival beyond 24 h but not for overall histological brain organization. Myt1l heterozygous mice became increasingly overweight and exhibited multifaceted behavioral alterations. In mouse pups, Myt1l haploinsufficiency caused mild alterations in early socio-affective communication through ultrasonic vocalizations. In adulthood, Myt1l heterozygous mice displayed hyperactivity due to impaired habituation learning. Motor performance was reduced in Myt1l heterozygous mice despite intact motor learning, possibly due to muscular hypotonia. While anxiety-related behavior was reduced, acoustic startle reactivity was enhanced, in line with higher sensitivity to loud sound. Finally, Myt1l haploinsufficiency had a negative impact on contextual fear memory retrieval, while cued fear memory retrieval appeared to be intact. In future studies, additional phenotypes might be identified and a detailed characterization of direct reciprocal social interaction behavior might help to reveal effects of Myt1l haploinsufficiency on social behavior in juvenile and adult mice. Behavioral alterations in Myt1l haploinsufficient mice recapitulate several clinical phenotypes observed in humans carrying heterozygous MYT1L mutations and thus serve as an informative model of the human MYT1L syndrome. |
| MYT1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/34856129  Title: Unraveling the mysteries of MYT1L: From reprogramming factor to multifaceted regulator of neuronal differentiation.  In this issue of Neuron, Chen et al. (2021) generated a mouse model for haploinsufficiency of MYT1L. MYT1L is widely used in neuronal reprogramming, and de novo mutations have been linked to a neurodevelopmental syndrome. Extensive characterization in this study better delineates MYT1L's role in transcriptional regulation and neuronal differentiation. |
| MYT1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/34614421  Title: A MYT1L syndrome mouse model recapitulates patient phenotypes and reveals altered brain development due to disrupted neuronal maturation.  Human genetics have defined a new neurodevelopmental syndrome caused by loss-of-function mutations in MYT1L, a transcription factor known for enabling fibroblast-to-neuron conversions. However, how MYT1L mutation causes intellectual disability, autism, ADHD, obesity, and brain anomalies is unknown. Here, we developed a Myt1l haploinsufficient mouse model that develops obesity, white-matter thinning, and microcephaly, mimicking common clinical phenotypes. During brain development we discovered disrupted gene expression, mediated in part by loss of Myt1l gene-target activation, and identified precocious neuronal differentiation as the mechanism for microcephaly. In contrast, in adults we discovered that mutation results in failure of transcriptional and chromatin maturation, echoed in disruptions in baseline physiological properties of neurons. Myt1l haploinsufficiency also results in behavioral anomalies, including hyperactivity, muscle weakness, and social alterations, with more severe phenotypes in males. Overall, our findings provide insight into the mechanistic underpinnings of this disorder and enable future preclinical studies. |
| MYT1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/32267091  Title: MYT1L: A systematic review of genetic variation encompassing schizophrenia and autism.  Variations in MYT1L, a gene encoding a transcription factor expressed in the brain, have been associated with autism, intellectual disability, and schizophrenia. Here we provide an updated review of published reports of neuropsychiatric correlates of loss of function and duplication of MYT1L. Of 27 duplications all were partial; 33% were associated exclusively with schizophrenia, and the chromosomal locations of schizophrenia-associated duplications exhibited a distinct difference in pattern-of-location from those associated with autism and/or intellectual disability. Of 51 published heterozygous loss of function variants, all but one were associated with intellectual disability, autism, or both, and one resulted in no neuropsychiatric diagnosis. There were no reports of schizophrenia associated with loss of function variants of MYT1L (Fisher's exact p < .00001, for contrast with all reported duplications). Although the precise function of the various mutations remains unspecified, these data collectively establish the candidacy of MYT1L as a reciprocal mutation, in which schizophrenia may be engendered by partial duplications, typically involving the 3' end of the gene, while developmental disability-notably autism-is associated with both loss of function and partial duplication. Future research on the specific effects of contrasting mutations in MYT1L may provide insight into the causal origins of autism and schizophrenia. |
| MYT1L |
| Url: https://pubmed.ncbi.nlm.nih.gov/22157634  Title: Germline mosaic transmission of a novel duplication of PXDN and MYT1L to two male half-siblings with autism.  Autism is a neurodevelopmental disorder with a strong genetic component to susceptibility. In this study, we report the molecular characterization of an apparent de-novo 281 kb duplication of chromosome 2p25.3 in two male half-siblings with autism. The 2p25.3 duplication was first identified through a low-density microarray, validated with fluorescent in-situ hybridization, and duplication breakpoints were delineated using an Affymetrix 6.0 single-nucleotide polymorphism microarray. The fluorescent in-situ hybridization results validated the novel copy number variant and revealed the mother to be mosaic, with ∼33% of her lymphoblast cells carrying the duplication. Therefore, the duplication was transmitted through the mechanism of germline mosaicism. In addition, duplication breakpoints were refined and showed that PXDN is fully duplicated, whereas seven exons of the terminal portion of the 25 exon gene MYT1L are within the duplicated region. MYT1L, a gene predominately expressed in the brain, has recently been linked with other neuropsychiatric illness such as schizophrenia and depression. Results from this study indicate that the 2p25.3 duplication disrupting PXDN and MYT1L is a potential autism-causing variant in the pedigree reported here and should receive further consideration as a candidate for autism. |
| NFIA |
| Url: https://pubmed.ncbi.nlm.nih.gov/33431980  Title: Exploring the biological role of postzygotic and germinal de novo mutations in ASD.  De novo mutations (DNMs), including germinal and postzygotic mutations (PZMs), are a strong source of causality for Autism Spectrum Disorder (ASD). However, the biological processes involved behind them remain unexplored. Our aim was to detect DNMs (germinal and PZMs) in a Spanish ASD cohort (360 trios) and to explore their role across different biological hierarchies (gene, biological pathway, cell and brain areas) using bioinformatic approaches. For the majority of the analysis, a combined ASD cohort (N = 2171 trios) was created using previously published data by the Autism Sequencing Consortium (ASC). New plausible candidate genes for ASD such as FMR1 and NFIA were found. In addition, genes harboring PZMs were significantly enriched for miR-137 targets in comparison with germinal DNMs that were enriched in GO terms related to synaptic transmission. The expression pattern of genes with PZMs was restricted to early mid-fetal cortex. In contrast, the analysis of genes with germinal DNMs revealed a spatio-temporal window from early to mid-fetal development stages, with expression in the amygdala, cerebellum, cortex and striatum. These results provide evidence of the pathogenic role of PZMs and suggest the existence of distinct mechanisms between PZMs and germinal DNMs that are influencing ASD risk. |
| NFIA, NFIB, NFIX |
| Url: https://pubmed.ncbi.nlm.nih.gov/33130023  Title: Pathogenic nonsense variant in NFIB in another patient with dysmorphism, Autism Spectrum Disorder, agenesis of the corpus callosum, and intellectual disability.  The Nuclear Factor I (NFI) transcription family (NFIA, NFIB and NFIX) have been implicated in a range of developmental pathologies, including corpus callosum, craniofacial, urinary tract abnormalities, as well in the development of a number of neurodevelopmental developmental phenotypes including muscular hypotonia, motor and speech delay, attention deficit disorder, autism spectrum disorder, and behavioural abnormalities. NFIB haploinsufficiency has only recently been presented as a cause for macrocephaly-intellectual disability syndrome, with comparable phenotypes to NFIA related disorder. We add another patient with a previously reported nonsense variant in the NFIB who has Autism Spectrum Disorder level 2, agenesis of the corpus callosum, ADHD, obsessive compulsive Disorder and an intellectual disability. A clinical exome analysis identified a nonsense variant, c.265C > T, p.(Arg89\*) involving exon 2 of NFIB (ClinVar variation ID: 424,344). A brain MRI demonstrated agenesis of the corpus callosum. |
| NFIA, NFIB, NFIX |
| Url: https://pubmed.ncbi.nlm.nih.gov/30388402  Title: NFIB Haploinsufficiency Is Associated with Intellectual Disability and Macrocephaly.  The nuclear factor I (NFI) family of transcription factors play an important role in normal development of multiple organs. Three NFI family members are highly expressed in the brain, and deletions or sequence variants in two of these, NFIA and NFIX, have been associated with intellectual disability (ID) and brain malformations. NFIB, however, has not previously been implicated in human disease. Here, we present a cohort of 18 individuals with mild ID and behavioral issues who are haploinsufficient for NFIB. Ten individuals harbored overlapping microdeletions of the chromosomal 9p23-p22.2 region, ranging in size from 225 kb to 4.3 Mb. Five additional subjects had point sequence variations creating a premature termination codon, and three subjects harbored single-nucleotide variations resulting in an inactive protein as determined using an in vitro reporter assay. All individuals presented with additional variable neurodevelopmental phenotypes, including muscular hypotonia, motor and speech delay, attention deficit disorder, autism spectrum disorder, and behavioral abnormalities. While structural brain anomalies, including dysgenesis of corpus callosum, were variable, individuals most frequently presented with macrocephaly. To determine whether macrocephaly could be a functional consequence of NFIB disruption, we analyzed a cortex-specific Nfib conditional knockout mouse model, which is postnatally viable. Utilizing magnetic resonance imaging and histology, we demonstrate that Nfib conditional knockout mice have enlargement of the cerebral cortex but preservation of overall brain structure and interhemispheric connectivity. Based on our findings, we propose that haploinsufficiency of NFIB causes ID with macrocephaly. |
| NFIA, NR4A2, SOX5, TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25600067  Title: T-Brain-1--A Potential Master Regulator in Autism Spectrum Disorders.  T-Brain-1 (TBR1), a causative gene in autism spectrum disorders (ASDs), encodes a brain-specific T-box transcription factor. It is therefore possible that TBR1 controls the expression of other autism risk factors. The downstream genes of TBR1 have been identified using microarray and promoter analyses. In this study, we annotated individual genes downstream of TBR1 and investigated any associations with ASDs through extensive literature searches. Of 124 TBR1 target genes, 23 were reported to be associated with ASDs. In addition, one gene, Kiaa0319, is a known causative gene for dyslexia, a disorder frequently associated with autism. A change in expression level in 10 of these 24 genes has been previously confirmed. We further validated the alteration of RNA expression levels of Kiaa0319, Baiap2, and Gad1 in Tbr1 deficient mice. Among these 24 genes, four transcription factors Auts2, Nfia, Nr4a2, and Sox5 were found, suggesting that TBR1 controls a transcriptional cascade relevant to autism pathogenesis. A further five of the 24 genes (Cd44, Cdh8, Cntn6, Gpc6, and Ntng1) encode membrane proteins that regulate cell adhesion and axonal outgrowth. These genes likely contribute to the role of TBR1 in regulation of neuronal migration and axonal extension. Besides, decreases in Grin2b expression and increases in Gad1 expression imply that neuronal activity may be aberrant in Tbr1 deficient mice. These analyses provide direction for future experiments to reveal the pathogenic mechanism of autism. |
| NKX2-2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34046991  Title: MacroH2A1.2 deficiency leads to neural stem cell differentiation defects and autism-like behaviors.  The development of the nervous system requires precise regulation. Any disturbance in the regulation process can lead to neurological developmental diseases, such as autism and schizophrenia. Histone variants are important components of epigenetic regulation. The function and mechanisms of the macroH2A (mH2A) histone variant during brain development are unknown. Here, we show that deletion of the mH2A isoform mH2A1.2 interferes with neural stem cell differentiation in mice. Deletion of mH2A1.2 affects neurodevelopment, enhances neural progenitor cell (NPC) proliferation, and reduces NPC differentiation in the developing mouse brain. mH2A1.2-deficient mice exhibit autism-like behaviors, such as deficits in social behavior and exploratory abilities. We identify NKX2.2 as an important downstream effector gene and show that NKX2.2 expression is reduced after mH2A1.2 deletion and that overexpression of NKX2.2 rescues neuronal abnormalities caused by mH2A1.2 loss. Our study reveals that mH2A1.2 reduces the proliferation of neural progenitors and enhances neuronal differentiation during embryonic neurogenesis and that these effects are at least in part mediated by NKX2.2. These findings provide a basis for studying the relationship between mH2A1.2 and neurological disorders. |
| NKX2-2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33931583  Title: UTMOST, a single and cross-tissue TWAS (Transcriptome Wide Association Study), reveals new ASD (Autism Spectrum Disorder) associated genes.  Autism spectrum disorders (ASD) is a complex neurodevelopmental disorder that may significantly impact on the affected individual's life. Common variation (SNPs) could explain about 50% of ASD heritability. Despite this fact and the large size of the last GWAS meta-analysis, it is believed that hundreds of risk genes in ASD have yet to be discovered. New tools, such as TWAS (Transcriptome Wide Association Studies) which integrate tissue expression and genetic data, are a great approach to identify new ASD susceptibility genes. The main goal of this study is to use UTMOST with the publicly available summary statistics from the largest ASD GWAS meta-analysis as genetic input. In addition, an in silico biological characterization for the novel associated loci was performed. Our results have shown the association of 4 genes at the brain level (CIPC, PINX1, NKX2-2, and PTPRE) and have highlighted the association of NKX2-2, MANBA, ERI1, and MITF at the gastrointestinal level. The gastrointestinal associations are quite relevant given the well-established but unexplored relationship between ASD and gastrointestinal symptoms. Cross-tissue analysis has shown the association of NKX2-2 and BLK. UTMOST-associated genes together with their in silico biological characterization seems to point to different biological mechanisms underlying ASD etiology. Thus, it would not be restricted to brain tissue and it will involve the participation of other body tissues such as the gastrointestinal. |
| NKX2-2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31447886  Title: Novel Gene-Based Analysis of ASD GWAS: Insight Into the Biological Role of Associated Genes.  Background: Autism spectrum disorder (ASD) is a neurodevelopmental disorder characterized by its significant social impact and high heritability. The latest meta-analysis of ASD GWAS (genome-wide association studies) has revealed the association of several SNPs that were replicated in additional sets of independent samples. However, summary statistics from GWAS can be used to perform a gene-based analysis (GBA). GBA allows to combine all genetic information across the gene to create a single statistic (p-value for each gene). Thus, PASCAL (Pathway scoring algorithm), a novel GBA tool, has been applied to the summary statistics from the latest meta-analysis of ASD. GBA approach (testing the gene as a unit) provides an advantage to perform an accurate insight into the biological ASD mechanisms. Therefore, a gene-network analysis and an enrichment analysis for KEGG and GO terms were carried out. GENE2FUNC was used to create gene expression heatmaps and to carry out differential expression analysis (DEA) across GTEx v7 tissues and Brainspan data. dbMDEGA was employed to perform a DEG analysis between ASD and brain control samples for the associated genes and interactors. Results: PASCAL has identified the following loci associated with ASD: XRN2, NKX2-4, PLK1S1, KCNN2, NKX2-2, CRHR1-IT1, C8orf74 and LOC644172. While some of these genes were previously reported by MAGMA (XRN2, PLK1S1, and KCNN2), PASCAL has been useful to highlight additional genes. The biological characterization of the ASD-associated genes and their interactors have demonstrated the association of several GO and KEGG terms. Moreover, DEA analysis has revealed several up- and down-regulated clusters. In addition, many of the ASD-associated genes and their interactors have shown association with ASD expression datasets. Conclusions: This study identifies several associations at a gene level in ASD. Most of them were previously reported by MAGMA. This fact proves that PASCAL is an efficient GBA tool to extract additional information from previous GWAS. In addition, this study has characterized for the first time the biological role of the ASD-associated genes across brain regions, neurodevelopmental stages, and ASD gene-expression datasets. |
| NR1D1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30973326  Title: Shank3 modulates sleep and expression of circadian transcription factors.  Autism Spectrum Disorder (ASD) is the most prevalent neurodevelopmental disorder in the United States and often co-presents with sleep problems. Sleep problems in ASD predict the severity of ASD core diagnostic symptoms and have a considerable impact on the quality of life of caregivers. Little is known, however, about the underlying molecular mechanisms of sleep problems in ASD. We investigated the role of Shank3, a high confidence ASD gene candidate, in sleep architecture and regulation. We show that mice lacking exon 21 of Shank3 have problems falling asleep even when sleepy. Using RNA-seq we show that sleep deprivation increases the differences in prefrontal cortex gene expression between mutants and wild types, downregulating circadian transcription factors Per3, Bhlhe41, Hlf, Tef, and Nr1d1. Shank3 mutants also have trouble regulating wheel-running activity in constant darkness. Overall, our study shows that Shank3 is an important modulator of sleep and clock gene expression. |
| NR1D1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29449373  Title: Tyrosine hydroxylase down-regulation after loss of Abelson helper integration site 1 (AHI1) promotes depression via the circadian clock pathway in mice.  Abelson helper integration site 1 (AHI1) is associated with several neuropsychiatric and brain developmental disorders, such as schizophrenia, depression, autism, and Joubert syndrome. Ahi1 deficiency in mice leads to behaviors typical of depression. However, the mechanisms by which AHI1 regulates behavior remain to be elucidated. Here, we found that down-regulation of expression of the rate-limiting enzyme in dopamine biosynthesis, tyrosine hydroxylase (TH), in the midbrains of Ahi1-knockout (KO) mice is responsible for Ahi1-deficiency-mediated depressive symptoms. We also found that Rev-Erbα, a TH transcriptional repressor and circadian regulator, is up-regulated in the Ahi1-KO mouse midbrains and Ahi1-knockdown Neuro-2a cells. Moreover, brain and muscle Arnt-like protein 1 (BMAL1), the Rev-Erbα transcriptional regulator, is also increased in the Ahi1-KO mouse midbrains and Ahi1-knockdown cells. Our results further revealed that AHI1 decreases BMAL1/Rev-Erbα expression by interacting with and repressing retinoic acid receptor-related orphan receptor α, a nuclear receptor and transcriptional regulator of circadian genes. Of note, Bmal1 deficiency reversed the reduction in TH expression induced by Ahi1 deficiency. Moreover, microinfusion of the Rev-Erbα inhibitor SR8278 into the ventral midbrain of Ahi1-KO mice significantly increased TH expression in the ventral tegmental area and improved their depressive symptoms. These findings provide a mechanistic explanation for a link between AHI1-related behaviors and the circadian clock pathway, indicating an involvement of circadian regulatory proteins in AHI1-regulated mood and behavior. |
| NR1D1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28262759  Title: Role of a circadian-relevant gene NR1D1 in brain development: possible involvement in the pathophysiology of autism spectrum disorders.  In our previous study, we screened autism spectrum disorder (ASD) patients with and without sleep disorders for mutations in the coding regions of circadian-relevant genes, and detected mutations in several clock genes including NR1D1. Here, we further screened ASD patients for NR1D1 mutations and identified three novel mutations including a de novo heterozygous one c.1499 G > A (p.R500H). We then analyzed the role of Nr1d1 in the development of the cerebral cortex in mice. Acute knockdown of mouse Nr1d1 with in utero electroporation caused abnormal positioning of cortical neurons during corticogenesis. This aberrant phenotype was rescued by wild type Nr1d1, but not by the c.1499 G > A mutant. Time-lapse imaging revealed characteristic abnormal migration phenotypes in Nr1d1-deficient cortical neurons. When Nr1d1 was knocked down, axon extension and dendritic arbor formation of cortical neurons were also suppressed while proliferation of neuronal progenitors and stem cells at the ventricular zone was not affected. Taken together, Nr1d1 was found to play a pivotal role in corticogenesis via regulation of excitatory neuron migration and synaptic network formation. These results suggest that functional defects in NR1D1 may be related to ASD etiology and pathophysiology. |
| NR1D1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24765068  Title: Medial prefrontal cortex: genes linked to bipolar disorder and schizophrenia have altered expression in the highly social maternal phenotype.  The transition to motherhood involves CNS changes that modify sociability and affective state. However, these changes also put females at risk for post-partum depression and psychosis, which impairs parenting abilities and adversely affects children. Thus, changes in expression and interactions in a core subset of genes may be critical for emergence of a healthy maternal phenotype, but inappropriate changes of the same genes could put women at risk for post-partum disorders. This study evaluated microarray gene expression changes in medial prefrontal cortex (mPFC), a region implicated in both maternal behavior and psychiatric disorders. Post-partum mice were compared to virgin controls housed with females and isolated for identical durations. Using the Modular Single-set Enrichment Test (MSET), we found that the genetic landscape of maternal mPFC bears statistical similarity to gene databases associated with schizophrenia (5 of 5 sets) and bipolar disorder (BPD, 3 of 3 sets). In contrast to previous studies of maternal lateral septum (LS) and medial preoptic area (MPOA), enrichment of autism and depression-linked genes was not significant (2 of 9 sets, 0 of 4 sets). Among genes linked to multiple disorders were fatty acid binding protein 7 (Fabp7), glutamate metabotropic receptor 3 (Grm3), platelet derived growth factor, beta polypeptide (Pdgfrb), and nuclear receptor subfamily 1, group D, member 1 (Nr1d1). RT-qPCR confirmed these gene changes as well as FMS-like tyrosine kinase 1 (Flt1) and proenkephalin (Penk). Systems-level methods revealed involvement of developmental gene networks in establishing the maternal phenotype and indirectly suggested a role for numerous microRNAs and transcription factors in mediating expression changes. Together, this study suggests that a subset of genes involved in shaping the healthy maternal brain may also be dysregulated in mental health disorders and put females at risk for post-partum psychosis with aspects of schizophrenia and BPD. |
| NR2F1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32320667  Title: Imbalance of Excitatory/Inhibitory Neuron Differentiation in Neurodevelopmental Disorders with an NR2F1 Point Mutation.  Recent studies have revealed an essential role for embryonic cortical development in the pathophysiology of neurodevelopmental disorders, including autism spectrum disorder (ASD). However, the genetic basis and underlying mechanisms remain unclear. Here, we generate mutant human embryonic stem cell lines (Mut hESCs) carrying an NR2F1-R112K mutation that has been identified in a patient with ASD features and investigate their neurodevelopmental alterations. Mut hESCs overproduce ventral telencephalic neuron progenitors (ventral NPCs) and underproduce dorsal NPCs, causing the imbalance of excitatory/inhibitory neurons. These alterations can be mainly attributed to the aberrantly activated Hedgehog signaling pathway. Moreover, the corresponding Nr2f1 point-mutant mice display a similar excitatory/inhibitory neuron imbalance and abnormal behaviors. Antagonizing the increased inhibitory synaptic transmission partially alleviates their behavioral deficits. Together, our results suggest that the NR2F1-dependent imbalance of excitatory/inhibitory neuron differentiation caused by the activated Hedgehog pathway is one precursor of neurodevelopmental disorders and may enlighten the therapeutic approaches. |
| NR2F1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32275123  Title: Phenotypic expansion of Bosch-Boonstra-Schaaf optic atrophy syndrome and further evidence for genotype-phenotype correlations.  Bosch-Boonstra-Schaaf Optic Atrophy Syndrome (BBSOAS) is an autosomal dominant neurodevelopmental disorder caused by loss-of-function variants in NR2F1 and characterized by visual impairment, developmental delay, and intellectual disability. Here we report 18 new cases, provide additional clinical information for 9 previously reported individuals, and review an additional 27 published cases to present a total of 54 patients. Among these are 22 individuals with point mutations or in-frame deletions in the DNA-binding domain (DBD), and 32 individuals with other types of variants including whole-gene deletions, nonsense and frameshift variants, and point mutations outside the DBD. We corroborate previously described clinical characteristics including developmental delay, intellectual disability, autism spectrum disorder diagnoses/features thereof, cognitive/behavioral anomalies, hypotonia, feeding difficulties, abnormal brain MRI findings, and seizures. We also confirm a vision phenotype that includes optic nerve hypoplasia, optic atrophy, and cortical visual impairment. Additionally, we expand the vision phenotype to include alacrima and manifest latent nystagmus (fusional maldevelopment), and we broaden the behavioral phenotypic spectrum to include a love of music, an unusually good long-term memory, sleep difficulties, a high pain tolerance, and touch sensitivity. Furthermore, we provide additional evidence for genotype-phenotype correlations, specifically supporting a more severe phenotype associated with DBD variants. |
| NR2F1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30777838  Title: Beyond protein-coding genes.  A long non-coding RNA called lnc-NR2F1 regulates several neuronal genes, including some involved in autism and intellectual disabilities. |
| NR2F1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30653836  Title: Hyperactive and anxiolytic-like behaviors result from loss of COUP-TFI/Nr2f1 in the mouse cortex.  The nuclear receptor COUP TFI (also known as Nr2f1) plays major roles in specifying distinct neuronal subtypes during patterning of the neocortical motor and somatosensory cortex, as well as in regulating the longitudinal growth of the hippocampus during development. In humans, mutations in the NR2F1 gene lead to a global developmental delay and intellectual disabilities. While more than 30% of patients show behavioral features of autism spectrum disorder, 16% of haploinsufficient children show signs of hyperactivity and impulsivity. Loss of COUP-TFI in the cortical mouse primordium results in altered area organization and serotonin distribution, abnormal coordination of voluntary movements and learning and memory deficits. Here, we asked whether absence of COUP-TFI affects locomotor activity, anxiety, as well as depression. Mice mutant for COUP-TFI have normal motor coordination, but significant traits of hyperactivity, which does not seem to respond to N-Methyl-D-aspartate (NMDA) antagonists. However, no changes in anxiety, despite increased locomotor performances, were observed in the open field task. On the contrary, elevated plus maze and dark-light test explorations indicate a decreased anxiety-like behavior in COUP-TFI mutant mice. Finally, significantly reduced immobility in the forced swim test and no changes in anhedonia in the sucrose preference task suggest no particular depressive behaviors in mutant mice. Taken together, our study shows that loss of COUP-TFI leads to increased locomotor activity but less anxiety and contributes in further deciphering the pathophysiology of patients haploinsufficient for NR2F1. |
| NR2F1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30628890  Title: The novel lncRNA lnc-NR2F1 is pro-neurogenic and mutated in human neurodevelopmental disorders.  Long noncoding RNAs (lncRNAs) have been shown to act as important cell biological regulators including cell fate decisions but are often ignored in human genetics. Combining differential lncRNA expression during neuronal lineage induction with copy number variation morbidity maps of a cohort of children with autism spectrum disorder/intellectual disability versus healthy controls revealed focal genomic mutations affecting several lncRNA candidate loci. Here we find that a t(5:12) chromosomal translocation in a family manifesting neurodevelopmental symptoms disrupts specifically lnc-NR2F1. We further show that lnc-NR2F1 is an evolutionarily conserved lncRNA functionally enhances induced neuronal cell maturation and directly occupies and regulates transcription of neuronal genes including autism-associated genes. Thus, integrating human genetics and functional testing in neuronal lineage induction is a promising approach for discovering candidate lncRNAs involved in neurodevelopmental diseases. |
| NR4A2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35687495  Title: Cannabis alters DNA methylation at maternally imprinted and autism candidate genes in spermatogenic cells.  Cannabis use in the United States is increasing, with highest consumption among men at their peak reproductive years. We previously demonstrated widespread changes in sperm DNA methylation with cannabis exposure in humans and rats, including genes important in neurodevelopment. Here, we use an in vitro human spermatogenesis model to recapitulate chronic cannabis use and assess DNA methylation at imprinted and autism spectrum disorder (ASD) candidate genes in spermatogonial stem cell (SSC)- and spermatid-like cells. Methylation at maternally imprinted genes SGCE and GRB10 was significantly altered in SSC- and spermatid-like cells, respectively, while PEG3 was significantly differentially methylated in spermatid-like cells. Two of ten randomly selected ASD candidate genes, HCN1 and NR4A2, had significantly altered methylation with cannabis exposure in SSC-like cells. These results support our findings in human cohorts and provide a new tool with which to gain mechanistic insights into the association between paternal cannabis use and risk of ASD in offspring. |
| PAX5 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35947077  Title: Biallelic PAX5 mutations cause hypogammaglobulinemia, sensorimotor deficits, and autism spectrum disorder.  The genetic causes of primary antibody deficiencies and autism spectrum disorder (ASD) are largely unknown. Here, we report a patient with hypogammaglobulinemia and ASD who carries biallelic mutations in the transcription factor PAX5. A patient-specific Pax5 mutant mouse revealed an early B cell developmental block and impaired immune responses as the cause of hypogammaglobulinemia. Pax5 mutant mice displayed behavioral deficits in all ASD domains. The patient and the mouse model showed aberrant cerebellar foliation and severely impaired sensorimotor learning. PAX5 deficiency also caused profound hypoplasia of the substantia nigra and ventral tegmental area due to loss of GABAergic neurons, thus affecting two midbrain hubs, controlling motor function and reward processing, respectively. Heterozygous Pax5 mutant mice exhibited similar anatomic and behavioral abnormalities. Lineage tracing identified Pax5 as a crucial regulator of cerebellar morphogenesis and midbrain GABAergic neurogenesis. These findings reveal new roles of Pax5 in brain development and unravel the underlying mechanism of a novel immunological and neurodevelopmental syndrome. |
| PAX5 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35094443  Title: Delineation of a novel neurodevelopmental syndrome associated with PAX5 haploinsufficiency.  PAX5 is a transcription factor associated with abnormal posterior midbrain and cerebellum development in mice. PAX5 is highly loss-of-function intolerant and missense constrained, and has been identified as a candidate gene for autism spectrum disorder (ASD). We describe 16 individuals from 12 families who carry deletions involving PAX5 and surrounding genes, de novo frameshift variants that are likely to trigger nonsense-mediated mRNA decay, a rare stop-gain variant, or missense variants that affect conserved amino acid residues. Four of these individuals were published previously but without detailed clinical descriptions. All these individuals have been diagnosed with one or more neurodevelopmental phenotypes including delayed developmental milestones (DD), intellectual disability (ID), and/or ASD. Seizures were documented in four individuals. No recurrent patterns of brain magnetic resonance imaging (MRI) findings, structural birth defects, or dysmorphic features were observed. Our findings suggest that PAX5 haploinsufficiency causes a neurodevelopmental disorder whose cardinal features include DD, variable ID, and/or ASD. |
| PAX5 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34589851  Title: Immunity and autoantibodies of a mouse strain with autistic-like behavior.  Female and male mice of the BTBR T + Itpr3 tf /J (BTBR) strain have behaviors that resemble autism spectrum disorder. In comparison to C57BL/6 (B6) mice, BTBR mice have elevated humoral immunity, in that they have naturally high serum IgG levels and generate high levels of IgG antibodies, including autoantibodies to brain antigens. This study focused on the specificities of autoantibodies and the immune cells and their transcription factors that might be responsible for the autoantibodies. BTBR IgG autoantibodies bind to neurons better than microglia and with highest titer to nuclear antigens. Two of the antigens identified were alpha-enolase (ENO1) and dihydrolipoyllysine-residue succinyltransferase component of 2-oxoglutarate dehydrogenase complex, mitochondrial (DLST). Surprisingly based on IgG levels, the blood and spleens of BTBR mice have more CD4+ and CD8+ T cells, but fewer B cells than B6 mice. The high levels of autoantibodies in BTBR relates to their splenic T follicular helper (Tfh) cell levels, which likely are responsible for the higher number of plasma cells in BTBR mice than B6 mice. BTBR mice have increased gene expression of interleukin-21 receptor (I l -21 r) and Paired Box 5 (Pax5), which are known to aid B cell differentiation to plasma cells, and an increased Lysine Demethylase 6B (Kdm6b)/DNA Methyltransferase 1 (Dnmt1) ratio, which increases gene expression. Identification of gene expression and immune activities of BTBR mice may aid understanding of mechanisms associated with autism since neuroimmune network interactions have been posited and induction of autoantibodies may drive the neuroinflammation associated with autism. |
| PHF21A |
| Url: https://pubmed.ncbi.nlm.nih.gov/31649809  Title: Disruption of PHF21A causes syndromic intellectual disability with craniofacial anomalies, epilepsy, hypotonia, and neurobehavioral problems including autism.  PHF21A has been associated with intellectual disability and craniofacial anomalies based on its deletion in the Potocki-Shaffer syndrome region at 11p11.2 and its disruption in three patients with balanced translocations. In addition, three patients with de novo truncating mutations in PHF21A were reported recently. Here, we analyze genomic data from seven unrelated individuals with mutations in PHF21A and provide detailed clinical descriptions, further expanding the phenotype associated with PHF21A haploinsufficiency. Diagnostic trio whole exome sequencing, Sanger sequencing, use of GeneMatcher, targeted gene panel sequencing, and MiSeq sequencing techniques were used to identify and confirm variants. RT-qPCR was used to measure the normal expression pattern of PHF21A in multiple human tissues including 13 different brain tissues. Protein-DNA modeling was performed to substantiate the pathogenicity of the missense mutation. We have identified seven heterozygous coding mutations, among which six are de novo (not maternal in one). Mutations include four frameshifts, one nonsense mutation in two patients, and one heterozygous missense mutation in the AT Hook domain, predicted to be deleterious and likely to cause loss of PHF21A function. We also found a new C-terminal domain composed of an intrinsically disordered region. This domain is truncated in six patients and thus likely to play an important role in the function of PHF21A, suggesting that haploinsufficiency is the likely underlying mechanism in the phenotype of seven patients. Our results extend the phenotypic spectrum of PHF21A mutations by adding autism spectrum disorder, epilepsy, hypotonia, and neurobehavioral problems. Furthermore, PHF21A is highly expressed in the human fetal brain, which is consistent with the neurodevelopmental phenotype. Deleterious nonsense, frameshift, and missense mutations disrupting the AT Hook domain and/or an intrinsically disordered region in PHF21A were found to be associated with autism spectrum disorder, epilepsy, hypotonia, neurobehavioral problems, tapering fingers, clinodactyly, and syndactyly, in addition to intellectual disability and craniofacial anomalies. This suggests that PHF21A is involved in autism spectrum disorder and intellectual disability, and its haploinsufficiency causes a diverse neurological phenotype. |
| PHF21A |
| Url: https://pubmed.ncbi.nlm.nih.gov/27855486  Title: Yin-yang actions of histone methylation regulatory complexes in the brain.  Dysregulation of histone methylation has emerged as a major driver of neurodevelopmental disorders including intellectual disabilities and autism spectrum disorders. Histone methyl writer and eraser enzymes generally act within multisubunit complexes rather than in isolation. However, it remains largely elusive how such complexes cooperate to achieve the precise spatiotemporal gene expression in the developing brain. Histone H3K4 methylation (H3K4me) is a chromatin signature associated with active gene-regulatory elements. We review a body of literature that supports a model in which the RAI1-containing H3K4me writer complex counterbalances the LSD1-containing H3K4me eraser complex to ensure normal brain development. This model predicts H3K4me as the nexus of previously unrelated neurodevelopmental disorders. |
| PIK3CA |
| Url: https://pubmed.ncbi.nlm.nih.gov/30953832  Title: Abnormal axon guidance signals and reduced interhemispheric connection via anterior commissure in neonates of marmoset ASD model.  In autism spectrum disorder (ASD), disrupted functional and structural connectivity in the social brain has been suggested as the core biological mechanism underlying the social recognition deficits of this neurodevelopmental disorder. In this study, we aimed to identify genetic and neurostructural abnormalities at birth in a non-human primate model of ASD, the common marmoset with maternal exposure to valproic acid (VPA), which has been reported to display social recognition deficit in adulthood. Using a comprehensive gene expression analysis, we found that 20 genes were significantly downregulated in VPA-exposed neonates. Of these, Frizzled3 (FZD3) and PIK3CA were identified in an axon guidance signaling pathway. FZD3 is essential for the normal development of the anterior commissure (AC) and corpus callosum (CC); hence, we performed diffusion tensor magnetic resonance imaging with a 7-Tesla scanner to measure the midsagittal sizes of these structures. We found that the AC size in VPA-exposed neonates was significantly smaller than that in age-matched controls, while the CC size did not differ. These results suggest that downregulation of the genes related to axon guidance and decreased AC size in neonatal primates may be linked to social brain dysfunctions that can happen later in life. |
| PIK3CA |
| Url: https://pubmed.ncbi.nlm.nih.gov/29296277  Title: Identification of mutations in the PI3K-AKT-mTOR signalling pathway in patients with macrocephaly and developmental delay and/or autism.  Macrocephaly, which is defined as a head circumference greater than or equal to + 2 standard deviations, is a feature commonly observed in children with developmental delay and/or autism spectrum disorder. Although PTEN is a well-known gene identified in patients with this syndromic presentation, other genes in the PI3K-AKT-mTOR signalling pathway have also recently been suggested to have important roles. The aim of this study is to characterise the mutation spectrum of this group of patients. We performed whole-exome sequencing of 21 patients with macrocephaly and developmental delay/autism spectrum disorder. Sources of genomic DNA included blood, buccal mucosa and saliva. Germline mutations were validated by Sanger sequencing, whereas somatic mutations were validated by droplet digital PCR. We identified ten pathogenic/likely pathogenic mutations in PTEN (n = 4), PIK3CA (n = 3), MTOR (n = 1) and PPP2R5D (n = 2) in ten patients. An additional PTEN mutation, which was classified as variant of unknown significance, was identified in a patient with a pathogenic PTEN mutation, making him harbour bi-allelic germline PTEN mutations. Two patients harboured somatic PIK3CA mutations, and the level of somatic mosaicism in blood DNA was low. Patients who tested positive for mutations in the PI3K-AKT-mTOR pathway had a lower developmental quotient than the rest of the cohort (DQ = 62.8 vs. 76.1, p = 0.021). Their dysmorphic features were non-specific, except for macrocephaly. Among the ten patients with identified mutations, brain magnetic resonance imaging was performed in nine, all of whom showed megalencephaly. We identified mutations in the PI3K-AKT-mTOR signalling pathway in nearly half of our patients with macrocephaly and developmental delay/autism spectrum disorder. These patients have subtle dysmorphic features and mild developmental issues. Clinically, patients with germline mutations are difficult to distinguish from patients with somatic mutations, and therefore, sequencing of buccal or saliva DNA is important to identify somatic mosaicism. Given the high diagnostic yield and the management implications, we suggest implementing comprehensive genetic testing in the PI3K-AKT-mTOR pathway in the clinical evaluation of patients with macrocephaly and developmental delay and/or autism spectrum disorder. |
| PIK3CA |
| Url: https://pubmed.ncbi.nlm.nih.gov/26462458  Title: Currently recognized genes for schizophrenia: High-resolution chromosome ideogram representation.  A large body of genetic data from schizophrenia-related research has identified an assortment of genes and disturbed pathways supporting involvement of complex genetic components for schizophrenia spectrum and other psychotic disorders. Advances in genetic technology and expanding studies with searchable genomic databases have led to multiple published reports, allowing us to compile a master list of known, clinically relevant, or susceptibility genes contributing to schizophrenia. We searched key words related to schizophrenia and genetics from peer-reviewed medical literature sources, authoritative public access psychiatric websites and genomic databases dedicated to gene discovery and characterization of schizophrenia. Our list of 560 genes were arranged in alphabetical order in tabular form with gene symbols placed on high-resolution human chromosome ideograms. Genome wide pathway analysis using GeneAnalytics was carried out on the resulting list of genes to assess the underlying genetic architecture for schizophrenia. Recognized genes of clinical relevance, susceptibility or causation impact a broad range of biological pathways and mechanisms including ion channels (e.g., CACNA1B, CACNA1C, CACNA1H), metabolism (e.g., CYP1A2, CYP2C19, CYP2D6), multiple targets of neurotransmitter pathways impacting dopamine, GABA, glutamate, and serotonin function, brain development (e.g., NRG1, RELN), signaling peptides (e.g., PIK3CA, PIK4CA) and immune function (e.g., HLA-DRB1, HLA-DQA1) and interleukins (e.g., IL1A, IL10, IL6). This summary will enable clinical and laboratory geneticists, genetic counselors, and other clinicians to access convenient pictorial images of the distribution and location of contributing genes to inform diagnosis and gene-based treatment as well as provide risk estimates for genetic counseling of families with affected relatives. |
| PIK3CA |
| Url: https://pubmed.ncbi.nlm.nih.gov/21837366  Title: Social Responsiveness Scale-aided analysis of the clinical impact of copy number variations in autism.  Recent array-based studies have detected a wealth of copy number variations (CNVs) in patients with autism spectrum disorders (ASD). Since CNVs also occur in healthy individuals, their contributions to the patient's phenotype remain largely unclear. In a cohort of children with symptoms of ASD, diagnosis of the index patient using ADOS-G and ADI-R was performed, and the Social Responsiveness Scale (SRS) was administered to the index patients, both parents, and all available siblings. CNVs were identified using SNP arrays and confirmed by FISH or array CGH. To evaluate the clinical significance of CNVs, we analyzed three families with multiple affected children (multiplex) and six families with a single affected child (simplex) in which at least one child carried a CNV with a brain-transcribed gene. CNVs containing genes that participate in pathways previously implicated in ASD, such as the phosphoinositol signaling pathway (PIK3CA, GIRDIN), contactin-based networks of cell communication (CNTN6), and microcephalin (MCPH1) were found not to co-segregate with ASD phenotypes. In one family, a loss of CNTN5 co-segregated with disease. This indicates that most CNVs may by themselves not be sufficient to cause ASD, but still may contribute to the phenotype by additive or epistatic interactions with inherited (transmitted) mutations or non-genetic factors. Our study extends the scope of genome-wide CNV profiling beyond de novo CNVs in sporadic patients and may aid in uncovering missing heritability in genome-wide screening studies of complex psychiatric disorders. |
| PRR12 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29556724  Title: De novo apparent loss-of-function mutations in PRR12 in three patients with intellectual disability and iris abnormalities.  PRR12 encodes a proline-rich protein nuclear factor suspected to be involved in neural development. Its nuclear expression in fetal brains and in the vision system supports its role in brain and eye development more specifically. However, its function and potential role in human disease has not been determined. Recently, a de novo t(10;19) (q22.3;q13.33) translocation disrupting the PRR12 gene was detected in a girl with intellectual disability and neuropsychiatric alterations. Here we report on three unrelated patients with heterozygous de novo apparent loss-of-function mutations in PRR12 detected by clinical whole exome sequencing: c.1918G>T (p.Glu640\*), c.4502\_4505delTGCC (p.Leu1501Argfs\*146) and c.903\_909dup (p.Pro304Thrfs\*46). All three patients had global developmental delay, intellectual disability, eye and vision abnormalities, dysmorphic features, and neuropsychiatric problems. Eye abnormalities were consistent among the three patients and consisted of stellate iris pattern and iris coloboma. Additional variable clinical features included hypotonia, skeletal abnormalities, sleeping problems, and behavioral issues such as autism and anxiety. In summary, we propose that haploinsufficiency of PRR12 is associated with this novel multisystem neurodevelopmental disorder. |
| RERE |
| Url: https://pubmed.ncbi.nlm.nih.gov/36053530  Title: Phenotypic variability in RERE-related disorders and the first report of an inherited variant.  RERE-related disorders, also known as Neurodevelopmental Disorders with or without Anomalies of the Brain, Eye, or Heart (NEDBEH), are caused by heterozygous pathogenic variants in the arginine-glutamic acid dipeptide repeats gene (RERE). Up-to-date, 20 cases have been reported with the core characteristics of developmental delay, intellectual disability, and/or autism spectrum disorder. Here, we describe three additional cases. In the first case, the patient was found to have a previously reported de novo missense variant; her clinical findings of global developmental delay, intellectual disability, autism spectrum disorder, vision abnormalities, musculoskeletal anomalies, dysmorphic facial features, and a congenital heart defect strengthen existing genotype-phenotype correlations. We also describe the first inherited variant in RERE, found in a patient (case 2) with developmental delay, autism, and hyperopia and his mother (case 3) with ADHD, myopia, and history of mild speech delay. Lastly, by summarizing the clinical features presented in the 23 cases now reported, we provide an updated review of the literature. |
| RERE |
| Url: https://pubmed.ncbi.nlm.nih.gov/27087320  Title: De Novo Mutations of RERE Cause a Genetic Syndrome with Features that Overlap Those Associated with Proximal 1p36 Deletions.  Deletions of chromosome 1p36 affect approximately 1 in 5,000 newborns and are associated with developmental delay, intellectual disability, and defects involving the brain, eye, ear, heart, and kidney. Arginine-glutamic acid dipeptide repeats (RERE) is located in the proximal 1p36 critical region. RERE is a widely-expressed nuclear receptor coregulator that positively regulates retinoic acid signaling. Animal models suggest that RERE deficiency might contribute to many of the structural and developmental birth defects and medical problems seen in individuals with 1p36 deletion syndrome, although human evidence supporting this role has been lacking. In this report, we describe ten individuals with intellectual disability, developmental delay, and/or autism spectrum disorder who carry rare and putatively damaging changes in RERE. In all cases in which both parental DNA samples were available, these changes were found to be de novo. Associated features that were recurrently seen in these individuals included hypotonia, seizures, behavioral problems, structural CNS anomalies, ophthalmologic anomalies, congenital heart defects, and genitourinary abnormalities. The spectrum of defects documented in these individuals is similar to that of a cohort of 31 individuals with isolated 1p36 deletions that include RERE and are recapitulated in RERE-deficient zebrafish and mice. Taken together, our findings suggest that mutations in RERE cause a genetic syndrome and that haploinsufficiency of RERE might be sufficient to cause many of the phenotypes associated with proximal 1p36 deletions. |
| RFX3, RFX4, RFX7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33658631  Title: Disruption of RFX family transcription factors causes autism, attention-deficit/hyperactivity disorder, intellectual disability, and dysregulated behavior.  We describe a novel neurobehavioral phenotype of autism spectrum disorder (ASD), intellectual disability, and/or attention-deficit/hyperactivity disorder (ADHD) associated with de novo or inherited deleterious variants in members of the RFX family of genes. RFX genes are evolutionarily conserved transcription factors that act as master regulators of central nervous system development and ciliogenesis. We assembled a cohort of 38 individuals (from 33 unrelated families) with de novo variants in RFX3, RFX4, and RFX7. We describe their common clinical phenotypes and present bioinformatic analyses of expression patterns and downstream targets of these genes as they relate to other neurodevelopmental risk genes. These individuals share neurobehavioral features including ASD, intellectual disability, and/or ADHD; other frequent features include hypersensitivity to sensory stimuli and sleep problems. RFX3, RFX4, and RFX7 are strongly expressed in developing and adult human brain, and X-box binding motifs as well as RFX ChIP-seq peaks are enriched in the cis-regulatory regions of known ASD risk genes. These results establish a likely role of deleterious variation in RFX3, RFX4, and RFX7 in cases of monogenic intellectual disability, ADHD and ASD, and position these genes as potentially critical transcriptional regulators of neurobiological pathways associated with neurodevelopmental disease pathogenesis. |
| RFX4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/18378158  Title: Immune transcriptome alterations in the temporal cortex of subjects with autism.  Autism is a severe disorder that involves both genetic and environmental factors. Expression profiling of the superior temporal gyrus of six autistic subjects and matched controls revealed increased transcript levels of many immune system-related genes. We also noticed changes in transcripts related to cell communication, differentiation, cell cycle regulation and chaperone systems. Critical expression changes were confirmed by qPCR (BCL6, CHI3L1, CYR61, IFI16, IFITM3, MAP2K3, PTDSR, RFX4, SPP1, RELN, NOTCH2, RIT1, SFN, GADD45B, HSPA6, HSPB8 and SERPINH1). Overall, these expression patterns appear to be more associated with the late recovery phase of autoimmune brain disorders, than with the innate immune response characteristic of neurodegenerative diseases. Moreover, a variance-based analysis revealed much greater transcript variability in brains from autistic subjects compared to the control group, suggesting that these genes may represent autism susceptibility genes and should be assessed in follow-up genetic studies. |
| RFX7 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36334883  Title: Phenotype expansion and neurological manifestations of neurobehavioural disease caused by a variant in RFX7.  The RFX7 gene is one of eight genes within the regulatory factor X family. RFX7 is highly expressed in the brain and plays an important role in cell maturation and differentiation. It has only recently been implicated in disease in humans. Reports from 15 individuals have described RFX-associated phenotype as a neurobehavioural disease, manifesting primarily with global developmental delay and intellectual disability. Autism spectrum disorder and attention deficit hyperactivity disorder have also been described in some children. Here we report a case of a 19-month-old with a de novo missense variant in RFX7 resulting in severe global developmental delay including significant speech delay, microcephaly, dyskinetic movements, and failure to thrive. This is the first association between variants in RFX7 and failure to thrive, expanding the phenotype of this newly described gene. In this report we will also show RFX7 associated progressive central nervous system involvement through serial brain imaging. |
| RORB |
| Url: https://pubmed.ncbi.nlm.nih.gov/25950944  Title: Burden analysis of rare microdeletions suggests a strong impact of neurodevelopmental genes in genetic generalised epilepsies.  Genetic generalised epilepsy (GGE) is the most common form of genetic epilepsy, accounting for 20% of all epilepsies. Genomic copy number variations (CNVs) constitute important genetic risk factors of common GGE syndromes. In our present genome-wide burden analysis, large (≥ 400 kb) and rare (< 1%) autosomal microdeletions with high calling confidence (≥ 200 markers) were assessed by the Affymetrix SNP 6.0 array in European case-control cohorts of 1,366 GGE patients and 5,234 ancestry-matched controls. We aimed to: 1) assess the microdeletion burden in common GGE syndromes, 2) estimate the relative contribution of recurrent microdeletions at genomic rearrangement hotspots and non-recurrent microdeletions, and 3) identify potential candidate genes for GGE. We found a significant excess of microdeletions in 7.3% of GGE patients compared to 4.0% in controls (P = 1.8 x 10-7; OR = 1.9). Recurrent microdeletions at seven known genomic hotspots accounted for 36.9% of all microdeletions identified in the GGE cohort and showed a 7.5-fold increased burden (P = 2.6 x 10-17) relative to controls. Microdeletions affecting either a gene previously implicated in neurodevelopmental disorders (P = 8.0 x 10-18, OR = 4.6) or an evolutionarily conserved brain-expressed gene related to autism spectrum disorder (P = 1.3 x 10-12, OR = 4.1) were significantly enriched in the GGE patients. Microdeletions found only in GGE patients harboured a high proportion of genes previously associated with epilepsy and neuropsychiatric disorders (NRXN1, RBFOX1, PCDH7, KCNA2, EPM2A, RORB, PLCB1). Our results demonstrate that the significantly increased burden of large and rare microdeletions in GGE patients is largely confined to recurrent hotspot microdeletions and microdeletions affecting neurodevelopmental genes, suggesting a strong impact of fundamental neurodevelopmental processes in the pathogenesis of common GGE syndromes. |
| SATB2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36708662  Title: MicroRNA-218 regulates neuronal radial migration and morphogenesis by targeting Satb2 in developing neocortex.  Neuronal migration and morphogenesis are fundamental processes for cortical development. Their defects may cause abnormities in neural circuit formation and even neuropsychiatric disorders. Many proteins, especially layer-specific transcription factors and adhesion molecules, have been reported to regulate the processes. However, the involvement of non-coding RNAs in cortical development has not been extensively studied. Here, we identified microRNA-218 (miR-218) as a layer V-specific microRNA in mouse brains. Expression of miR-218 was elevated in patients with autism spectrum disorder (ASD) and schizophrenia. We found in this study that miR-218 overexpression in developing mouse cortex led to severe defects in radial migration, morphogenesis, and spatial distribution of the cortical neurons. Moreover, we identified Satb2, an upper-layer marker, as a molecular target repressed by miR-218. These results suggest an underlying mechanism of miR-218 involvement in neuropsychiatric disorders, and the interactions of layer-specific non-coding RNAs and proteins in regulating cortical development. |
| SATB2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31868578  Title: Aberrant calcium channel splicing drives defects in cortical differentiation in Timothy syndrome.  The syndromic autism spectrum disorder (ASD) Timothy syndrome (TS) is caused by a point mutation in the alternatively spliced exon 8A of the calcium channel Cav1.2. Using mouse brain and human induced pluripotent stem cells (iPSCs), we provide evidence that the TS mutation prevents a normal developmental switch in Cav1.2 exon utilization, resulting in persistent expression of gain-of-function mutant channels during neuronal differentiation. In iPSC models, the TS mutation reduces the abundance of SATB2-expressing cortical projection neurons, leading to excess CTIP2+ neurons. We show that expression of TS-Cav1.2 channels in the embryonic mouse cortex recapitulates these differentiation defects in a calcium-dependent manner and that in utero Cav1.2 gain-and-loss of function reciprocally regulates the abundance of these neuronal populations. Our findings support the idea that disruption of developmentally regulated calcium channel splicing patterns instructively alters differentiation in the developing cortex, providing important in vivo insights into the pathophysiology of a syndromic ASD. |
| SATB2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30809123  Title: Loss of Satb2 in the Cortex and Hippocampus Leads to Abnormal Behaviors in Mice.  Satb2-associated syndrome (SAS) is a genetic disorder that results from the deletion or mutation of one allele within the Satb2 locus. Patients with SAS show behavioral abnormalities, including developmental delay/intellectual disability, hyperactivity, and symptoms of autism. To address the role of Satb2 in SAS-related behaviors and generate an SAS mouse model, Satb2 was deleted in the cortex and hippocampus of Emx1-Cre; Satb2flox/flox [Satb2 conditional knockout (CKO)] mice. Satb2 CKO mice showed hyperactivity, increased impulsivity, abnormal social novelty, and impaired spatial learning and memory. Furthermore, we also found that the development of neurons in cortical layer IV was defective in Satb2 CKO mice, as shown by the loss of layer-specific gene expression and abnormal thalamocortical projections. In summary, the abnormal behaviors revealed in Satb2 CKO mice may reflect the SAS symptoms associated with Satb2 mutation in human patients, possibly due to defective development of cortical neurons in multiple layers including alterations of their inputs/outputs. |
| SATB2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30040823  Title: Genes regulated by SATB2 during neurodevelopment contribute to schizophrenia and educational attainment.  SATB2 is associated with schizophrenia and is an important transcription factor regulating neocortical organization and circuitry. Rare mutations in SATB2 cause a syndrome that includes developmental delay, and mouse studies identify an important role for SATB2 in learning and memory. Interacting partners BCL11B and GATAD2A are also schizophrenia risk genes indicating that other genes interacting with or are regulated by SATB2 are making a contribution to schizophrenia and cognition. We used data from Satb2 mouse models to generate three gene-sets that contain genes either functionally related to SATB2 or targeted by SATB2 at different stages of development. Each was tested for enrichment using the largest available genome-wide association studies (GWAS) datasets for schizophrenia and educational attainment (EA) and enrichment analysis was also performed for schizophrenia and other neurodevelopmental disorders using data from rare variant sequencing studies. These SATB2 gene-sets were enriched for genes containing common variants associated with schizophrenia and EA, and were enriched for genes containing rare variants reported in studies of schizophrenia, autism and intellectual disability. In the developing cortex, genes targeted by SATB2 based on ChIP-seq data, and functionally affected when SATB2 is not expressed based on differential expression analysis using RNA-seq data, show strong enrichment for genes associated with EA. For genes expressed in the hippocampus or at the synapse, those targeted by SATB2 are more strongly enriched for genes associated EA than gene-sets not targeted by SATB2. This study demonstrates that single gene findings from GWAS can provide important insights to pathobiological processes. In this case we find evidence that genes influenced by SATB2 and involved in synaptic transmission, axon guidance and formation of the corpus callosum are contributing to schizophrenia and cognition. |
| SATB2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24109430  Title: Lack of serotonin reuptake during brain development alters rostral raphe-prefrontal network formation.  Besides its "classical" neurotransmitter function, serotonin (5-HT) has been found to also act as a neurodevelopmental signal. During development, the 5-HT projection system, besides an external placental source, represents one of the earliest neurotransmitter systems to innervate the brain. One of the targets of the 5-HT projection system, originating in the brainstem raphe nuclei, is the medial prefrontal cortex (mPFC), an area involved in higher cognitive functions and important in the etiology of many neurodevelopmental disorders. Little is known, however, about the exact role of 5-HT and its signaling molecules in the formation of the raphe-prefrontal network. Using explant essays, we here studied the role of the 5-HT transporter (5-HTT), an important modulator of the 5-HT signal, in rostral raphe-prefrontal network formation. We found that the chemotrophic nature of the interaction between the origin (rostral raphe cluster) and a target (mPFC) of the 5-HT projection system was affected in rats lacking the 5-HTT (5-HTT(-/-)). While 5-HTT deficiency did not affect the dorsal raphe 5-HT-positive outgrowing neurites, the median raphe 5-HT neurites switched from a strong repulsive to an attractive interaction when co-cultured with the mPFC. Furthermore, the fasciculation of the mPFC outgrowing neurites was dependent on the amount of 5-HTT. In the mPFC of 5-HTT(-/-) pups, we observed clear differences in 5-HT innervation and the identity of a class of projection neurons of the mPFC. In the absence of the 5-HTT, the 5-HT innervation in all subareas of the early postnatal mPFC increased dramatically and the number of Satb2-positive callosal projection neurons was decreased. Together, these results suggest a 5-HTT dependency during early development of these brain areas and in the formation of the raphe-prefrontal network. The tremendous complexity of the 5-HT projection system and its role in several neurodevelopmental disorders highlights the need for further research in this largely unexplored area. |
| SETD2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36537238  Title: CHD8 suppression impacts on histone H3 lysine 36 trimethylation and alters RNA alternative splicing.  Disruptive mutations in the chromodomain helicase DNA-binding protein 8 gene (CHD8) have been recurrently associated with autism spectrum disorders (ASDs). Here we investigated how chromatin reacts to CHD8 suppression by analyzing a panel of histone modifications in induced pluripotent stem cell-derived neural progenitors. CHD8 suppression led to significant reduction (47.82%) in histone H3K36me3 peaks at gene bodies, particularly impacting on transcriptional elongation chromatin states. H3K36me3 reduction specifically affects highly expressed, CHD8-bound genes and correlates with altered alternative splicing patterns of 462 genes implicated in 'regulation of RNA splicing' and 'mRNA catabolic process'. Mass spectrometry analysis uncovered a novel interaction between CHD8 and the splicing regulator heterogeneous nuclear ribonucleoprotein L (hnRNPL), providing the first mechanistic insights to explain the CHD8 suppression-derived splicing phenotype, partly implicating SETD2, a H3K36me3 methyltransferase. In summary, our results point toward broad molecular consequences of CHD8 suppression, entailing altered histone deposition/maintenance and RNA processing regulation as important regulatory processes in ASD. |
| SETD2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34014281  Title: Neuronal SETD2 activity links microtubule methylation to an anxiety-like phenotype in mice.  Gene discovery efforts in autism spectrum disorder have identified heterozygous defects in chromatin remodeller genes, the 'readers, writers and erasers' of methyl marks on chromatin, as major contributors to this disease. Despite this advance, a convergent aetiology between these defects and aberrant chromatin architecture or gene expression has remained elusive. Recently, data have begun to emerge that chromatin remodellers also function directly on the cytoskeleton. Strongly associated with autism spectrum disorder, the SETD2 histone methyltransferase for example, has now been shown to directly methylate microtubules of the mitotic spindle. However, whether microtubule methylation occurs in post-mitotic cells, for example on the neuronal cytoskeleton, is not known. We found the SETD2 α-tubulin lysine 40 trimethyl mark occurs on microtubules in the brain and in primary neurons in culture, and that the SETD2 C-terminal SRI domain is required for binding and methylation of α-tubulin. A CRISPR knock-in of a pathogenic SRI domain mutation (Setd2SRI) that disables microtubule methylation revealed at least one wild-type allele was required in mice for survival, and while viable, heterozygous Setd2SRI/wtmice exhibited an anxiety-like phenotype. Finally, whereas RNA-sequencing (RNA-seq) and chromatin immunoprecipitation-sequencing (ChIP-seq) showed no concomitant changes in chromatin methylation or gene expression in Setd2SRI/wtmice, primary neurons exhibited structural deficits in axon length and dendritic arborization. These data provide the first demonstration that microtubules of neurons are methylated, and reveals a heterozygous chromatin remodeller defect that specifically disables microtubule methylation is sufficient to drive an autism-associated phenotype. |
| SETD2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32234480  Title: FMRP Control of Ribosome Translocation Promotes Chromatin Modifications and Alternative Splicing of Neuronal Genes Linked to Autism.  Silencing of FMR1 and loss of its gene product, FMRP, results in fragile X syndrome (FXS). FMRP binds brain mRNAs and inhibits polypeptide elongation. Using ribosome profiling of the hippocampus, we find that ribosome footprint levels in Fmr1-deficient tissue mostly reflect changes in RNA abundance. Profiling over a time course of ribosome runoff in wild-type tissue reveals a wide range of ribosome translocation rates; on many mRNAs, the ribosomes are stalled. Sucrose gradient ultracentrifugation of hippocampal slices after ribosome runoff reveals that FMRP co-sediments with stalled ribosomes, and its loss results in decline of ribosome stalling on specific mRNAs. One such mRNA encodes SETD2, a lysine methyltransferase that catalyzes H3K36me3. Chromatin immunoprecipitation sequencing (ChIP-seq) demonstrates that loss of FMRP alters the deployment of this histone mark. H3K36me3 is associated with alternative pre-RNA processing, which we find occurs in an FMRP-dependent manner on transcripts linked to neural function and autism spectrum disorders. |
| SETD2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29294103  Title: Brain-specific deletion of histone variant H2A.z results in cortical neurogenesis defects and neurodevelopmental disorder.  Defects in neurogenesis alter brain circuit formations and may lead to neurodevelopmental disorders such as autism and schizophrenia. Histone H2A.z, a variant of histone H2A, plays critical roles in chromatin structure and epigenetic regulation, but its function and mechanism in brain development remain largely unknown. Here, we find that the deletion of H2A.z results in enhanced proliferation of neural progenitors but reduced neuronal differentiation. In addition, neurons in H2A.z knockout mice exhibit abnormal dendrites during brain development. Furthermore, H2A.zcKO mice exhibit serial behavioral deficits, such as decreased exploratory activity and impaired learning and memory. Mechanistically, H2A.z regulates embryonic neurogenesis by targeting Nkx2-4 through interaction with Setd2, thereby promoting H3K36me3 modification to activate the transcription of Nkx2-4. Furthermore, enforced expression of Nkx2-4 can rescue the defective neurogenesis in the H2A.z-knockdown embryonic brain. Together, our findings implicate the epigenetic regulation by H2A.z in embryonic neurogenesis and provide a framework for understanding how disruption in the H2A.z gene may contribute to neurological disorders. |
| SETDB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35682642  Title: Enhanced Expression of Human Endogenous Retroviruses, TRIM28 and SETDB1 in Autism Spectrum Disorder.  Human endogenous retroviruses (HERVs) are relics of ancestral infections and represent 8% of the human genome. They are no longer infectious, but their activation has been associated with several disorders, including neuropsychiatric conditions. Enhanced expression of HERV-K and HERV-H envelope genes has been found in the blood of autism spectrum disorder (ASD) patients, but no information is available on syncytin 1 (SYN1), SYN2, and multiple sclerosis-associated retrovirus (MSRV), which are thought to be implicated in brain development and immune responses. HERV activation is regulated by TRIM28 and SETDB1, which are part of the epigenetic mechanisms that organize the chromatin architecture in response to external stimuli and are involved in neural cell differentiation and brain inflammation. We assessed, through a PCR realtime Taqman amplification assay, the transcription levels of pol genes of HERV-H, -K, and -W families, of env genes of SYN1, SYN2, and MSRV, as well as of TRIM28 and SETDB1 in the blood of 33 ASD children (28 males, median 3.8 years, 25-75% interquartile range 3.0-6.0 y) and healthy controls (HC). Significantly higher expressions of TRIM28 and SETDB1, as well as of all the HERV genes tested, except for HERV-W-pol, were found in ASD, as compared with HC. Positive correlations were observed between the mRNA levels of TRIM28 or SETDB1 and every HERV gene in ASD patients, but not in HC. Overexpression of TRIM28/SETDB1 and several HERVs in children with ASD and the positive correlations between their transcriptional levels suggest that these may be main players in pathogenetic mechanisms leading to ASD. |
| SETDB1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33279625  Title: Histone lysine methyltransferase SETDB1 as a novel target for central nervous system diseases.  Epigenetic changes that regulate chromatin structure have a major impact in genome stabilization and maintenance of cellular homeostasis, been recently implicated in the pathophysiology of central nervous system (CNS). Aberrant expression and dysregulation of histone modification enzymes has been associated with the development of several CNS disorders, revealing these enzymes as putative targets for drug development and novel therapeutic approaches. SETDB1 is a histone lysine methyltransferase responsible for the di- and tri-methylation of histone 3 (H3) at lysine (K) 9 in euchromatic regions further promoting gene silencing through heterochromatin formation. By this way, SETDB1 has been shown to regulate gene expression and influence normal cellular homeostasis required for nervous system function while it is also implicated in the pathogenesis of CNS disorders. Among them, brain tumors, schizophrenia, Huntington's disease, autism spectrum disorders along with alcohol-induced fetal neurobehavioral deficits and Prader-Willi syndrome are representative examples, indicating the aberrant expression and function of SETDB1 as a common pathogenic factor. In this review, we focus on SETDB1-associated molecular mechanisms implicated in CNS physiology and disease while we further discuss current pharmacological approaches targeting SETDB1 enzymatic activity with beneficial effects. |
| SHOX |
| Url: https://pubmed.ncbi.nlm.nih.gov/27073233  Title: Microduplications at the pseudoautosomal SHOX locus in autism spectrum disorders and related neurodevelopmental conditions.  The pseudoautosomal short stature homeobox-containing (SHOX) gene encodes a homeodomain transcription factor involved in cell-cycle and growth regulation. SHOX/SHOX enhancers deletions cause short stature and skeletal abnormalities in a female-dominant fashion; duplications appear to be rare. Neurodevelopmental disorders (NDDs), such as autism spectrum disorders (ASDs), are complex disorders with high heritability and skewed sex ratio; several rare (<1% frequency) CNVs have been implicated in risk. We analysed data from a discovery series of 90 adult ASD cases, who underwent clinical genetic testing by array-comparative genomic hybridisation (CGH). Twenty-seven individuals harboured CNV abnormalities, including two unrelated females with microduplications affecting SHOX. To determine the prevalence of SHOX duplications and delineate their associated phenotypic spectrum, we subsequently examined array-CGH data from a follow-up sample of 26 574 patients, including 18 857 with NDD (3541 with ASD). We found a significant enrichment of SHOX microduplications in the NDD cases (p=0.00036; OR 2.21) and, particularly, in those with ASD (p=9.18×10(-7); OR 3.63) compared with 12 594 population-based controls. SHOX duplications affecting the upstream or downstream enhancers were enriched only in females with NDD (p=0.0043; OR 2.69/p=0.00020; OR 7.20), but not in males (p=0.404; OR 1.38/p=0.096; OR 2.21). Microduplications at the SHOX locus are a low penetrance risk factor for ASD/NDD, with increased risk in both sexes. However, a concomitant duplication of SHOX enhancers may be required to trigger a NDD in females. Since specific SHOX isoforms are exclusively expressed in the developing foetal brain, this may reflect the pathogenic effect of altered SHOX protein dosage on neurodevelopment. |
| SOX5, TCF4, YY1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34947998  Title: Autism-Related Transcription Factors Underlying the Sex-Specific Effects of Prenatal Bisphenol A Exposure on Transcriptome-Interactome Profiles in the Offspring Prefrontal Cortex.  Bisphenol A (BPA) is an environmental risk factor for autism spectrum disorder (ASD). BPA exposure dysregulates ASD-related genes in the hippocampus and neurological functions of offspring. However, whether prenatal BPA exposure has an impact on genes in the prefrontal cortex, another brain region highly implicated in ASD, and through what mechanisms have not been investigated. Here, we demonstrated that prenatal BPA exposure disrupts the transcriptome-interactome profiles of the prefrontal cortex of neonatal rats. Interestingly, the list of BPA-responsive genes was significantly enriched with known ASD candidate genes, as well as genes that were dysregulated in the postmortem brain tissues of ASD cases from multiple independent studies. Moreover, several differentially expressed genes in the offspring's prefrontal cortex were the targets of ASD-related transcription factors, including AR, ESR1, and RORA. The hypergeometric distribution analysis revealed that BPA may regulate the expression of such genes through these transcription factors in a sex-dependent manner. The molecular docking analysis of BPA and ASD-related transcription factors revealed novel potential targets of BPA, including RORA, SOX5, TCF4, and YY1. Our findings indicated that prenatal BPA exposure disrupts ASD-related genes in the offspring's prefrontal cortex and may increase the risk of ASD through sex-dependent molecular mechanisms, which should be investigated further. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36678498  Title: The Role of Zinc and NMDA Receptors in Autism Spectrum Disorders.  NMDA-type glutamate receptors are critical for synaptic plasticity in the central nervous system. Their unique properties and age-dependent arrangement of subunit types underpin their role as a coincidence detector of pre- and postsynaptic activity during brain development and maturation. NMDAR function is highly modulated by zinc, which is co-released with glutamate and concentrates in postsynaptic spines. Both NMDARs and zinc have been strongly linked to autism spectrum disorders (ASDs), suggesting that NMDARs are an important player in the beneficial effects observed with zinc in both animal models and children with ASDs. Significant evidence is emerging that these beneficial effects occur via zinc-dependent regulation of SHANK proteins, which form the backbone of the postsynaptic density. For example, dietary zinc supplementation enhances SHANK2 or SHANK3 synaptic recruitment and rescues NMDAR deficits and hypofunction in Shank3ex13-16-/- and Tbr1+/- ASD mice. Across multiple studies, synaptic changes occur in parallel with a reversal of ASD-associated behaviours, highlighting the zinc-dependent regulation of NMDARs and glutamatergic synapses as therapeutic targets for severe forms of ASDs, either pre- or postnatally. The data from rodent models set a strong foundation for future translational studies in human cells and people affected by ASDs. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35303947  Title: Dietary zinc supplementation rescues fear-based learning and synaptic function in the Tbr1+/- mouse model of autism spectrum disorders.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder characterised by a dyad of behavioural symptoms-social and communication deficits and repetitive behaviours. Multiple aetiological genetic and environmental factors have been identified as causing or increasing the likelihood of ASD, including serum zinc deficiency. Our previous studies revealed that dietary zinc supplementation can normalise impaired social behaviours, excessive grooming, and heightened anxiety in a Shank3 mouse model of ASD, as well as the amelioration of synapse dysfunction. Here, we have examined the efficacy and breadth of dietary zinc supplementation as an effective therapeutic strategy utilising a non-Shank-related mouse model of ASD-mice with Tbr1 haploinsufficiency. We performed behavioural assays, amygdalar slice whole-cell patch-clamp electrophysiology, and immunohistochemistry to characterise the synaptic mechanisms underlying the ASD-associated behavioural deficits observed in Tbr1+/- mice and the therapeutic potential of dietary zinc supplementation. Two-way analysis of variance (ANOVA) with Šídák's post hoc test and one-way ANOVA with Tukey's post hoc multiple comparisons were performed for statistical analysis. Our data show that dietary zinc supplementation prevents impairments in auditory fear memory and social interaction, but not social novelty, in the Tbr1+/- mice. Tbr1 haploinsufficiency did not induce excessive grooming nor elevate anxiety in mice. At the synaptic level, dietary zinc supplementation reversed α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPAR) and N-methyl-D-aspartate receptor (NMDAR) hypofunction and normalised presynaptic function at thalamic-lateral amygdala (LA) synapses that are crucial for auditory fear memory. In addition, the zinc supplemented diet significantly restored the synaptic puncta density of the GluN1 subunit essential for functional NMDARs as well as SHANK3 expression in both the basal and lateral amygdala (BLA) of Tbr1+/- mice. The therapeutic effect of dietary zinc supplementation observed in rodent models may not reproduce the same effects in human patients. The effect of dietary zinc supplementation on synaptic function in other brain structures affected by Tbr1 haploinsufficiency including olfactory bulb and anterior commissure will also need to be examined. Our data further the understanding of the molecular mechanisms underlying the effect of dietary zinc supplementation and verify the efficacy and breadth of its application as a potential treatment strategy for ASD. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35123407  Title: LiCl treatment leads to long-term restoration of spine maturation and synaptogenesis in adult Tbr1 mutants.  Tbr1 encodes a T-box transcription factor and is considered a high confidence autism spectrum disorder (ASD) gene. Tbr1 is expressed in the postmitotic excitatory neurons of the deep neocortical layers 5 and 6. Postnatally and neonatally, Tbr1 conditional mutants (CKOs) have immature dendritic spines and reduced synaptic density. However, an understanding of Tbr1's function in the adult mouse brain remains elusive. We used conditional mutagenesis to interrogate Tbr1's function in cortical layers 5 and 6 of the adult mouse cortex. Adult Tbr1 CKO mutants have dendritic spine and synaptic deficits as well as reduced frequency of mEPSCs and mIPSCs. LiCl, a WNT signaling agonist, robustly rescues the dendritic spine maturation, synaptic defects, and excitatory and inhibitory synaptic transmission deficits. LiCl treatment could be used as a therapeutic approach for some cases of ASD with deficits in synaptic transmission. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34409402  Title: Mid-pregnancy maternal immune activation increases Pax6-positive and Tbr2-positive neural progenitor cells and causes integrated stress response in the fetal brain in a mouse model of maternal viral infection.  Maternal immune activation (MIA) in midpregnancy is a risk factor for neurodevelopmental disorders. Improper brain development may cause malformations of the brain; maldevelopment induced by MIA may lead to a pathology-related phenotype. In this study, a single intraperitoneal injection of 20 mg/kg polyriboinosinic-polyribocytidylic acid [poly(I:C)] was administered to C57BL/6J mice on embryonic day (E) 12.5 to mimic maternal viral infection. Histopathological analysis of neurogenesis was performed using markers for Pax6, Tbr2, and Tbr1. In these fetuses, significant increases were observed in the proportion of Pax6-positive neural progenitor cells and Pax6/Tbr2 double-positive cells 24 h after poly(I:C) injection. There were no differences in the proportion of Tbr1-positive postmitotic neurons 48 h after poly(I:C) injection. At E18.5, there were more Pax6-positive and Tbr2-positive neural progenitor cells in the poly(I:C)-injected group than in the saline-injected group. Gene ontology enrichment analysis of poly(I:C)-induced differentially expressed genes in the fetal brain at E12.5 demonstrated that these genes were enriched in terms including response to cytokine, response to decreased oxygen levels in the category of biological process. At E13.5, activating transcription factor 4 (Atf4), which is an effector of integrated stress response, was significantly upregulated in the fetal brain. Our results show that poly(I:C)-induced MIA at E12.5 leads to dysregulated neurogenesis and upregulates Atf4 in the fetal brain. These findings provide a new insight in the mechanism of MIA causing improper brain development and subsequent neurodevelopmental disorders. |
| TBR1, ZBTB16 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33895774  Title: Zbtb16 regulates social cognitive behaviors and neocortical development.  Zinc finger and BTB domain containing 16 (ZBTB16) play the roles in the neural progenitor cell proliferation and neuronal differentiation during development, however, how the function of ZBTB16 is involved in brain function and behaviors unknown. Here we show the deletion of Zbtb16 in mice leads to social impairment, repetitive behaviors, risk-taking behaviors, and cognitive impairment. To elucidate the mechanism underlying the behavioral phenotypes, we conducted histological analyses and observed impairments in thinning of neocortical layer 6 (L6) and a reduction of TBR1+ neurons in Zbtb16 KO mice. Furthermore, we found increased dendritic spines and microglia, as well as developmental defects in oligodendrocytes and neocortical myelination in the prefrontal cortex (PFC) of Zbtb16 KO mice. Using genomics approaches, we identified the Zbtb16 transcriptome that includes genes involved in neocortical maturation such as neurogenesis and myelination, and both autism spectrum disorder (ASD) and schizophrenia (SCZ) pathobiology. Co-expression networks further identified Zbtb16-correlated modules that are unique to ASD or SCZ, respectively. Our study provides insight into the novel roles of ZBTB16 in behaviors and neocortical development related to the disorders. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32948248  Title: A de novo frameshift pathogenic variant in TBR1 identified in autism without intellectual disability.  In order to be able to provide accurate genetic counseling to patients with Autism Spectrum Disorder (ASD), it is crucial to identify correlations between heterogeneous phenotypes and genetic alterations. Among the hundreds of de novo pathogenic variants reported in ASD, single-nucleotide variations and small insertions/deletions were reported in TBR1. This gene encodes a transcription factor that plays a key role in brain development. Pathogenic variants in TBR1 are often associated with severe forms of ASD, including intellectual disability and language impairment. Adults diagnosed with ASD but without intellectual disability (diagnosis of Asperger syndrome, according to the DSM-IV) took part in a genetic consultation encompassing metabolic assessments, a molecular karyotype and the screening of a panel of 268 genes involved in intellectual disability, ASD and epilepsy. In addition, the patient reported here went through a neuropsychological assessment, structural magnetic resonance imaging and magnetic resonance spectroscopy measurements. Here, we report the case of a young adult male who presents with a typical form of ASD. Importantly, this patient presents with no intellectual disability or language impairment, despite a de novo heterozygous frameshift pathogenic variant in TBR1, leading to an early premature termination codon (c.26del, p.(Pro9Leufs\*12)). Based on this case report, we discuss the role of TBR1 in general brain development, language development, intellectual disability and other symptoms of ASD. Providing a detailed clinical description of the individuals with such pathogenic variants should help to understand the genotype-phenotype relationships in ASD. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32144139  Title: Central neurogenetic signatures of the visuomotor integration system.  Visuomotor impairments characterize numerous neurological disorders and neurogenetic syndromes, such as autism spectrum disorder (ASD) and Dravet, Fragile X, Prader-Willi, Turner, and Williams syndromes. Despite recent advances in systems neuroscience, the biological basis underlying visuomotor functional impairments associated with these clinical conditions is poorly understood. In this study, we used neuroimaging connectomic approaches to map the visuomotor integration (VMI) system in the human brain and investigated the topology approximation of the VMI network to the Allen Human Brain Atlas, a whole-brain transcriptome-wide atlas of cortical genetic expression. We found the genetic expression of four genes-TBR1, SCN1A, MAGEL2, and CACNB4-to be prominently associated with visuomotor integrators in the human cortex. TBR1 gene transcripts, an ASD gene whose expression is related to neural development of the cortex and the hippocampus, showed a central spatial allocation within the VMI system. Our findings delineate gene expression traits underlying the VMI system in the human cortex, where specific genes, such as TBR1, are likely to play a central role in its neuronal organization, as well as on specific phenotypes of neurogenetic syndromes. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32005960  Title: De novo TBR1 variants cause a neurocognitive phenotype with ID and autistic traits: report of 25 new individuals and review of the literature.  TBR1, a T-box transcription factor expressed in the cerebral cortex, regulates the expression of several candidate genes for autism spectrum disorders (ASD). Although TBR1 has been reported as a high-confidence risk gene for ASD and intellectual disability (ID) in functional and clinical reports since 2011, TBR1 has only recently been recorded as a human disease gene in the OMIM database. Currently, the neurodevelopmental disorders and structural brain anomalies associated with TBR1 variants are not well characterized. Through international data sharing, we collected data from 25 unreported individuals and compared them with data from the literature. We evaluated structural brain anomalies in seven individuals by analysis of MRI images, and compared these with anomalies observed in TBR1 mutant mice. The phenotype included ID in all individuals, associated to autistic traits in 76% of them. No recognizable facial phenotype could be identified. MRI analysis revealed a reduction of the anterior commissure and suggested new features including dysplastic hippocampus and subtle neocortical dysgenesis. This report supports the role of TBR1 in ID associated with autistic traits and suggests new structural brain malformations in humans. We hope this work will help geneticists to interpret TBR1 variants and diagnose ASD probands. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31680851  Title: A TBR1-K228E Mutation Induces Tbr1 Upregulation, Altered Cortical Distribution of Interneurons, Increased Inhibitory Synaptic Transmission, and Autistic-Like Behavioral Deficits in Mice.  Mutations in Tbr1, a high-confidence ASD (autism spectrum disorder)-risk gene encoding the transcriptional regulator TBR1, have been shown to induce diverse ASD-related molecular, synaptic, neuronal, and behavioral dysfunctions in mice. However, whether Tbr1 mutations derived from autistic individuals cause similar dysfunctions in mice remains unclear. Here we generated and characterized mice carrying the TBR1-K228E de novo mutation identified in human ASD and identified various ASD-related phenotypes. In heterozygous mice carrying this mutation (Tbr1 +/K228E mice), levels of the TBR1-K228E protein, which is unable to bind target DNA, were strongly increased. RNA-Seq analysis of the Tbr1 +/K228E embryonic brain indicated significant changes in the expression of genes associated with neurons, astrocytes, ribosomes, neuronal synapses, and ASD risk. The Tbr1 +/K228E neocortex also displayed an abnormal distribution of parvalbumin-positive interneurons, with a lower density in superficial layers but a higher density in deep layers. These changes were associated with an increase in inhibitory synaptic transmission in layer 6 pyramidal neurons that was resistant to compensation by network activity. Behaviorally, Tbr1 +/K228E mice showed decreased social interaction, increased self-grooming, and modestly increased anxiety-like behaviors. These results suggest that the human heterozygous TBR1-K228E mutation induces ASD-related transcriptomic, protein, neuronal, synaptic, and behavioral dysfunctions in mice. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30792833  Title: Haploinsufficiency of autism causative gene Tbr1 impairs olfactory discrimination and neuronal activation of the olfactory system in mice.  Autism spectrum disorders (ASD) exhibit two clusters of core symptoms, i.e., social and communication impairment, and repetitive behaviors and sensory abnormalities. Our previous study demonstrated that TBR1, a causative gene of ASD, controls axonal projection and neuronal activation of amygdala and regulates social interaction and vocal communication in a mouse model. Behavioral defects caused by Tbr1 haploinsufficiency can be ameliorated by increasing neural activity via D-cycloserine treatment, an N-methyl-D-aspartate receptor (NMDAR) coagonist. In this report, we investigate the role of TBR1 in regulating olfaction and test whether D-cycloserine can also improve olfactory defects in Tbr1 mutant mice. We used Tbr1+/- mice as a model to investigate the function of TBR1 in olfactory sensation and discrimination of non-social odors. We employed a behavioral assay to characterize the olfactory defects of Tbr1+/- mice. Magnetic resonance imaging (MRI) and histological analysis were applied to characterize anatomical features. Immunostaining was performed to further analyze differences in expression of TBR1 subfamily members (namely TBR1, TBR2, and TBX21), interneuron populations, and dendritic abnormalities in olfactory bulbs. Finally, C-FOS staining was used to monitor neuronal activation of the olfactory system upon odor stimulation. Tbr1+/- mice exhibited smaller olfactory bulbs and anterior commissures, reduced interneuron populations, and an abnormal dendritic morphology of mitral cells in the olfactory bulbs. Tbr1 haploinsufficiency specifically impaired olfactory discrimination but not olfactory sensation. Neuronal activation upon odorant stimulation was reduced in the glomerular layer of Tbr1+/- olfactory bulbs. Furthermore, although the sizes of piriform and perirhinal cortices were not affected by Tbr1 deficiency, neuronal activation was reduced in these two cortical regions in response to odorant stimulation. These results suggest an impairment of neuronal activation in olfactory bulbs and defective connectivity from olfactory bulbs to the upper olfactory system in Tbr1+/- mice. Systemic administration of D-cycloserine, an NMDAR co-agonist, ameliorated olfactory discrimination in Tbr1+/- mice, suggesting that increased neuronal activity has a beneficial effect on Tbr1 deficiency. Tbr1 regulates neural circuits and activity in the olfactory system to control olfaction. Tbr1+/- mice can serve as a suitable model for revealing how an autism causative gene controls neuronal circuits, neural activity, and autism-related behaviors. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30318412  Title: Neonatal Tbr1 Dosage Controls Cortical Layer 6 Connectivity.  An understanding of how heterozygous loss-of-function mutations in autism spectrum disorder (ASD) risk genes, such as TBR1, contribute to ASD remains elusive. Conditional Tbr1 deletion during late mouse gestation in cortical layer 6 neurons (Tbr1layer6 mutants) provides novel insights into its function, including dendritic patterning, synaptogenesis, and cell-intrinsic physiology. These phenotypes occur in heterozygotes, providing insights into mechanisms that may underlie ASD pathophysiology. Restoring expression of Wnt7b largely rescues the synaptic deficit in Tbr1layer6 mutant neurons. Furthermore, Tbr1layer6 heterozygotes have increased anxiety-like behavior, a phenotype seen ASD. Integrating TBR1 chromatin immunoprecipitation sequencing (ChIP-seq) and RNA sequencing (RNA-seq) data from layer 6 neurons and activity of TBR1-bound candidate enhancers provides evidence for how TBR1 regulates layer 6 properties. Moreover, several putative TBR1 targets are ASD risk genes, placing TBR1 in a central position both for ASD risk and for regulating transcriptional circuits that control multiple steps in layer 6 development essential for the assembly of neural circuits. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28595950  Title: Luteolin attenuates interleukin-6-mediated astrogliosis in human iPSC-derived neural aggregates: A candidate preventive substance for maternal immune activation-induced abnormalities.  Maternal infection during pregnancy increases the risk of neurodevelopmental conditions such as autism spectrum disorders and schizophrenia in offspring. Several previous animal studies have indicated that maternal immune activation (MIA), rather than a specific pathogen, alters fetal brain development. Among them, prenatal exposure to interleukin-6 (IL-6) has been associated with behavioral and neuropathological abnormalities, though such findings remain to be elucidated in humans. We developed a human cell-based model of MIA by exposing human induced pluripotent stem cells (hiPSCs)-derived neural aggregates to IL-6 and investigated whether luteolin-a naturally occurring flavonoid found in edible plants-could prevent MIA-induced abnormalities. We generated neural aggregates from hiPSCs using the serum-free floating culture of embryoid body-like aggregates with quick reaggregation (SFEBq) method, following which aggregates were cultured in suspension. We then exposed the aggregates to IL-6 (100ng/ml) for 24h at day 51. Transient IL-6 exposure significantly increased the area ratio of astrocytes (GFAP-positive area ratio) and decreased the area ratio of early-born neurons (TBR1-positive or CTIP2-positive area ratio) relative to controls. In addition, western blot analysis revealed that levels of phosphorylated STAT3 were significantly elevated in IL-6-exposed neural aggregates. Luteolin treatment inhibited STAT3 phosphorylation and counteracted IL-6-mediated increases of GFAP-positive cells and reductions of TBR1-positive and CTIP2-positive cells. Our observations suggest that the flavonoid luteolin may attenuate or prevent MIA-induced neural abnormalities. As we observed increased apoptosis at high concentrations of luteolin, further studies are required to determine the optimal intake dosage and duration for pregnant women. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28234597  Title: Calcium/calmodulin-dependent serine protein kinase (CASK), a protein implicated in mental retardation and autism-spectrum disorders, interacts with T-Brain-1 (TBR1) to control extinction of associative memory in male mice.  Human genetic studies have indicated that mutations in calcium/calmodulin-dependent serine protein kinase (CASK) result in X-linked mental retardation and autism-spectrum disorders. We aimed to establish a mouse model to study how Cask regulates mental ability. Because Cask encodes a multidomain scaffold protein, a possible strategy to dissect how CASK regulates mental ability and cognition is to disrupt specific protein-protein interactions of CASK in vivo and then investigate the impact of individual specific protein interactions. Previous in vitro analyses indicated that a rat CASK T724A mutation reduces the interaction between CASK and T-brain-1 (TBR1) in transfected COS cells. Because TBR1 is critical for glutamate receptor, ionotropic, N-methyl-D-aspartate receptor subunit 2B (Grin2b) expression and is a causative gene for autism and intellectual disability, we then generated CASK T740A (corresponding to rat CASK T724A) mutant mice using a gene-targeting approach. Immunoblotting, coimmunoprecipitation, histological methods and behavioural assays (including home cage, open field, auditory and contextual fear conditioning and conditioned taste aversion) were applied to investigate expression of CASK and its related proteins, the protein-protein interactions of CASK, and anatomic and behavioural features of CASK T740A mice. The CASK T740A mutation attenuated the interaction between CASK and TBR1 in the brain. However, CASK T740A mice were generally healthy, without obvious defects in brain morphology. The most dramatic defect among the mutant mice was in extinction of associative memory, though acquisition was normal. The functions of other CASK protein interactions cannot be addressed using CASK T740A mice. Disruption of the CASK and TBR1 interaction impairs extinction, suggesting the involvement of CASK in cognitive flexibility. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28057268  Title: Control of Neuronal Development by T-Box Genes in the Brain.  T-box transcription factors play key roles in the regulation of developmental processes such as cell differentiation and migration. Mammals have 17 T-box genes, of which several regulate brain development. The Tbr1 subfamily of T-box genes is particularly important in development of the cerebral cortex, olfactory bulbs (OBs), and cerebellum. This subfamily is comprised of Tbr1, Tbr2 (also known as Eomes), and Tbx21. In developing cerebral cortex, Tbr2 and Tbr1 are expressed during successive stages of differentiation in the pyramidal neuron lineage, from Tbr2+ intermediate progenitors to Tbr1+ postmitotic glutamatergic neurons. At each stage, Tbr2 and Tbr1 regulate laminar and regional identity of cortical projection neurons, cell migration, and axon guidance. In the OB, Tbr1 subfamily genes regulate neurogenesis of mitral and tufted cells, and glutamatergic juxtaglomerular interneurons. Tbr2 is also prominent in the development of retinal ganglion cells in nonimage-forming pathways. Other regions that require Tbr2 or Tbr1 in development or adulthood include the cerebellum and adult dentate gyrus. In humans, de novo mutations in TBR1 are important causes of sporadic autism and intellectual disability. Further studies of T-box transcription factors will enhance our understanding of neurodevelopmental disorders and inform approaches to new therapies. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27325115  Title: TBR1 regulates autism risk genes in the developing neocortex.  Exome sequencing studies have identified multiple genes harboring de novo loss-of-function (LoF) variants in individuals with autism spectrum disorders (ASD), including TBR1, a master regulator of cortical development. We performed ChIP-seq for TBR1 during mouse cortical neurogenesis and show that TBR1-bound regions are enriched adjacent to ASD genes. ASD genes were also enriched among genes that are differentially expressed in Tbr1 knockouts, which together with the ChIP-seq data, suggests direct transcriptional regulation. Of the nine ASD genes examined, seven were misexpressed in the cortices of Tbr1 knockout mice, including six with increased expression in the deep cortical layers. ASD genes with adjacent cortical TBR1 ChIP-seq peaks also showed unusually low levels of LoF mutations in a reference human population and among Icelanders. We then leveraged TBR1 binding to identify an appealing subset of candidate ASD genes. Our findings highlight a TBR1-regulated network of ASD genes in the developing neocortex that are relatively intolerant to LoF mutations, indicating that these genes may play critical roles in normal cortical development. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/26578866  Title: Brain-specific transcriptional regulator T-brain-1 controls brain wiring and neuronal activity in autism spectrum disorders.  T-brain-1 (TBR1) is a brain-specific T-box transcription factor. In 1995, Tbr1 was first identified from a subtractive hybridization that compared mouse embryonic and adult telencephalons. Previous studies of Tbr1 (-∕-) mice have indicated critical roles for TBR1 in the development of the cerebral cortex, amygdala, and olfactory bulb. Neuronal migration and axonal projection are two important developmental features controlled by TBR1. Recently, recurrent de novo disruptive mutations in the TBR1 gene have been found in patients with autism spectrum disorders (ASDs). Human genetic studies have identified TBR1 as a high-confidence risk factor for ASDs. Because only one allele of the TBR1 gene is mutated in these patients, Tbr1 (+∕-) mice serve as a good genetic mouse model to explore the mechanism by which de novo TBR1 mutation leads to ASDs. Although neuronal migration and axonal projection defects of cerebral cortex are the most prominent phenotypes in Tbr1 (-∕-) mice, these features are not found in Tbr1 (+∕-) mice. Instead, inter- and intra-amygdalar axonal projections and NMDAR expression and activity in amygdala are particularly susceptible to Tbr1 haploinsufficiency. The studies indicated that both abnormal brain wiring (abnormal amygdalar connections) and excitation/inhibition imbalance (NMDAR hypoactivity), two prominent models for ASD etiology, are present in Tbr1 (+∕-) mice. Moreover, calcium/calmodulin-dependent serine protein kinase (CASK) was found to interact with TBR1. The CASK-TBR1 complex had been shown to directly bind the promoter of the Grin2b gene, which is also known as Nmdar2b, and upregulate Grin2b expression. This molecular function of TBR1 provides an explanation for NMDAR hypoactivity in Tbr1 (+∕-) mice. In addition to Grin2b, cell adhesion molecules-including Ntng1, Cdh8, and Cntn2-are also regulated by TBR1 to control axonal projections of amygdala. Taken together, the studies of Tbr1 provide an integrated picture of ASD etiology at the cellular and circuit levels. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25981743  Title: Trans-synaptic zinc mobilization improves social interaction in two mouse models of autism through NMDAR activation.  Genetic aspects of autism spectrum disorders (ASDs) have recently been extensively explored, but environmental influences that affect ASDs have received considerably less attention. Zinc (Zn) is a nutritional factor implicated in ASDs, but evidence for a strong association and linking mechanism is largely lacking. Here we report that trans-synaptic Zn mobilization rapidly rescues social interaction in two independent mouse models of ASD. In mice lacking Shank2, an excitatory postsynaptic scaffolding protein, postsynaptic Zn elevation induced by clioquinol (a Zn chelator and ionophore) improves social interaction. Postsynaptic Zn is mainly derived from presynaptic pools and activates NMDA receptors (NMDARs) through postsynaptic activation of the tyrosine kinase Src. Clioquinol also improves social interaction in mice haploinsufficient for the transcription factor Tbr1, which accompanies NMDAR activation in the amygdala. These results suggest that trans-synaptic Zn mobilization induced by clioquinol rescues social deficits in mouse models of ASD through postsynaptic Src and NMDAR activation. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25309323  Title: Neuronal excitation upregulates Tbr1, a high-confidence risk gene of autism, mediating Grin2b expression in the adult brain.  The activity-regulated gene expression of transcription factors is required for neural plasticity and function in response to neuronal stimulation. T-brain-1 (TBR1), a critical neuron-specific transcription factor for forebrain development, has been recognized as a high-confidence risk gene for autism spectrum disorders. Here, we show that in addition to its role in brain development, Tbr1 responds to neuronal activation and further modulates the Grin2b expression in adult brains and mature neurons. The expression levels of Tbr1 were investigated using both immunostaining and quantitative reverse transcription polymerase chain reaction (RT-PCR) analyses. We found that the mRNA and protein expression levels of Tbr1 are induced by excitatory synaptic transmission driven by bicuculline or glutamate treatment in cultured mature neurons. The upregulation of Tbr1 expression requires the activation of both α-amino-3-hydroxy-5-methyl-4-isoxazole-propionic acid (AMPA) and N-methyl-D-aspartate (NMDA) receptors. Furthermore, behavioral training triggers Tbr1 induction in the adult mouse brain. The elevation of Tbr1 expression is associated with Grin2b upregulation in both mature neurons and adult brains. Using Tbr1-deficient neurons, we further demonstrated that TBR1 is required for the induction of Grin2b upon neuronal activation. Taken together with the previous studies showing that TBR1 binds the Grin2b promoter and controls expression of luciferase reporter driven by Grin2b promoter, the evidence suggests that TBR1 directly controls Grin2b expression in mature neurons. We also found that the addition of the calcium/calmodulin-dependent protein kinase II (CaMKII) antagonist KN-93, but not the calcium-dependent phosphatase calcineurin antagonist cyclosporin A, to cultured mature neurons noticeably inhibited Tbr1 induction, indicating that neuronal activation upregulates Tbr1 expression in a CaMKII-dependent manner. In conclusion, our study suggests that Tbr1 plays an important role in adult mouse brains in response to neuronal activation to modulate the activity-regulated gene transcription required for neural plasticity. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/24441682  Title: Tbr1 haploinsufficiency impairs amygdalar axonal projections and results in cognitive abnormality.  The neuron-specific transcription factor T-box brain 1 (TBR1) regulates brain development. Disruptive mutations in the TBR1 gene have been repeatedly identified in patients with autism spectrum disorders (ASDs). Here, we show that Tbr1 haploinsufficiency results in defective axonal projections of amygdalar neurons and the impairment of social interaction, ultrasonic vocalization, associative memory and cognitive flexibility in mice. Loss of a copy of the Tbr1 gene altered the expression of Ntng1, Cntn2 and Cdh8 and reduced both inter- and intra-amygdalar connections. These developmental defects likely impair neuronal activation upon behavioral stimulation, which is indicated by fewer c-FOS-positive neurons and lack of GRIN2B induction in Tbr1(+/-) amygdalae. We also show that upregulation of amygdalar neuronal activity by local infusion of a partial NMDA receptor agonist, d-cycloserine, ameliorates the behavioral defects of Tbr1(+/-) mice. Our study suggests that TBR1 is important in the regulation of amygdalar axonal connections and cognition. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/23112752  Title: Investigation of TBR1 Hemizygosity: Four Individuals with 2q24 Microdeletions.  TBR1 encodes a transcription factor with critical roles in corticogenesis, including cortical neuron migration and axon pathfinding, establishment of regional and laminar identity of cortical neurons, and control of glutamatergic neuronal cell fate. Based upon TBR1's role in cortical development, we sought to investigate TBR1 hemizygosity in individuals referred for genetic evaluation of intellectual disability and developmental delay. We describe 4 patients with microdeletions identified by molecular cytogenetic techniques, encompassing TBR1 and spanning 2q24.1q31.1, ranging in size from 2.17 to 12.34 Mb. Only the patient with the largest deletion had a possible cortical malformation. Mild ventriculomegaly is the only common brain anomaly, present in all patients; a Chiari I malformation is seen in 2 patients, and mega cisterna magna is seen in a third. Our findings are consistent with Tbr1 mouse models showing that hemizygosity of the gene requires additional genetic factors for the manifestation of severe structural brain malformations. Other syndromic features are present in these patients, including autism spectrum disorders, ocular colobomas, and craniosynostosis, features that are likely affected by the deletion of genes other than TBR1. |
| TBR1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/19948250  Title: Autism susceptibility candidate 2 (Auts2) encodes a nuclear protein expressed in developing brain regions implicated in autism neuropathology.  Autism susceptibility candidate 2 (Auts2) is a gene associated with autism and mental retardation, whose function is unknown. Expression of Auts2 mRNA and protein were studied in the developing mouse brain by in situ hybridization, immunohistochemistry, and western blotting. Auts2 mRNA was highly expressed in the developing cerebral cortex and cerebellum, regions often affected by neuropathological changes in autism, and a few other brain regions. On embryonic day (E) 12, Auts2 mRNA was expressed in the cortical preplate, where it colocalized with Tbr1, a transcription factor specific for postmitotic projection neurons. From E16 to postnatal day 21, Auts2 was expressed most abundantly in frontal cortex, hippocampus and cerebellum, including Purkinje cells and deep nuclei. High levels of Auts2 were also detected in developing dorsal thalamus, olfactory bulb, inferior colliculus and substantia nigra. Auts2 protein showed similar regional expression patterns as the mRNA. At the cellular level, Auts2 protein was localized in the nuclei of neurons and some neuronal progenitors. |
| TCF4, TCF7L2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36782064  Title: TCF7L2 acts as a molecular switch in midbrain to control mammal vocalization through its DNA binding domain but not transcription activation domain.  Vocalization is an essential medium for social signaling in birds and mammals. Periaqueductal gray (PAG) a conserved midbrain structure is believed to be responsible for innate vocalizations, but its molecular regulation remains largely unknown. Here, through a mouse forward genetic screening we identified one of the key Wnt/β-catenin effectors TCF7L2/TCF4 controls ultrasonic vocalization (USV) production and syllable complexity during maternal deprivation and sexual encounter. Early developmental expression of TCF7L2 in PAG excitatory neurons is necessary for the complex trait, while TCF7L2 loss reduces neuronal gene expressions and synaptic transmission in PAG. TCF7L2-mediated vocal control is independent of its β-catenin-binding domain but dependent of its DNA binding ability. Patient mutations associated with developmental disorders, including autism spectrum disorders, disrupt the transcriptional repression effect of TCF7L2, while mice carrying those mutations display severe USV impairments. Therefore, we conclude that TCF7L2 orchestrates gene expression in midbrain to control vocal production through its DNA binding but not transcription activation domain. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36407762  Title: Expression of alternative transcription factor 4 mRNAs and protein isoforms in the developing and adult rodent and human tissues.  Transcription factor 4 (TCF4) belongs to the class I basic helix-loop-helix family of transcription factors (also known as E-proteins) and is vital for the development of the nervous system. Aberrations in the TCF4 gene are associated with several neurocognitive disorders such as schizophrenia, intellectual disability, post-traumatic stress disorder, depression, and Pitt-Hopkins Syndrome, a rare but severe autism spectrum disorder. Expression of the human TCF4 gene can produce at least 18 N-terminally distinct protein isoforms, which activate transcription with different activities and thus may vary in their function during development. We used long-read RNA-sequencing and western blot analysis combined with the analysis of publicly available short-read RNA-sequencing data to describe both the mRNA and protein expression of the many distinct TCF4 isoforms in rodent and human neural and nonneural tissues. We show that TCF4 mRNA and protein expression is much higher in the rodent brain compared to nonneural tissues. TCF4 protein expression is highest in the rodent cerebral cortex and hippocampus, where expression peaks around birth, and in the rodent cerebellum, where expression peaks about a week after birth. In human, highest TCF4 expression levels were seen in the developing brain, although some nonneural tissues displayed comparable expression levels to adult brain. In addition, we show for the first time that out of the many possible TCF4 isoforms, the main TCF4 isoforms expressed in the rodent and human brain and other tissues are TCF4-B, -C, -D, -A, and-I. Taken together, our isoform specific analysis of TCF4 expression in different tissues could be used for the generation of gene therapy applications for patients with TCF4-associated diseases. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36224259  Title: Evaluation of Nav1.8 as a therapeutic target for Pitt Hopkins Syndrome.  Pitt Hopkins Syndrome (PTHS) is a rare syndromic form of autism spectrum disorder (ASD) caused by autosomal dominant mutations in the Transcription Factor 4 (TCF4) gene. TCF4 is a basic helix-loop-helix transcription factor that is critical for neurodevelopment and brain function through its binding to cis-regulatory elements of target genes. One potential therapeutic strategy for PTHS is to identify dysregulated target genes and normalize their dysfunction. Here, we propose that SCN10A is an important target gene of TCF4 that is an applicable therapeutic approach for PTHS. Scn10a encodes the voltage-gated sodium channel Nav1.8 and is consistently shown to be upregulated in PTHS mouse models. In this perspective, we review prior literature and present novel data that suggests inhibiting Nav1.8 in PTHS mouse models is effective at normalizing neuron function, brain circuit activity and behavioral abnormalities and posit this therapeutic approach as a treatment for PTHS. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35501322  Title: Transcription Factor 4 loss-of-function is associated with deficits in progenitor proliferation and cortical neuron content.  Transcription Factor 4 (TCF4) has been associated with autism, schizophrenia, and other neuropsychiatric disorders. However, how pathological TCF4 mutations affect the human neural tissue is poorly understood. Here, we derive neural progenitor cells, neurons, and brain organoids from skin fibroblasts obtained from children with Pitt-Hopkins Syndrome carrying clinically relevant mutations in TCF4. We show that neural progenitors bearing these mutations have reduced proliferation and impaired capacity to differentiate into neurons. We identify a mechanism through which TCF4 loss-of-function leads to decreased Wnt signaling and then to diminished expression of SOX genes, culminating in reduced progenitor proliferation in vitro. Moreover, we show reduced cortical neuron content and impaired electrical activity in the patient-derived organoids, phenotypes that were rescued after correction of TCF4 expression or by pharmacological modulation of Wnt signaling. This work delineates pathological mechanisms in neural cells harboring TCF4 mutations and provides a potential target for therapeutic strategies for genetic disorders associated with this gene. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34780749  Title: Brain injury and inflammation genes common to a number of neurological diseases and the genes involved in the genesis of GABAnergic neurons are altered in monoamine oxidase B knockout mice.  Monoamine oxidase B (MAO B) oxidizes trace amine phenylethylamine (PEA), and neurotransmitters serotonin and dopamine in the brain. We reported previously that PEA levels increased significantly in all brain regions, but serotonin and dopamine levels were unchanged in MAO B knockout (KO) mice. PEA and dopamine are both synthesized from phenylalanine by aromatic L-amino acid decarboxylase in dopaminergic neurons in the striatum. A high concentration of PEA in the striatum may cause dopaminergic neuronal death in the absence of MAO B. We isolated the RNA from brain tissue of MAO B KO mice (2-month old) and age-matched wild type (WT) male mice and analyzed the altered genes by Affymetrix microarray. Differentially expressed genes (DEGs) in MAO B KO compared to WT mice were analyzed by Partek Genomics Suite, followed by Ingenuity Pathway Analysis (IPA) to assess their functional relationships. DEGs in MAO B KO mice are involved in brain inflammation and the genesis of GABAnergic neurons. The significant DEGs include four brain injury or inflammation genes (upregulated: Ido1, TSPO, AVP, Tdo2), five gamma-aminobutyric acid (GABA) receptors (down-regulated: GABRA2, GABRA3, GABRB1, GABRB3, GABRG3), five transcription factors related to adult neurogenesis (upregulated: Wnt7b, Hes5; down-regulated: Pax6, Tcf4, Dtna). Altered brain injury and inflammation genes in MAO B knockout mice are involved in various neurological disorders: attention deficit hyperactive disorder, panic disorder, obsessive compulsive disorder, autism, amyotrophic lateral sclerosis, Parkinson's diseases, Alzheimer's disease, bipolar affective disorder. Many were commonly involved in these disorders, indicating that there are overlapping molecular pathways. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34645823  Title: Disordered breathing in a Pitt-Hopkins syndrome model involves Phox2b-expressing parafacial neurons and aberrant Nav1.8 expression.  Pitt-Hopkins syndrome (PTHS) is a rare autism spectrum-like disorder characterized by intellectual disability, developmental delays, and breathing problems involving episodes of hyperventilation followed by apnea. PTHS is caused by functional haploinsufficiency of the gene encoding transcription factor 4 (Tcf4). Despite the severity of this disease, mechanisms contributing to PTHS behavioral abnormalities are not well understood. Here, we show that a Tcf4 truncation (Tcf4tr/+) mouse model of PTHS exhibits breathing problems similar to PTHS patients. This behavioral deficit is associated with selective loss of putative expiratory parafacial neurons and compromised function of neurons in the retrotrapezoid nucleus that regulate breathing in response to tissue CO2/H+. We also show that central Nav1.8 channels can be targeted pharmacologically to improve respiratory function at the cellular and behavioral levels in Tcf4tr/+ mice, thus establishing Nav1.8 as a high priority target with therapeutic potential in PTHS. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/33228529  Title: Enriched environment ameliorates adult hippocampal neurogenesis deficits in Tcf4 haploinsufficient mice.  Transcription factor 4 (TCF4) has been linked to human neurodevelopmental disorders such as intellectual disability, Pitt-Hopkins Syndrome (PTHS), autism, and schizophrenia. Recent work demonstrated that TCF4 participates in the control of a wide range of neurodevelopmental processes in mammalian nervous system development including neural precursor proliferation, timing of differentiation, migration, dendritogenesis and synapse formation. TCF4 is highly expressed in the adult hippocampal dentate gyrus - one of the few brain regions where neural stem / progenitor cells generate new functional neurons throughout life. We here investigated whether TCF4 haploinsufficiency, which in humans causes non-syndromic forms of intellectual disability and PTHS, affects adult hippocampal neurogenesis, a process that is essential for hippocampal plasticity in rodents and potentially in humans. Young adult Tcf4 heterozygote knockout mice showed a major reduction in the level of adult hippocampal neurogenesis, which was at least in part caused by lower stem/progenitor cell numbers and impaired maturation and survival of adult-generated neurons. Interestingly, housing in an enriched environment was sufficient to enhance maturation and survival of new neurons and to substantially augment neurogenesis levels in Tcf4 heterozygote knockout mice. The present findings indicate that haploinsufficiency for the intellectual disability- and PTHS-linked transcription factor TCF4 not only affects embryonic neurodevelopment but impedes neurogenesis in the hippocampus of adult mice. These findings suggest that TCF4 haploinsufficiency may have a negative impact on hippocampal function throughout adulthood by impeding hippocampal neurogenesis. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32971458  Title: Generation of 10 patient-specific induced pluripotent stem cells (iPSCs) to model Pitt-Hopkins Syndrome.  Autosomal dominant mutations in transcription factor 4 (TCF4) are associated with a rare syndromic form of Autism Spectrum Disorder (ASD) called Pitt-Hopkins Syndrome (PTHS). Here, we report the generation of a collection of induced pluripotent stem cells (iPSCs) from 5 patients diagnosed with PTHS and 5 familial controls. These patient-derived iPSCs contain a variety of mutations within the TCF4 gene, possess a normal karyotype and express all the appropriate pluripotent stem cell markers. These novel patient lines will be a useful resource for the research community to study PTHS and the function of TCF4. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32765228  Title: Region and Cell Type Distribution of TCF4 in the Postnatal Mouse Brain.  Transcription factor 4 is a class I basic helix-loop-helix transcription factor regulating gene expression. Altered TCF4 gene expression has been linked to non-syndromic intellectual disability, schizophrenia, and a severe neurodevelopmental disorder known as Pitt-Hopkins syndrome. An understanding of the cell types expressing TCF4 protein in the mouse brain is needed to help identify potential pathophysiological mechanisms and targets for therapeutic delivery in TCF4-linked disorders. Here we developed a novel green fluorescent protein reporter mouse to visualize TCF4-expressing cells throughout the brain. Using this TCF4 reporter mouse, we observed prominent expression of TCF4 in the pallial region and cerebellum of the postnatal brain. At the cellular level, both glutamatergic and GABAergic neurons express TCF4 in the cortex and hippocampus, while only a subset of GABAergic interneurons express TCF4 in the striatum. Among glial cell groups, TCF4 is present in astrocytes and immature and mature oligodendrocytes. In the cerebellum, cells in the granule and molecular layer express TCF4. Our findings greatly extend our knowledge of the spatiotemporal and cell type-specific expression patterns of TCF4 in the brain, and hence, lay the groundwork to better understand TCF4-linked neurological disorders. Any effort to restore TCF4 functions through small molecule or genetic therapies should target these brain regions and cell groups to best recapitulate TCF4 expression patterns. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32510763  Title: TCF20 dysfunction leads to cortical neurogenesis defects and autistic-like behaviors in mice.  Recently, de novo mutations of transcription factor 20 (TCF20) were found in patients with autism by large-scale exome sequencing. However, how TCF20 modulates brain development and whether its dysfunction causes ASD remain unclear. Here, we show that TCF20 deficits impair neurogenesis in mouse. TCF20 deletion significantly reduces the number of neurons, which leads to abnormal brain functions. Furthermore, transcriptome analysis and ChIP-qPCR reveal that the DNA demethylation factor TDG is a downstream target gene of TCF20. As a nonspecific DNA demethylation factor, TDG potentially affects many genes. Combined TDG ChIP-seq and GO analysis of TCF20 RNA-Seq identifies T-cell factor 4 (TCF-4) as a common target. TDG controls the DNA methylation level in the promoter area of TCF-4, affecting TCF-4 expression and modulating neural differentiation. Overexpression of TDG or TCF-4 rescues the deficient neurogenesis of TCF20 knockdown brains. Together, our data reveal that TCF20 is essential for neurogenesis and we suggest that defects in neurogenesis caused by TCF20 loss are associated with ASD. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32474139  Title: Tcf4 is required for correct brain development during embryogenesis.  Tcf4 has been linked to autism, schizophrenia, and Pitt-Hopkins Syndrome (PTHS) in humans, suggesting a role for Tcf4 in brain development and importantly cortical development. However, the mechanisms behind its role in disease and brain development are still elusive. We provide evidence that Tcf4 has a critical function in the differentiation of cortical regions, corpus callosum and anterior commissure formation, and development of the hippocampus during murine embryonic development. In the present study, we show that Tcf4 is expressed throughout the developing brain at the peak of neurogenesis. Deletion of Tcf4 results in mis-specification of the cortical neurons, malformation of the corpus callosum and anterior commissure, and hypoplasia of the hippocampus. Furthermore, the Tcf4 mutant shows an absence of midline remodeling, underlined by the loss of GFAP-expressing midline glia in the indusium griseum and callosal wedge and midline zipper glia in the telencephalic midline. RNA-sequencing on E14.5 cortex material shows that Tcf4 functions as a transcriptional activator and loss of Tcf4 results in downregulation of genes linked to neurogenesis and neuronal maturation. Furthermore, many genes that are differentially expressed after Tcf4 ablation are linked to other neurodevelopmental disorders. Taken together, we show that correct brain development and neuronal differentiation are severely affected in Tcf4 mutants, phenocopying morphological brain defects detected in PTHS patients. The presented data identifies new leads to understand the mechanisms behind brain and specifically cortical development and can provide novel insights in developmental mechanisms underlying human brain defects. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/32266943  Title: Transcription factor Tcf4 is the preferred heterodimerization partner for Olig2 in oligodendrocytes and required for differentiation.  Development of oligodendrocytes and myelin formation in the vertebrate central nervous system is under control of several basic helix-loop-helix transcription factors such as Olig2, Ascl1, Hes5 and the Id proteins. The class I basic helix-loop-helix proteins Tcf3, Tcf4 and Tcf12 represent potential heterodimerization partners and functional modulators for all, but have not been investigated in oligodendrocytes so far. Using mouse mutants, organotypic slice and primary cell cultures we here show that Tcf4 is required in a cell-autonomous manner for proper terminal differentiation and myelination in vivo and ex vivo. Partial compensation is provided by the paralogous Tcf3, but not Tcf12. On the mechanistic level Tcf4 was identified as the preferred heterodimerization partner of the central regulator of oligodendrocyte development Olig2. Both genetic studies in the mouse as well as functional studies on enhancer regions of myelin genes confirmed the relevance of this physical interaction for oligodendrocyte differentiation. Considering that alterations in TCF4 are associated with syndromic and non-syndromic forms of intellectual disability, schizophrenia and autism in humans, our findings point to the possibility of an oligodendroglial contribution to these disorders. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31081034  Title: Structural basis for preferential binding of human TCF4 to DNA containing 5-carboxylcytosine.  The psychiatric risk-associated transcription factor 4 (TCF4) is linked to schizophrenia. Rare TCF4 coding variants are found in individuals with Pitt-Hopkins syndrome-an intellectual disability and autism spectrum disorder. TCF4 contains a C-terminal basic-helix-loop-helix (bHLH) DNA binding domain which recognizes the enhancer-box (E-box) element 5'-CANNTG-3' (where N = any nucleotide). A subset of the TCF4-occupancy sites have the expanded consensus binding specificity 5'-C(A/G)-CANNTG-3', with an added outer Cp(A/G) dinucleotide; for example in the promoter for CNIH3, a gene involved in opioid dependence. In mammalian genomes, particularly brain, the CpG and CpA dinucleotides can be methylated at the 5-position of cytosine (5mC), and then may undergo successive oxidations to the 5-hydroxymethyl (5hmC), 5-formyl (5fC), and 5-carboxyl (5caC) forms. We find that, in the context of 5'-0CG-1CA-2CG-3TG-3'(where the numbers indicate successive dinucleotides), modification of the central E-box 2CG has very little effect on TCF4 binding, E-box 1CA modification has a negative influence on binding, while modification of the flanking 0CG, particularly carboxylation, has a strong positive impact on TCF4 binding to DNA. Crystallization of TCF4 in complex with unmodified or 5caC-modified oligonucleotides revealed that the basic region of bHLH domain adopts multiple conformations, including an extended loop going through the DNA minor groove, or the N-terminal portion of a long helix binding in the DNA major groove. The different protein conformations enable arginine 576 (R576) to interact, respectively, with a thymine in the minor groove, a phosphate group of DNA backbone, or 5caC in the major groove. The Pitt-Hopkins syndrome mutations affect five arginine residues in the basic region, two of them (R569 and R576) involved in 5caC recognition. Our analyses indicate, and suggest a structural basis for, the preferential recognition of 5caC by a transcription factor centrally important in brain development. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30375316  Title: Pitt-Hopkins Syndrome: A Unique Case Study.  Pitt-Hopkins syndrome (PTHS) is a rare genetic disorder caused by insufficient expression of the TCF4 gene. Most cases are characterized by severe intellectual disability, absent speech, motor delays, and autism spectrum disorder. Many have abnormal brain imaging, dysmorphic facial features, and medical comorbidities: myopia, constipation, epilepsy, and apneic spells. The present case study expands existing understanding of this disorder by presenting a unique phenotype with higher cognitive abilities and fewer medical comorbidities. The present case study reports on a 13-year-old, Caucasian male with a recent diagnosis of PTHS following genetic testing (i.e., whole exome sequencing). He was referred for a neuropsychological evaluation to document his neurocognitive functioning to assist with intervention planning. Evaluation of intellectual, attention/executive, memory, visual-motor/fine-motor, academic, adaptive, and emotional/behavioral functioning revealed global impairment across all areas of functioning. However, he demonstrated abilities beyond what has been detailed in the literature, including use of full sentences, capacity to learn and solve novel problems, basic academic functioning, and independent ambulation. Children with PTHS may demonstrate a spectrum of abilities beyond what has been documented in the literature thus far. Failure to recognize this spectrum can result in late identification of an accurate diagnosis. (JINS, 2018, 24, 995-1002). |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29228394  Title: The Psychiatric Risk Gene Transcription Factor 4 (TCF4) Regulates Neurodevelopmental Pathways Associated With Schizophrenia, Autism, and Intellectual Disability.  Common genetic variants in and around the gene encoding transcription factor 4 (TCF4) are associated with an increased risk of schizophrenia. Conversely, rare damaging TCF4 mutations cause Pitt-Hopkins syndrome and have also been found in individuals with intellectual disability (ID) and autism spectrum disorder (ASD). Chromatin immunoprecipitation and next generation sequencing were used to identify the genomic targets of TCF4. These data were integrated with expression, epigenetic and disease gene sets using a range of computational tools. We identify 10604 TCF4 binding sites in the genome that were assigned to 5437 genes. De novo motif enrichment found that most TCF4 binding sites contained at least one E-box (5'-CAtcTG). Approximately 77% of TCF4 binding sites overlapped with the H3K27ac histone modification for active enhancers. Enrichment analysis on the set of TCF4 targets identified numerous, highly significant functional clusters for pathways including nervous system development, ion transport and signal transduction, and co-expression modules for genes associated with synaptic function and brain development. Importantly, we found that genes harboring de novo mutations in schizophrenia (P = 5.3 × 10-7), ASD (P = 2.5 × 10-4), and ID (P = 7.6 × 10-3) were also enriched among TCF4 targets. TCF4 binding sites were also found at other schizophrenia risk loci including the nicotinic acetylcholine receptor cluster, CHRNA5/CHRNA3/CHRNB4 and SETD1A. These data demonstrate that TCF4 binding sites are found in a large number of neuronal genes that include many genetic risk factors for common neurodevelopmental disorders. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/29222403  Title: Common Pathophysiology in Multiple Mouse Models of Pitt-Hopkins Syndrome.  Mutations or deletions of the transcription factor TCF4 are linked to Pitt-Hopkins syndrome (PTHS) and schizophrenia, suggesting that the precise pathogenic mutations dictate cellular, synaptic, and behavioral consequences. Here, we generated two novel mouse models of PTHS, one that mimics the most common pathogenic TCF4 point mutation (human R580W, mouse R579W) and one that deletes three pathogenic arginines, and explored phenotypes of these lines alongside models of pan-cellular or CNS-specific heterozygous Tcf4 disruption. We used mice of both sexes to show that impaired Tcf4 function results in consistent microcephaly, hyperactivity, reduced anxiety, and deficient spatial learning. All four PTHS mouse models demonstrated exaggerated hippocampal long-term potentiation (LTP), consistent with deficits in hippocampus-mediated behaviors. We further examined R579W mutant mice and mice with pan-cellular Tcf4 heterozygosity and found that they exhibited hippocampal NMDA receptor hyperfunction, which likely drives the enhanced LTP. Together, our data pinpoint convergent neurobiological features in PTHS mouse models and provide a foundation for preclinical studies and a rationale for testing whether NMDAR antagonists might be used to treat PTHS.SIGNIFICANCE STATEMENT Pitt-Hopkins syndrome (PTHS) is a rare neurodevelopmental disorder associated with TCF4 mutations/deletions. Despite this genetic insight, there is a need to identify the function of TCF4 in the brain. Toward this goal, we developed two mouse lines, including one harboring the most prevalent pathogenic point mutation, and compared them with two existing models that conditionally delete Tcf4 Our data identify a set of overlapping phenotypes that may serve as outcome measures for preclinical studies of PTHS treatments. We also discovered penetrant enhanced synaptic plasticity across mouse models that may be linked to increased NMDA receptor function. These data reveal convergent neurobiological characteristics of PTHS mouse models and support the further investigation of NMDA receptor antagonists as a possible PTHS treatment. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28733602  Title: Distinct selective forces and Neanderthal introgression shaped genetic diversity at genes involved in neurodevelopmental disorders.  In addition to high intelligence, humans evolved specialized social-cognitive skills, which are specifically affected in children with autism spectrum disorder (ASD). Genes affected in ASD represent suitable candidates to study the evolution of human social cognition. We performed an evolutionary analysis on 68 genes associated to neurodevelopmental disorders; our data indicate that genetic diversity was shaped by distinct selective forces, including natural selection and introgression from archaic hominins. We discuss the possibility that segregation distortion during spermatogenesis accounts for a subset of ASD mutations. Finally, we detected modern-human-specific alleles in DYRK1A and TCF4. These variants are located within regions that display chromatin features typical of transcriptional enhancers in several brain areas, strongly suggesting a regulatory role. These SNPs thus represent candidates for association with neurodevelopmental disorders, and await experimental validation in future studies. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28289282  Title: The schizophrenia- and autism-associated gene, transcription factor 4 regulates the columnar distribution of layer 2/3 prefrontal pyramidal neurons in an activity-dependent manner.  Disruption of the laminar and columnar organization of the brain is implicated in several psychiatric disorders. Here, we show in utero gain-of-function of the psychiatric risk gene transcription factor 4 (TCF4) severely disrupts the columnar organization of medial prefrontal cortex (mPFC) in a transcription- and activity-dependent manner. This morphological phenotype was rescued by co-expression of TCF4 plus calmodulin in a calcium-dependent manner and by dampening neuronal excitability through co-expression of an inwardly rectifying potassium channel (Kir2.1). For we believe the first time, we show that N-methyl-d-aspartate (NMDA) receptor-dependent Ca2+ transients are instructive to minicolumn organization because Crispr/Cas9-mediated mutation of NMDA receptors rescued TCF4-dependent morphological phenotypes. Furthermore, we demonstrate that the transcriptional regulation by the psychiatric risk gene TCF4 enhances NMDA receptor-dependent early network oscillations. Our novel findings indicate that TCF4-dependent transcription directs the proper formation of prefrontal cortical minicolumns by regulating the expression of genes involved in early spontaneous neuronal activity, and thus our results provides insights into potential pathophysiological mechanisms of TCF4-associated psychiatric disorders. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/30775158  Title: Molecular Mechanisms of Transcription Factor 4 in Pitt Hopkins Syndrome.  Pitt Hopkins syndrome (PTHS) is a rare neurodevelopmental disorder that results from mutations of the clinically pleiotropic Transcription Factor 4 (TCF4) gene. Mutations in the genomic locus of TCF4 on chromosome 18 have been linked to multiple disorders including 18q syndrome, schizophrenia, Fuch's corneal dystrophy, and sclerosing cholangitis. For PTHS, TCF4 mutation or deletion leads to the production of a dominant negative TCF4 protein and/or haploinsufficiency that results in abnormal brain development. The biology of TCF4 has been studied for several years in regards to its role in immune cell differentiation, although its role in neurodevelopment and the mechanisms resulting in the severe symptoms of PTHS are not well studied. Here, we summarize the current understanding of PTHS and recent findings that have begun to describe the biological implications of TCF4 deficiency during brain development and into adulthood. In particular, we focus on recent work that has looked at the role of TCF4 biology within the context of PTHS and highlight the potential for identification of therapeutic targets for PTHS. PTHS research continues to uncover mutations in TCF4 that underlie the genetic cause of this rare disease, and emerging evidence for molecular mechanisms that TCF4 regulates in brain development and neuronal function is contributing to a more complete picture of how pathology arises from this genetic basis, with important implications for the potential of future clinical care. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/28032012  Title: Neurodevelopmental models of transcription factor 4 deficiency converge on a common ion channel as a potential therapeutic target for Pitt Hopkins syndrome.  The clinically pleiotropic gene, Transcription Factor 4 (TCF4), is a broadly expressed basic helix-loop-helix (bHLH) transcription factor linked to multiple neurodevelopmental disorders, including schizophrenia, 18q deletion syndrome, and Pitt Hopkins syndrome (PTHS). In vivo suppression of Tcf4 by shRNA or mutation by CRISPR/Cas9 in the developing rat prefrontal cortex resulted in attenuated action potential output. To explain this intrinsic excitability deficit, we demonstrated that haploinsufficiency of TCF4 lead to the ectopic expression of two ion channels, Scn10a and Kcnq1. These targets of TCF4 regulation were identified through molecular profiling experiments that used translating ribosome affinity purification to enrich mRNA from genetically manipulated neurons. Using a mouse model of PTHS (Tcf4+/tr), we observed a similar intrinsic excitability deficit, however the underlying mechanism appeared slightly different than our rat model - as Scn10a expression was similarly increased but Kcnq1 expression was decreased. Here, we show that the truncated TCF4 protein expressed in our PTHS mouse model binds to wild-type TCF4 protein, and we suggest the difference in Kcnq1 expression levels between these two rodent models appears to be explained by a dominant-negative function of the truncated TCF4 protein. Despite the differences in the underlying molecular mechanisms, we observed common underlying intrinsic excitability deficits that are consistent with ectopic expression of Scn10a. The converging molecular function of TCF4 across two independent rodent models indicates SCN10a is a potential therapeutic target for Pitt-Hopkins syndrome. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/27265882  Title: Prostaglandin E2 promotes neural proliferation and differentiation and regulates Wnt target gene expression.  Prostaglandin E2 (PGE2 ) is an endogenous lipid molecule that regulates important physiological functions, including calcium signaling, neuronal plasticity, and immune responses. Exogenous factors such as diet, exposure to immunological agents, toxic chemicals, and drugs can influence PGE2 levels in the developing brain and have been associated with autism disorders. This study seeks to determine whether changes in PGE2 level can alter the behavior of undifferentiated and differentiating neuroectodermal (NE-4C) stem cells and whether PGE2 signaling impinges on the Wnt/β-catenin pathways. We show that PGE2 increases proliferation of undifferentiated NE-4C stem cells. PGE2 also promotes the progression of NE-4C stem cell differentiation into neuronal-lineage cells, which is apparent by accelerated appearance of neuronal clusters (neurospheres) and earlier expression of the neuronal marker microtubule-associated protein tau. Furthermore, PGE2 alters the expression of downstream Wnt-regulated genes previously associated with neurodevelopmental disorders. In undifferentiated stem cells, PGE2 downregulates Ptgs2 expression and upregulates Mmp9 and Ccnd1 expression. In differentiating neuronal cells, PGE2 causes upregulation of Wnt3, Tcf4, and Ccnd1. The convergence of the PGE2 and the Wnt pathways is also apparent through increased expression of active β-catenin, a key signaling component of the Wnt/β-catenin pathways. This study provides novel evidence that PGE2 influences progression of neuronal development and influences Wnt target gene expression. We discuss how these findings could have potential implications for neurodevelopmental disorders such as autism. © 2016 Wiley Periodicals, Inc. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25728630  Title: Pitt-Hopkins Mouse Model has Altered Particular Gastrointestinal Transits In Vivo.  Pitt-Hopkins syndrome (PTHS) is a neurodevelopmental disorder, classified as an autism spectrum disorder that is caused by the haploinsufficiency of Transcription Factor 4 (TCF4). The most common non-neurological symptoms in PTHS patients are gastrointestinal (GI) disturbances, mainly gastroesophageal reflux and severe constipation (in about 30 and 75% of PTHS patients, respectively). We hypothesized that the recently recognized mouse model of PTHS will exhibit problems with their gut function. We conducted series of in vivo tests on 15- to 19- week old male mice, heterozygous for the TCF4 functional deletion, mimicking the TCF4 haploinsufficiency in PTHS patients, and their wild type littermates. Data collection and initial analysis were performed blindly, that is, the genotyping key was received after the mean values were calculated for each individual animal, and then mean/median of each group was subsequently calculated. Body weight, fecal pellet output, and fluid content were similar between the groups, indicating normal gross growth of PTHS mice and their overall physiological GI motility and intestinal secretion/absorption. There were no significant differences in gut length and gross appearance pointing out that PTHS mice have normal gut in gross anatomical terms. However, the assessment of gut transit indicates that, while whole-gut transit velocity was similar between the groups, the upper GI and distal colon transit velocities were significantly reduced in the PTHS mice. This is the first evidence of specific gut related problems in the PTHS mice. Our study also validates the TCF4 functional knockout mice as an animal model to study PTHS-associated GI disturbances. |
| TCF4 |
| Url: https://pubmed.ncbi.nlm.nih.gov/22584867  Title: Convergent functional genomics of schizophrenia: from comprehensive understanding to genetic risk prediction.  We have used a translational convergent functional genomics (CFG) approach to identify and prioritize genes involved in schizophrenia, by gene-level integration of genome-wide association study data with other genetic and gene expression studies in humans and animal models. Using this polyevidence scoring and pathway analyses, we identify top genes (DISC1, TCF4, MBP, MOBP, NCAM1, NRCAM, NDUFV2, RAB18, as well as ADCYAP1, BDNF, CNR1, COMT, DRD2, DTNBP1, GAD1, GRIA1, GRIN2B, HTR2A, NRG1, RELN, SNAP-25, TNIK), brain development, myelination, cell adhesion, glutamate receptor signaling, G-protein-coupled receptor signaling and cAMP-mediated signaling as key to pathophysiology and as targets for therapeutic intervention. Overall, the data are consistent with a model of disrupted connectivity in schizophrenia, resulting from the effects of neurodevelopmental environmental stress on a background of genetic vulnerability. In addition, we show how the top candidate genes identified by CFG can be used to generate a genetic risk prediction score (GRPS) to aid schizophrenia diagnostics, with predictive ability in independent cohorts. The GRPS also differentiates classic age of onset schizophrenia from early onset and late-onset disease. We also show, in three independent cohorts, two European American and one African American, increasing overlap, reproducibility and consistency of findings from single-nucleotide polymorphisms to genes, then genes prioritized by CFG, and ultimately at the level of biological pathways and mechanisms. Finally, we compared our top candidate genes for schizophrenia from this analysis with top candidate genes for bipolar disorder and anxiety disorders from previous CFG analyses conducted by us, as well as findings from the fields of autism and Alzheimer. Overall, our work maps the genomic and biological landscape for schizophrenia, providing leads towards a better understanding of illness, diagnostics and therapeutics. It also reveals the significant genetic overlap with other major psychiatric disorder domains, suggesting the need for improved nosology. |
| TCF7L2 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36738649  Title: Shared genetics of psychiatric disorders and type 2 diabetes:a large-scale genome-wide cross-trait analysis.  Individuals with psychiatric disorders have elevated rates of type 2 diabetes comorbidity. Although little is known about the shared genetics and causality of this association. Thus, we aimed to investigate shared genetics and causal link between different type 2 diabetes and psychiatric disorders. We conducted a large-scale genome-wide cross-trait association study(GWAS) to investigate genetic overlap between type 2 diabetes and anorexia nervosa, attention deficit/hyperactivity disorder, autism spectrum disorder, bipolar disorder, major depressive disorder, obsessive-compulsive disorder, schizophrenia, anxiety disorders and Tourette syndrome. By post-GWAS functional analysis, we identify variants genes expression in various tissues. Enrichment pathways, potential protein interaction and mendelian randomization also provided to research the relationship between type 2 diabetes and psychiatric disorders. We discovered that type 2 diabetes and psychiatric disorders had a significant correlation. We identified 138 related loci, 32 were novel loci. Post-GWAS analysis revealed that 86 differentially expressed genes were located in different brain regions and peripheral blood in type 2 diabetes and related psychiatric disorders. MAPK signaling pathway plays an important role in neural development and insulin signaling. In addition, there is a causal relationship between T2D and mental disorders. In PPI analysis, the central genes of the DEG PPI network were FTO and TCF7L2. This large-scale genome-wide cross-trait analysis identified shared genetics andpotential causal links between type 2 diabetes and related psychiatric disorders, suggesting potential new biological functions in common among them. |
| TSHZ3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35292625  Title: Targeted Tshz3 deletion in corticostriatal circuit components segregates core autistic behaviors.  We previously linked TSHZ3 haploinsufficiency to autism spectrum disorder (ASD) and showed that embryonic or postnatal Tshz3 deletion in mice results in behavioral traits relevant to the two core domains of ASD, namely social interaction deficits and repetitive behaviors. Here, we provide evidence that cortical projection neurons (CPNs) and striatal cholinergic interneurons (SCINs) are two main and complementary players in the TSHZ3-linked ASD syndrome. In the cerebral cortex, TSHZ3 is expressed in CPNs and in a proportion of GABAergic interneurons, but not in cholinergic interneurons or glial cells. In the striatum, TSHZ3 is expressed in all SCINs, while its expression is absent or partial in the other main brain cholinergic systems. We then characterized two new conditional knockout (cKO) models generated by crossing Tshz3flox/flox with Emx1-Cre (Emx1-cKO) or Chat-Cre (Chat-cKO) mice to decipher the respective role of CPNs and SCINs. Emx1-cKO mice show altered excitatory synaptic transmission onto CPNs and impaired plasticity at corticostriatal synapses, with neither cortical neuron loss nor abnormal layer distribution. These animals present social interaction deficits but no repetitive patterns of behavior. Chat-cKO mice exhibit no loss of SCINs but changes in the electrophysiological properties of these interneurons, associated with repetitive patterns of behavior without social interaction deficits. Therefore, dysfunction in either CPNs or SCINs segregates with a distinct ASD behavioral trait. These findings provide novel insights onto the implication of the corticostriatal circuitry in ASD by revealing an unexpected neuronal dichotomy in the biological background of the two core behavioral domains of this disorder. |
| TSHZ3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/34919690  Title: Haploinsufficiency of the mouse Tshz3 gene leads to kidney defects.  Renal tract defects and autism spectrum disorder (ASD) deficits represent the phenotypic core of the 19q12 deletion syndrome caused by the loss of one copy of the TSHZ3 gene. Although a proportion of Tshz3 heterozygous (Tshz3+/lacZ) mice display ureteral defects, no kidney defects have been reported in these mice. The purpose of this study was to characterize the expression of Tshz3 in adult kidney as well as the renal consequences of embryonic haploinsufficiency of Tshz3 by analyzing the morphology and function of Tshz3 heterozygous adult kidney. Here, we described Tshz3 expression in the smooth muscle and stromal cells lining the renal pelvis, the papilla and glomerular endothelial cells (GEnCs) of the adult kidney as well as in the proximal nephron tubules in neonatal mice. Histological analysis showed that Tshz3+/lacZ adult kidney had an average of 29% fewer glomeruli than wild-type kidney. Transmission electron microscopy of Tshz3+/lacZ glomeruli revealed a reduced thickness of the glomerular basement membrane and a larger foot process width. Compared to wild type, Tshz3+/lacZ mice showed lower blood urea, phosphates, magnesium and potassium at 2 months of age. At the molecular level, transcriptome analysis identified differentially expressed genes related to inflammatory processes in Tshz3+/lacZ compare to wild-type (control) adult kidneys. Lastly, analysis of the urinary peptidome revealed 33 peptides associated with Tshz3+/lacZ adult mice. These results provide the first evidence that in the mouse Tshz3 haploinsufficiency leads to cellular, molecular and functional abnormalities in the adult mouse kidney. |
| TSHZ3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31542842  Title: Construct Validity and Cross Validity of a Test Battery Modeling Autism Spectrum Disorder (ASD) in Mice.  Modeling in other organism species is one of the crucial stages in ascertaining the association between gene and psychiatric disorder. Testing Autism Spectrum Disorder (ASD) in mice is very popular but construct validity of the batteries is not available. We presented here the first factor analysis of a behavioral model of ASD-like in mice coupled with empirical validation. We defined fourteen measures aligning mouse-behavior measures with the criteria defined by DSM-5 for the diagnostic of ASD. Sixty-five mice belonging to a heterogeneous pool of genotypes were tested. Reliability coefficients vary from .68 to .81. The factor analysis resulted in a three- factor solution in line with DSM criteria: social behavior, stereotypy and narrowness of the field of interest. The empirical validation with mice sharing a haplo-insufficiency of the zinc-finger transcription factor TSHZ3/Tshz3 associated with ASD shows the discriminant power of the highly loaded items. |
| TSHZ3 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31060802  Title: Postnatal Tshz3 Deletion Drives Altered Corticostriatal Function and Autism Spectrum Disorder-like Behavior.  Heterozygous deletion of the TSHZ3 gene, encoding for the teashirt zinc-finger homeobox family member 3 (TSHZ3) transcription factor that is highly expressed in cortical projection neurons (CPNs), has been linked to an autism spectrum disorder (ASD) syndrome. Similarly, mice with Tshz3 haploinsufficiency show ASD-like behavior, paralleled by molecular changes in CPNs and corticostriatal synaptic dysfunctions. Here, we aimed at gaining more insight into "when" and "where" TSHZ3 is required for the proper development of the brain, and its deficiency crucial for developing this ASD syndrome. We generated and characterized a novel mouse model of conditional Tshz3 deletion, obtained by crossing Tshz3flox/flox with CaMKIIalpha-Cre mice, in which Tshz3 is deleted in CPNs from postnatal day 2 to 3 onward. We characterized these mice by a multilevel approach combining genetics, cell biology, electrophysiology, behavioral testing, and bioinformatics. These conditional Tshz3 knockout mice exhibit altered cortical expression of more than 1000 genes, ∼50% of which have their human orthologue involved in ASD, in particular genes encoding for glutamatergic synapse components. Consistently, we detected electrophysiological and synaptic changes in CPNs and impaired corticostriatal transmission and plasticity. Furthermore, these mice showed strong ASD-like behavioral deficits. Our study reveals a crucial postnatal role of TSHZ3 in the development and functioning of the corticostriatal circuitry and provides evidence that dysfunction in these circuits might be determinant for ASD pathogenesis. Our conditional Tshz3 knockout mouse constitutes a novel ASD model, opening the possibility for an early postnatal therapeutic window for the syndrome linked to TSHZ3 haploinsufficiency. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/36855792  Title: Autism-like behavior of murine offspring induced by prenatal exposure to progestin is associated with gastrointestinal dysfunction due to claudin-1 suppression.  Autism spectrum disorders (ASD) are associated with the contribution of many prenatal risk factors; in particular, the sex hormone progestin and vitamin D receptor (VDR) are associated with gastrointestinal (GI) symptoms in ASD development, although the related mechanism remains unclear. We investigated the possible role and mechanism of progestin 17-hydroxyprogesterone caproate (17-OHPC) exposure-induced GI dysfunction and autism-like behaviours (ALB) in mouse offspring. An intestine-specific VDR-deficient mouse model was established for prenatal treatment, while transplantation of haematopoietic stem cells (HSCT) with related gene manipulation was used for postnatal treatment for 17-OHPC exposure-induced GI dysfunction and ALB in mouse offspring. The in vivo mouse experiments found that VDR deficiency mimics prenatal 17-OHPC exposure-mediated GI dysfunction, but has no effect on 17-OHPC-mediated autism-like behaviours (ALB) in mouse offspring. Furthermore, prenatal 17-OHPC exposure induces CLDN1 suppression in intestine epithelial cells, and transplantation of HSCT with CLDN1 expression ameliorates prenatal 17-OHPC exposure-mediated GI dysfunction, but has no effect on 17-OHPC-mediated ALB in offspring. In conclusion, prenatal 17-OHPC exposure triggers GI dysfunction in autism-like mouse offspring via CLDN1 suppression, providing a possible explanation for the involvement of CLDN1 and VDR in prenatal 17-OHPC exposure-mediated GI dysfunction with ASD. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/34835929  Title: Role of Vitamin D in Cognitive Dysfunction: New Molecular Concepts and Discrepancies between Animal and Human Findings.  increasing evidence suggests that besides the several metabolic, endocrine, and immune functions of 1alpha,25-dihydroxyvitamin D (1,25(OH)2D), the neuronal effects of 1,25(OH)2D should also be considered an essential contributor to the development of cognition in the early years and its maintenance in aging. The developmental disabilities induced by vitamin D deficiency (VDD) include neurological disorders (e.g., attention deficit hyperactivity disorder, autism spectrum disorder, schizophrenia) characterized by cognitive dysfunction. On the other hand, VDD has frequently been associated with dementia of aging and neurodegenerative diseases (e.g., Alzheimer's, Parkinson's disease). various cells (i.e., neurons, astrocytes, and microglia) within the central nervous system (CNS) express vitamin D receptors (VDR). Moreover, some of them are capable of synthesizing and catabolizing 1,25(OH)2D via 25-hydroxyvitamin D 1alpha-hydroxylase (CYP27B1) and 25-hydroxyvitamin D 24-hydroxylase (CYP24A1) enzymes, respectively. Both 1,25(OH)2D and 25-hydroxyvitamin D were determined from different areas of the brain and their uneven distribution suggests that vitamin D signaling might have a paracrine or autocrine nature in the CNS. Although both cholecalciferol and 25-hydroxyvitamin D pass the blood-brain barrier, the influence of supplementation has not yet demonstrated to have a direct impact on neuronal functions. So, this review summarizes the existing evidence for the action of vitamin D on cognitive function in animal models and humans and discusses the possible pitfalls of therapeutic clinical translation. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/33368246  Title: Defining vitamin D receptor expression in the brain using a novel VDRCre mouse.  Vitamin D action has been linked to several diseases regulated by the brain including obesity, diabetes, autism, and Parkinson's. However, the location of the vitamin D receptor (VDR) in the brain is not clear due to conflicting reports. We found that two antibodies previously published as specific in peripheral tissues are not specific in the brain. We thus created a new knockin mouse with cre recombinase expression under the control of the endogenous VDR promoter (VDRCre ). We demonstrated that the cre activity in the VDRCre mouse brain (as reported by a cre-dependent tdTomato expression) is highly overlapping with endogenous VDR mRNAs. These VDR-expressing cells were enriched in multiple brain regions including the cortex, amygdala, caudate putamen, and hypothalamus among others. In the hypothalamus, VDR partially colocalized with vasopressin, oxytocin, estrogen receptor-α, and β-endorphin to various degrees. We further functionally validated our model by demonstrating that the endogenous VDR agonist 1,25-dihydroxyvitamin D activated all tested tdTomato+ neurons in the paraventricular hypothalamus but had no effect on neurons without tdTomato fluorescence. Thus, we have generated a new mouse tool that allows us to visualize VDR-expressing cells and to characterize their functions. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/32634371  Title: Gut instincts: vitamin D/vitamin D receptor and microbiome in neurodevelopment disorders.  The gut microbiome regulates a relationship with the brain known as the gut-microbiota-brain (GMB) axis. This interaction is influenced by immune cells, microbial metabolites and neurotransmitters. Recent findings show gut dysbiosis is prevalent in autism spectrum disorder (ASD) as well as attention deficit hyperactivity disorder (ADHD). There are previously established negative correlations among vitamin D, vitamin D receptor (VDR) levels and severity of ASD as well as ADHD. Both vitamin D and VDR are known to regulate homeostasis in the brain and the intestinal microbiome. This review summarizes the growing relationship between vitamin D/VDR signalling and the GMB axis in ASD and ADHD. We focus on current publications and summarize the progress of GMB in neurodevelopmental disorders, describe effects and mechanisms of vitamin D/VDR in regulating the microbiome and synoptically highlight the potential applications of targeting vitamin D/VDR signalling in neurodevelopment disorders. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/32083397  Title: Vitamin D Receptor Polymorphisms Associated with Autism Spectrum Disorder.  Vitamin D is endowed with a number of biological properties, including down-regulation of inflammation, and might contribute to the pathogenesis of autism spectrum disorders (ASD). Vitamin D binds to the vitamin D Receptor (VDR); the biological activity of the ensuing complex depends on VDR FokI, BsmI, ApaI, and TaqI gene polymorphisms. We evaluated such Single Nucletoide Polymorphismsm (SNPs) in a cohort of 100 Italian families with ASD children. FokI genotype distribution was skewed in ASD children compared with their healthy sibs (Pc = 0.03 2 df) and to a group of 170 Italian healthy women (HC) (Pc = 0.04 2 df). FokI genotype and allelic distribution skewing were also observed in mothers of ASD children compared to HC (Pc = 0.04 2 df). Both Transmission Disequilibrium Test for single loci and haplotype analysis distribution revealed a major FokI (C) allele-mediated protective effect, which was more frequently transmitted (73%) than not transmitted to healthy sibs (P = 0.02). A protective FokI-, BsmI-, ApaI-, and TaqI (CCAG) haplotype was more frequently carried by healthy sibs than by ASD children (P = 1 × 10-4 ; OR: 0.1, 95% CI: 0.03-0.4) too. Finally, a strong gene-dose association of FokI (T) allele with both higher Childhood Autism Rating Scale score (Pc = 0.01) and, particularly, with hyperactivity behavior (Pc = 0.006) emerged in ASD children. Because the protein produced by the FokI (T) allele is transcriptionally less active than that produced by the FokI (C) allele, the reduced biological activity of the vitamin D/VDR complex prevalent in ASD could favor ASD- and maternal immune activation- associated inflammation. Vitamin D supplementation might be useful in preventative and rehabilitation protocols for ASD. Autism Res 2020, 13: 680-690. © 2020 International Society for Autism Research, Wiley Periodicals, Inc. LAY SUMMARY: Vitamin D deficiency and Vitamin D receptor (VDR) polymorphisms are associated with structural and functional brain abnormalities and behavioral disorders. We analyzed the association of VDR gene polymorphisms in a cohort of 100 Italian families with ASD children. A strong correlation between one of the VDR polymorphisms and hyperactivity behavior was evidenced in ASD children. In healthy mothers, the same VDR polymorphism was also correlated with an increased risk of giving birth to children with ASD. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/31142227  Title: Vitamin D and the nervous system.  Objective: to summarise the activities that Vitamin D (VD) carries out in the brain and to clarify the potential role of VD in neurological diseases. Methods: a literature research has been performed in Pubmed using the following keywords: 'Vitamin D', 'nervous system', 'brain'. Results: the studies reviewed show that VD contributes to cerebral activity in both embryonic and adult brain, helping the connectivity of neural circuits responsible for locomotor, emotional and reward-dependent behavior. Low VD serum levels have been found in patients affected by Alzheimer Disease, Parkinson Disease, Multiple Sclerosis, Autism Spectrum Disorders, Sleep Disorders and Schizophrenia. Discussion: findings are controversial and should be interpreted with caution, since most of the studies performed have observational study set and few interventional studies are available, producing conflicting results. Overall, it can be stated that the potential role of Vitamin D in neurological diseases is mostly unclear and further randomised controlled trials are needed to understand better whether Vitamin D supplementation treatment can be useful in brain disorders. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/30410834  Title: Fok-I, Bsm-I, and Taq-I Variants of Vitamin D Receptor Polymorphism in the Development of Autism Spectrum Disorder: A Literature Review.  The role of vitamin D in the development of autism spectrum disorder (ASD) is of intensified interest in medical science in recent years. Vitamin D has a significant role in neurogenesis, neuroprotection, and neurodevelopment. Due to the close association of vitamin D with the brain, it has been found that in the pathophysiology of several neuropsychiatric disorders vitamin D receptor (VDR) polymorphism plays a significant role. In this review article, we looked for a relation between VDR polymorphism and ASD. We systemically reviewed all the potential articles on the relation between VDR polymorphism and ASD. We found that several VDR variants FokI, BsmI, and TaqI polymorphisms are related to ASD. Even paternal VDR polymorphism can be a causative factor for ASD in the offspring. The relation between FokI (ff) genotype polymorphism and increased level of serum 1,25(OH)D3 in ASD patients is a very significant finding. Variation of ASD-related genotypes in different ethnic population raises a big question on whether the environmental factors also can do changes in human genotypes leading to ASD. |
| VDR |
| Url: https://pubmed.ncbi.nlm.nih.gov/26071405  Title: 1,25-Dihydroxyvitamin D regulates expression of the tryptophan hydroxylase 2 and leptin genes: implication for behavioral influences of vitamin D.  To investigate vitamin D-related control of brain-expressed genes, candidate vitamin D responsive elements (VDREs) at -7/-10 kb in human tryptophan hydroxylase (TPH)2 were probed. Both VDREs bound the vitamin D receptor (VDR)-retinoid X receptor (RXR) complex and drove reporter gene transcription in response to 1,25-dihydroxyvitamin D3 (1,25D). Brain TPH2 mRNA, encoding the rate-limiting enzyme in serotonin synthesis, was induced 2.2-fold by 10 nM 1,25D in human U87 glioblastoma cells and 47.8-fold in rat serotonergic RN46A-B14 cells. 1,25D regulation of leptin (Lep), encoding a serotoninlike satiety factor, was also examined. In mouse adipocytes, 1,25D repressed leptin mRNA levels by at least 84%, whereas 1,25D induced leptin mRNA 15.1-fold in human glioblastoma cells. Chromatin immunoprecipitation sequencing analysis of the mouse Lep gene in response to 1,25D revealed a cluster of regulatory sites (cis-regulatory module; CRM) at -28 kb that 1,25D-dependently docked VDR, RXR, C/EBPβ, and RUNX2. This CRM harbored 3 VDREs and single C/EBPβ and RUNX2 sites. Therefore, the expression of human TPH2 and mouse Lep are governed by 1,25D, potentially via respective VDREs located at -7/-10 kb and -28 kb. These results imply that vitamin D affects brain serotonin concentrations, which may be relevant to psychiatric disorders, such as autism, and may control leptin levels and affect eating behavior. |
| WAC |
| Url: https://pubmed.ncbi.nlm.nih.gov/35766809  Title: Phenotypic and Brain Imaging Findings Associated With a 10p Proximal Deletion Including the WAC Gene: Case Report and Literature Review.  Microarray-based techniques are an important testing method in etiological studies of intellectual disability and autism spectrum disorder. Interstitial deletion in the p11-p12 region of chromosome 10 is rare, having been reported in just 12 cases to date. Intellectual disability associated with the WAC gene in this region is referred to as DeSanto-Shinawi syndrome . Although all individuals with p11-p12 region of chromosome 10 deletion share a common phenotype involving intellectual disability and dysmorphic features, individuals with DeSanto-Shinawi syndrome usually do not experience the cardiac and neurologic abnormalities or cryptorchidism associated with a 10p11-p12 deletion. With this case report, we aim to expand the phenotypic spectrum of 10p11-p12 deletion. Our patient was a 9-year-old boy with intellectual disability, autism symptoms, dysmorphic features, and behavioral abnormalities. He had no cardiac problems or neurologic symptoms such as hypotonia, feeding difficulties, or seizures. However, he presented cryptorchidism in addition to symptoms that are consistent with DeSanto-Shinawi syndrome. Array comparative genomic hybridization of genomic DNA isolated from a peripheral blood sample revealed a heterozygous deletion in 10p11.23-p12.1, which contains the WAC gene. We discuss our case in the context of a literature review of candidate genes. It is still difficult to establish genotype-phenotype correlations for neurologic, cardiac, and visual symptoms, and cryptorchidism, in individuals with a 10p11-p12 deletion. As more individuals are diagnosed with deletion in this chromosomal region, the associated phenotypes will become clearer. |
| YY1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/35199341  Title: Ying Yang 1 engagement in brain pathology.  Herein, we discuss data concerning the involvement of transcription factor Yin Yang 1 (YY1) in the development of brain diseases, highlighting mechanisms of its pathological actions. YY1 plays an important role in the developmental and adult pathology of the nervous system. YY1 is essential for neurulation as well as maintenance and differentiation of neuronal progenitor cells and oligodendrocytes regulating both neural and glial tissues of the brain. Lack of a YY1 gene causes many developmental abnormalities and anatomical malformations of the central nervous system (CNS). Once dysregulated, YY1 exerts multiple neuropathological actions being involved in the induction of many brain disorders like stroke, epilepsy, Alzheimer's and Parkinson's diseases, autism spectrum disorder, dystonia, and brain tumors. A better understanding of YY1's dysfunction in the nervous system may lead to the development of novel therapeutic strategies related to YY1's actions. |
| YY1 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31838722  Title: Identification of De Novo JAK2 and MAPK7 Mutations Related to Autism Spectrum Disorder Using Whole-Exome Sequencing in a Chinese Child and Adolescent Trio-Based Sample.  Autism spectrum disorder (ASD) is a neurodevelopmental disorder with high phenotypic and genetic heterogeneity. Whole-exome sequencing studies have shown that de novo single-nucleotide variations (SNVs) play an important role in sporadic ASD. The present study aimed to search for de novo SNVs using whole-exome sequencing in 59 unrelated Chinese ASD sporadic trios, and found 24 genes (including five reported ASD candidate genes CACNA1D, ACHE, YY1, TTN, and FBXO11) with de novo harmful SNVs. Five genes (CACNA1D, JAK2, ACHE, MAPK7, and PRKAG2) classified as "medium-confidence" genes were found to be related to ASD using the Phenolyzer gene analysis tool, which predicts the correlation between the candidate genes and the ASD phenotype. De novo SNVs in JAK2, MAPK7, and PRKAG2 were first found in ASD. Both JAK2 and MAPK7 were involved in the regulation of the MAPK signaling pathway. Gene co-expression and inter-gene interaction networks were constructed and gene expression data in different brain regions were further extracted, revealing that JAK2 and MAPK7 genes were associated with certain previously reported ASD genes and played an important role in early brain development. The findings of this study suggest that the aforementioned five reported ASD genes and JAK2 and MAPK7 may be related to ASD susceptibility. Further investigations of expression studies in cellular and animal models are needed to explore the mechanism underlying the involvement of JAK2 and MAPK7 in ASD. |
| ZBTB20 |
| Url: https://pubmed.ncbi.nlm.nih.gov/25062845  Title: Neurodevelopmental disorders associated with dosage imbalance of ZBTB20 correlate with the morbidity spectrum of ZBTB20 candidate target genes.  Recently, a number of patients have been described with structural rearrangements at 3q13.31, delineating a novel microdeletion syndrome with common clinical features including developmental delay and other neurodevelopmental disorders (NDD). A smallest region of overlapping deletions (SRO) involved five RefSeq genes, including the transcription factor gene ZBTB20 and the dopamine receptor gene DRD3, considered as candidate genes for the syndrome. We used array comparative genomic hybridization and next-generation mate-pair sequencing to identify key structural rearrangements involving ZBTB20 in two patients with NDD. In a patient with developmental delay, attention-deficit hyperactivity disorder, psychosis, Tourette's syndrome and autistic traits, a de novo balanced t(3;18) translocation truncated ZBTB20. The other breakpoint did not disrupt any gene. In a second patient with developmental delay and autism, we detected the first microdeletion at 3q13.31, which truncated ZBTB20 but did not involve DRD3 or the other genes within the previously defined SRO. Zbtb20 directly represses 346 genes in the developing murine brain. Of the 342 human orthologous ZBTB20 candidate target genes, we found 68 associated with NDD. Using chromatin immunoprecipitation and quantitative PCR, we validated the in vivo binding of Zbtb20 in evolutionary conserved regions in six of these genes (Cntn4, Gad1, Nrxn1, Nrxn3, Scn2a, Snap25). Our study links dosage imbalance of ZBTB20 to a range of neurodevelopmental, cognitive and psychiatric disorders, likely mediated by dysregulation of multiple ZBTB20 target genes, and provides new knowledge on the genetic background of the NDD seen in the 3q13.31 microdeletion syndrome. |
| ZNF292 |
| Url: https://pubmed.ncbi.nlm.nih.gov/31723249  Title: De novo and inherited variants in ZNF292 underlie a neurodevelopmental disorder with features of autism spectrum disorder.  Intellectual disability (ID) and autism spectrum disorder (ASD) are genetically heterogeneous neurodevelopmental disorders. We sought to delineate the clinical, molecular, and neuroimaging spectrum of a novel neurodevelopmental disorder caused by variants in the zinc finger protein 292 gene (ZNF292). We ascertained a cohort of 28 families with ID due to putatively pathogenic ZNF292 variants that were identified via targeted and exome sequencing. Available data were analyzed to characterize the canonical phenotype and examine genotype-phenotype relationships. Probands presented with ID as well as a spectrum of neurodevelopmental features including ASD, among others. All ZNF292 variants were de novo, except in one family with dominant inheritance. ZNF292 encodes a highly conserved zinc finger protein that acts as a transcription factor and is highly expressed in the developing human brain supporting its critical role in neurodevelopment. De novo and dominantly inherited variants in ZNF292 are associated with a range of neurodevelopmental features including ID and ASD. The clinical spectrum is broad, and most individuals present with mild to moderate ID with or without other syndromic features. Our results suggest that variants in ZNF292 are likely a recurrent cause of a neurodevelopmental disorder manifesting as ID with or without ASD. |
| ZNF462 |
| Url: https://pubmed.ncbi.nlm.nih.gov/36604593  Title: ZFP462 safeguards neural lineage specification by targeting G9A/GLP-mediated heterochromatin to silence enhancers.  ZNF462 haploinsufficiency is linked to Weiss-Kruszka syndrome, a genetic disorder characterized by neurodevelopmental defects, including autism. Though conserved in vertebrates and essential for embryonic development, the molecular functions of ZNF462 remain unclear. We identified its murine homologue ZFP462 in a screen for mediators of epigenetic gene silencing. Here we show that ZFP462 safeguards neural lineage specification of mouse embryonic stem cells (ESCs) by targeting the H3K9-specific histone methyltransferase complex G9A/GLP to silence meso-endodermal genes. ZFP462 binds to transposable elements that are potential enhancers harbouring pluripotency and meso-endoderm transcription factor binding sites. Recruiting G9A/GLP, ZFP462 seeds heterochromatin, restricting transcription factor binding. Loss of ZFP462 in ESCs results in increased chromatin accessibility at target sites and ectopic expression of meso-endodermal genes. Taken together, ZFP462 confers lineage and locus specificity to the broadly expressed epigenetic regulator G9A/GLP. Our results suggest that aberrant activation of lineage non-specific genes in the neuronal lineage underlies ZNF462-associated neurodevelopmental pathology. |
| ZNF804A |
| Url: https://pubmed.ncbi.nlm.nih.gov/33446247  Title: Schizophrenia risk ZNF804A interacts with its associated proteins to modulate dendritic morphology and synaptic development.  Schizophrenia (SZ) is a devastating brain disease that affects about 1% of world population. Among the top genetic associations, zinc finger protein 804A (ZNF804A) gene encodes a zinc finger protein, associated with SZ and biolar disorder (BD). Copy number variants (CNVs) of ZNF804A have been observed in patients with autism spectrum disorders (ASDs), anxiety disorder, and BD, suggesting that ZNF804A is a dosage sensitive gene for brain development. However, its molecular functions have not been fully determined. Our previous interactomic study revealed that ZNF804A interacts with multiple proteins to control protein translation and neural development. ZNF804A is localized in the cytoplasm and neurites in the human cortex and is expressed in various types of neurons, including pyramidal, dopaminergic, GABAergic, and Purkinje neurons in mouse brain. To further examine the effect of gene dosage of ZNF804A on neurite morphology, both knockdown and overexpression of ZNF804A in primary neuronal cells significantly attenuate dendritic complex and spine formation. To determine the factors mediating these phenotypes, interestingly, three binding proteins of ZNF804A, galectin 1 (LGALS1), fasciculation and elongation protein zeta 1 (FEZ1) and ribosomal protein SA (RPSA), show different effects on reversing the deficits. LGALS1 and FEZ1 stimulate neurite outgrowth at basal level but RPSA shows no effect. Intriguingly, LGALS1 but not FEZ1, reverses the neurite outgrowth deficits induced by ZNF804A knockdown. However, FEZ1 and RPSA but not LGALS1, can ameliorate ZNF804A overexpression-mediated dendritic abnormalities. Thus, our results uncover a critical post-mitotic role of ZNF804A in neurite and synaptic development relevant to neurodevelopmental pathologies. |
| ZNF804A |
| Url: https://pubmed.ncbi.nlm.nih.gov/30670685  Title: A promoter variant in ZNF804A decreasing its expression increases the risk of autism spectrum disorder in the Han Chinese population.  Synaptic pathology may be one of the cellular substrates underlying autism spectrum disorder (ASD). ZNF804A is a transcription factor that can affect or regulate the expression of many candidate genes involved in ASD. It also localizes at synapses and regulates neuronal and synaptic morphology. So far, few reports have addressed possible associations between ZNF804A polymorphisms and ASD. This study aimed to investigate whether ZNF804A genetic variants contribute to ASD susceptibility and its possible pathological role in the disorder. We analyzed the relationship of two polymorphisms (rs10497655 and rs34714481) in ZNF804A promoter region with ASD in 854 cases versus 926 controls. The functional analyses of rs10497655 were then performed using real-time quantitative polymerase chain reaction, electrophoretic mobility shift assays, chromatin immunoprecipitation and dual-luciferase assays. The variant rs10497655 was significantly associated with ASD (P = 0.007851), which had a significant effect on ZNF804A expression, with the T risk allele homozygotes related with reduced ZNF804A expression in human fetal brains. HSF2 acted as a suppressor by down-regulating ZNF804A expression and had a stronger binding affinity for the T allele of rs10497655 than for the C allele. This was the first experiment to elucidate the process in which a disease-associated SNP affects the level of ZNF804A expression by binding with the upstream regulation factor HSF2. This result indicates that the rs10497655 allelic expression difference of ZNF804A during the critical period of brain development may have an effect on postnatal phenotypes of ASD. It reveals new roles of ZNF804A polymorphisms in the pathogenesis of psychiatric disorders. |
| ZNF804A |
| Url: https://pubmed.ncbi.nlm.nih.gov/20553308  Title: REVIEW: Genome-wide findings in schizophrenia and the role of gene-environment interplay.  The recent advent of genome-wide mass-marker technology has resulted in renewed optimism to unravel the genetic architecture of psychotic disorders. Genome-wide association studies have identified a number of common polymorphisms robustly associated with schizophrenia, in ZNF804A, transcription factor 4, major histocompatibility complex, and neurogranin. In addition, copy number variants (CNVs) in 1q21.1, 2p16.3, 15q11.2, 15q13.3, 16p11.2, and 22q11.2 were convincingly implicated in schizophrenia risk. Furthermore, these studies have suggested considerable genetic overlap with bipolar disorder (particularly for common polymorphisms) and neurodevelopmental disorders such as autism (particularly for CNVs). The influence of these risk variants on relevant intermediate phenotypes needs further study. In addition, there is a need for etiological models of psychosis integrating genetic risk with environmental factors associated with the disorder, focusing specifically on environmental impact on gene expression (epigenetics) and convergence of genes and environment on common biological pathways bringing about larger effects than those of genes or environment in isolation (gene-environment interaction). Collaborative efforts that bring together expertise in statistics, genetics, epidemiology, experimental psychiatry, brain imaging, and clinical psychiatry will be required to succeed in this challenging task. |