Investigating Differential Epigenetic Regulation Across Brain Regions In BIPOLAR DISORDER (BAPD) Patients

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What is Bipolar Disorder?

- Bipolar mood or affective disorder is characterised by recurrent episodes of manic and depression in the same patient at different times
- Earlier known as, manic depressive psychosis
- In the United States, about 4.4 % of adults experience Bipolar Disorder at some point.
- The causes are not clearly understood, but both environmental and genetic factors play a role. Many genes, each with small effects, contribute to the disorder

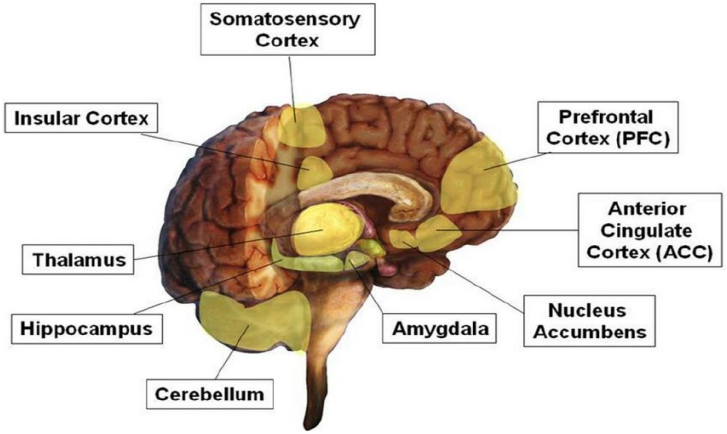
Types of Bipolar Disorder:

1. Bipolar Disorder 1
2. Bipolar Disorder 2
3. Cyclothymic disorder
4. Other types

Motivation For The Problem Addressed

- Increasing evidences of gene-environment interactions during critical periods of the lifespan manifesting as mental illness are emerging in the field of neuropsychiatry(Kendler2001: McEwen 2000)
- Epigenetics offers a potential mechanism by which environmental influences convey molecular factors contributing to disease risk (Petronis, 2010)
- There is increasing evidence for epigenetic dysregulation associated with neuropsychiatric disease (Lunnon et al., 2014, Hannon et al., 2016a, Sun et al., 2016)
- Identifying a mechanistic role of epigenetic factors in psychiatric disease will be valuable for understanding disease aetiology and discovering protective factors or novel psychopharmacological treatments (Boks et al., 2012)
- Histone acetylation represents a valid therapeutic target in depression(Tsankova N1, Renthal W 2007)

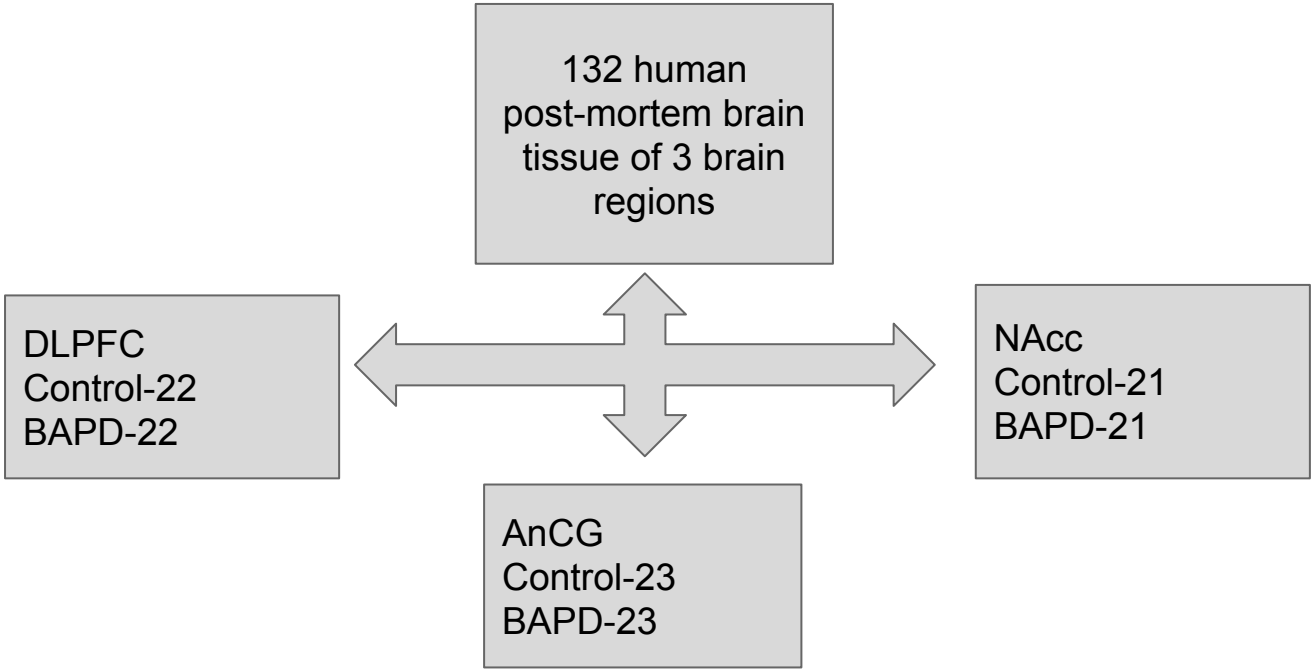
Questions Addressed



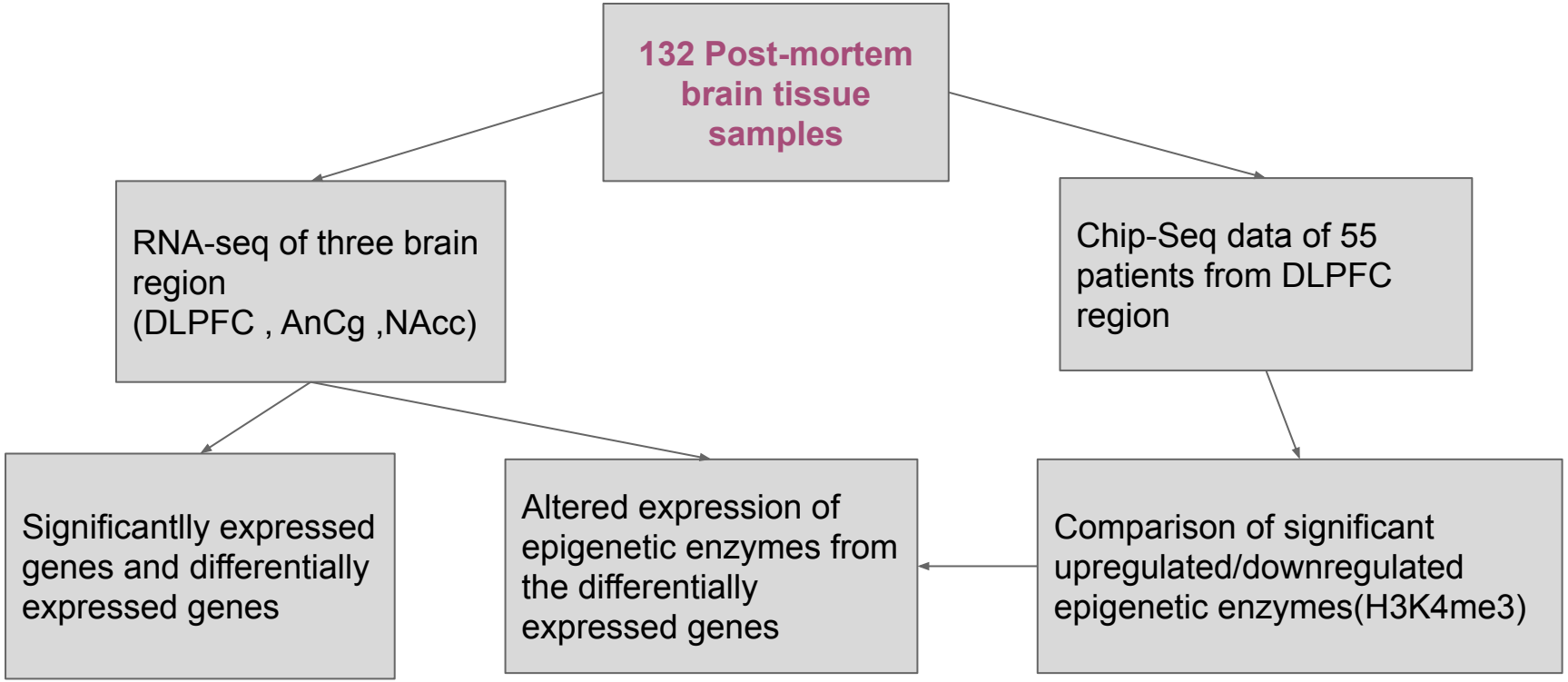
- To investigate differential epigenetic regulation across different brain regions (anterior cingulate, dorsolateral prefrontal cortex, nuclear accumbens) in 132 BPAD patients of Caucasian ethnicity from open source transcriptome data
- Correlating genome-wide enrichment of epigenetic (chromatin) marks (active marks: H3K4me3) in these patients from ChIP sequencing data
- Identifying upregulated and downregulated pathways associated with differentially expressed genes across three brain regions
- Identifying alterations in gene expression profiles of epigenetic enzymes such as HAT, HDAC and KMT

Data-Source

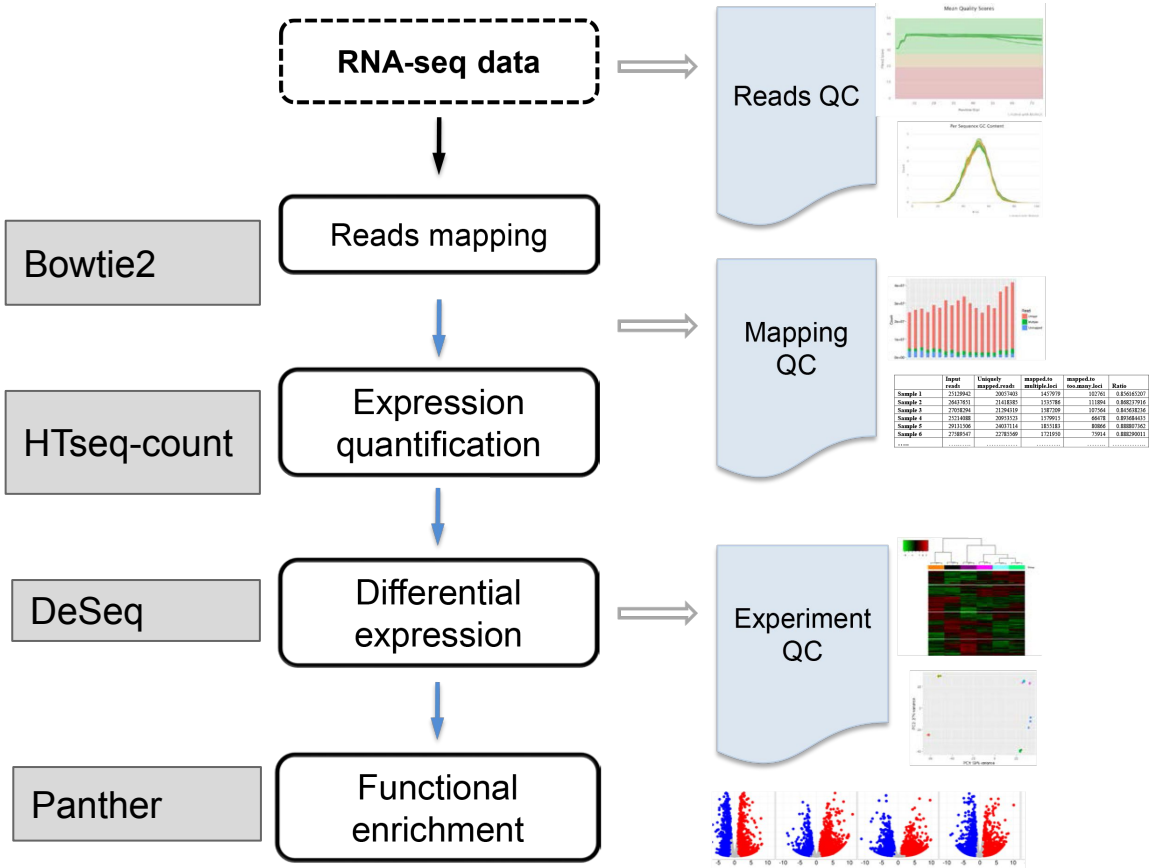
- Accession ID- GSE80655
- Clinically-annotated human post-mortem brain tissue of 3 brain regions of bipolar disorder patients -Dorsolateral prefrontal cortex(DLPFC) ,Anterior cingulate(AnCG) and Nuclear accumbans(NAcc)



Study Design

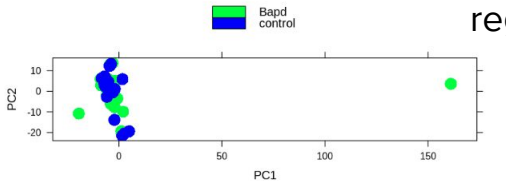


Methodology for RNA-Seq Analysis

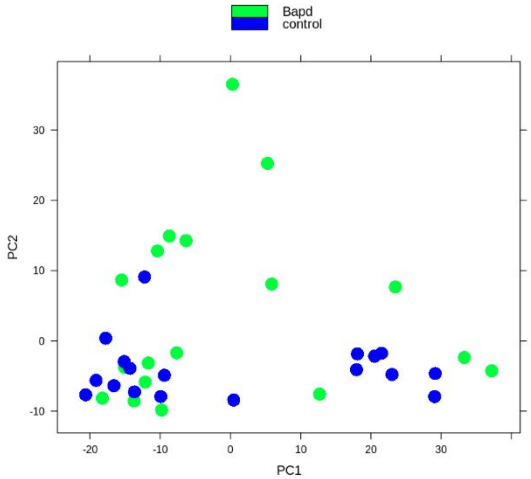


PCA plots depicting variations among samples

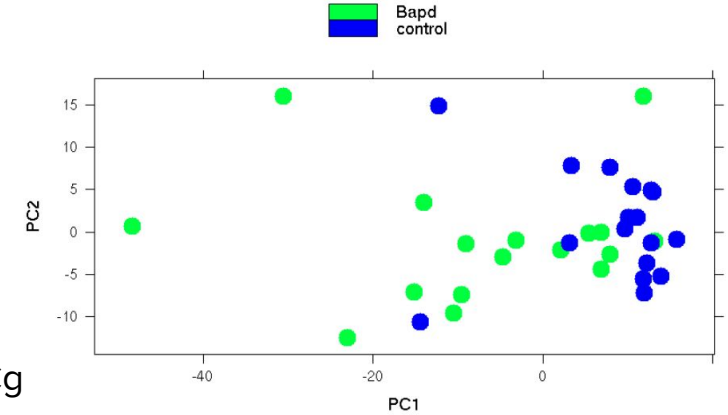
NAcc region



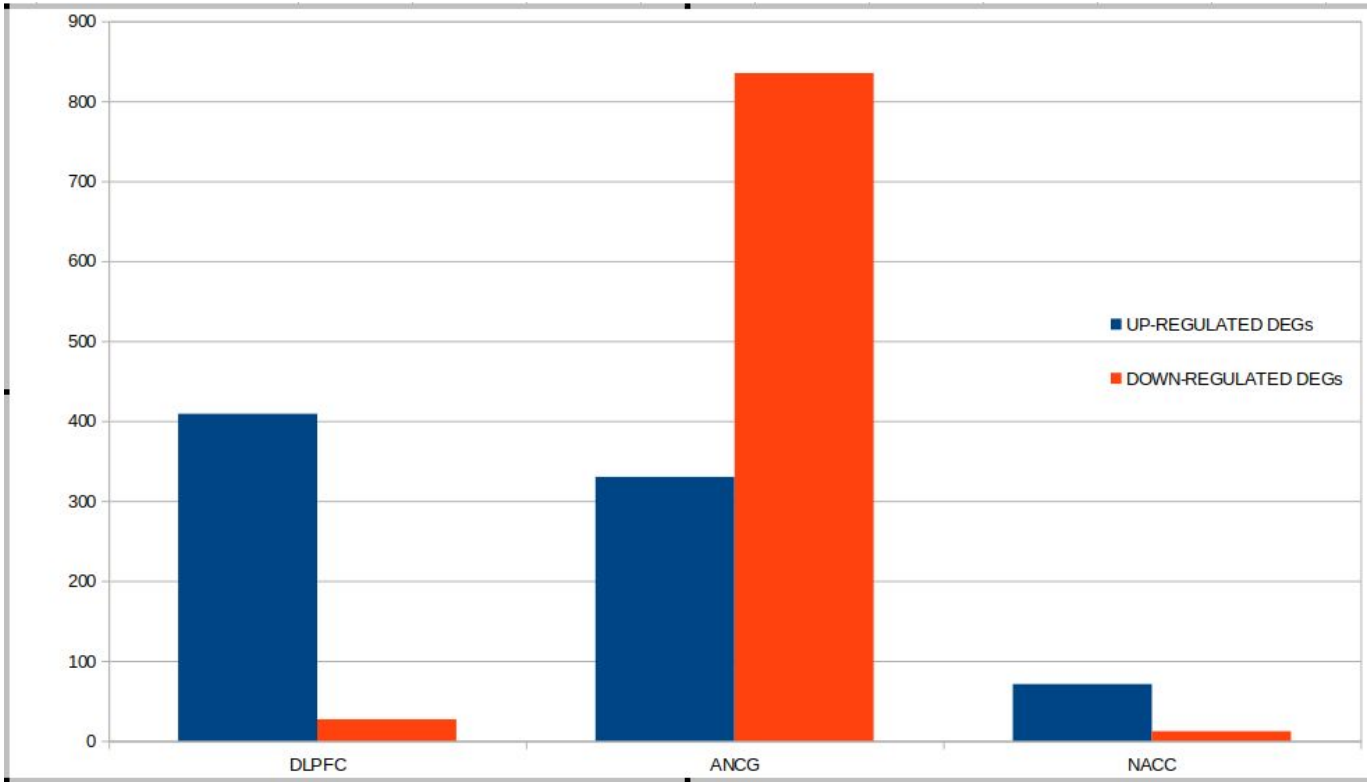
DLPFC region



AnCg region



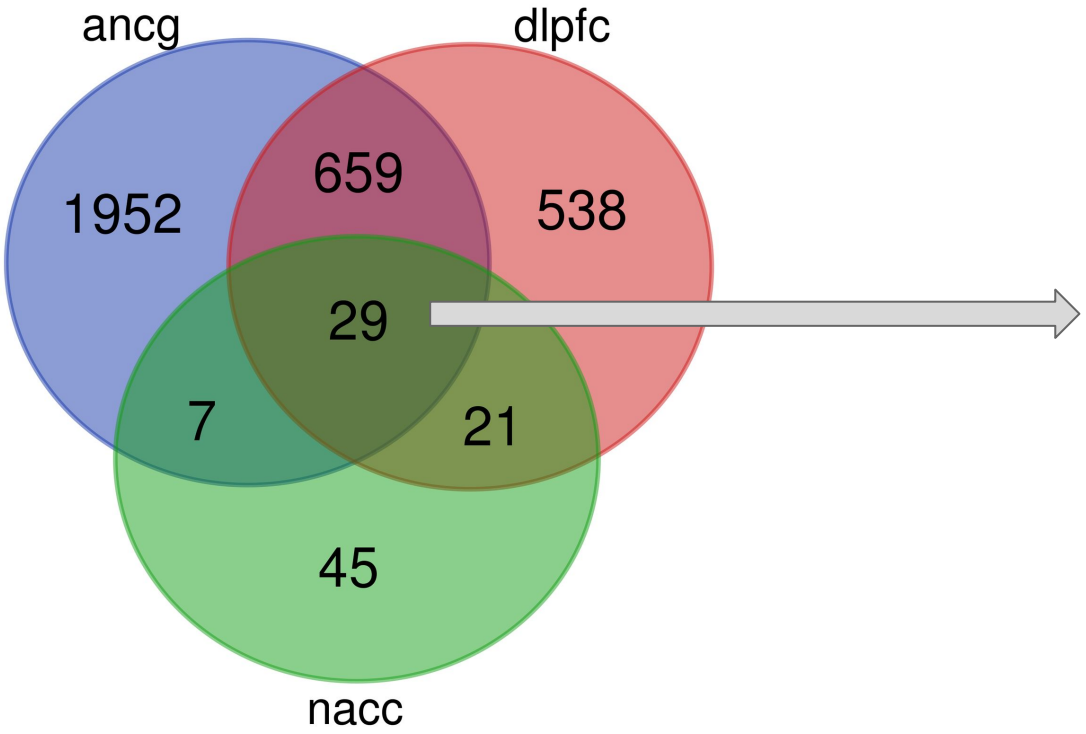
Differentially Expressed Genes Across 3 brain regions



Pathway-Analysis for Up-regulated and Down-regulated genes

	Dorsolateralprefrontalcortex	Nuclear Accumbens	Anterior Cingulate
Pathway-Analysis of Up-regulated genes	1.p53 pathway feedback loops2(PRB1)	1.Alzheimer disease-presenilin pathway(wnt1,wnt10B) 2.Cadherin signalling pathway 3.Wnt signalling pathways	1. Wnt signaling pathway(DVL1,wnt1) 2.Alzheimer disease-presenilin pathway(wnt1,LRP3,Notch3)
Pathway-Analysis of Down-regulated genes	1.Heterotrimeric G-protein signalling Gq and Go alpha mediated pathway 2.Heterotrimeric G-protein signalling Gi alpha and Gs alpha mediated pathway(OPRKI) 3.opiod prodynorphin pathway(OPRKI: kaapa type opoid receptor)	1.Interleukin signalling pathway 2.Alzheimer disease-presenilin pathway(LRP2)	1.Alzheimer disease presenilin pathway(wnt,PCDH8) 2. Angiogenesis Pathway 3. Cadherin signaling pathway 4. Wnt signaling pathway(wnt1,PCDH8)

Common DEGs across brain regions



RALYL, PAPSS2 ,PRR5 ,GRIP1
PCBP3,SGCZ, SLC44A3-AS1
MTHFD1L,CACNB2 , TP53, TPST1
MACROD2,MCTP1 ,SLC2A13,
TENM2, CSMD1, STXBP5L,
LRRTM4, ADGRB3, CDH9, GRM5,
CNTNAP3, RYR2, ALG1L9P
MSRA, XKR6, RIMS2,CHSY3
LOC645513

Top Pathways Of Common DEGs Across Three Brain Regions

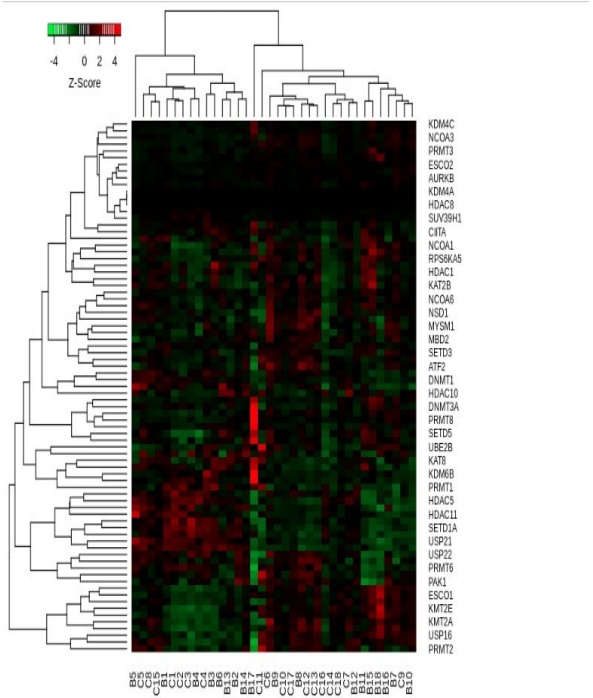
- **5HT2-TYPE receptor-mediated signalling pathway** : 5-hydroxy tryptamine/serotonin receptor(HTR2A) encodes for 5HT2A, 5HT2B, 5HT2C receptors. 5HT2A is responsible for addiction,mood,learning, anxiety, cognition, apetite, imagination. 5HT2B is responsible for sleep, cardiovascular pathway. 5HT2C is responsible for locomotion and sexual behaviour
- **Alzheimer Disease-amyloid secretase pathway**: CACNB2 gene encodes for β subunit of voltage-gated calcium channel. There are neural projections between hippocampus and Prefrontal cortex, alterations is responsible for dysregulation in cognition and perception
- **Beta1 adrenergic receptor signalling pathway**: (GRM5)Glutamatergic dysregulation is implicated in the neuropathology of bipolar disorder. These receptors are responsible for mood elevation and psychosis
- Wnt signalling pathway
- Beta 2 adrenergic receptor signalling pathway
- CCKR signalling map pathway
- p53 pathway

Epigenetic Enzymes Investigated In Neuropsychiatric Disorders

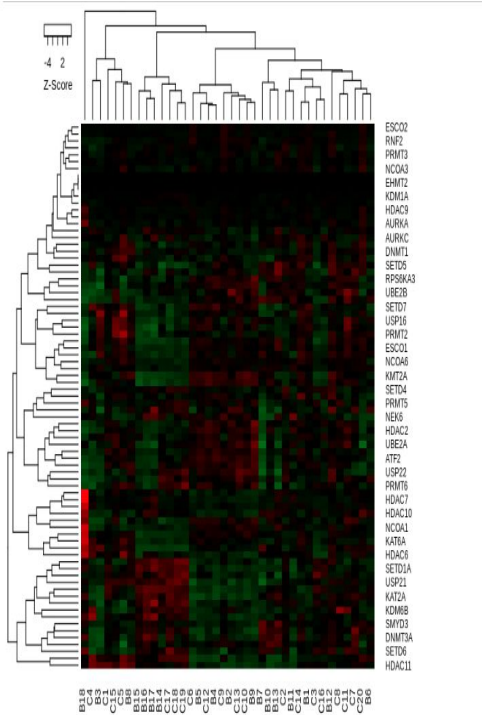
Category	No of Enzymes	Detail of Enzymes
Histone Methyl Transferases	15	CARM1/PRMT4, DOT1L, EHMT2, KMT2A, KMT2C, PRMT1, PRMT2, PRMT5, PRMT7, PRMT8, PRMT3, PRMT6, SETDB2, SMYD3, SUV39H1
Histone Demethylases	7	KDM5C, KDM6B, KDM4C, KDM5B, KDM1A, KDM4A, MBD2
SET Domain Proteins	15	ASH1L, SETD6, SETD4, SETD8, SETD1B, SETDB1, SETD3, SETD1A, SETD2, SETD7, KMT2E, NSD1, SETD5, SUV420H1, WHSC1
Histone Acetyl Transferases	17	ATF2, CDYL, CIITA, CSRP2BP, ESCO1, HAT1, KAT2A, KAT5, KAT6A, KAT7, KAT8, KAT2B, KAT6B, NCOA1, NCOA6, ESCO2, NCOA3
Histone Deacetylases	11	HDAC5, HDAC4, HDAC11, HDAC9, HDAC6, HDAC10, HDAC1, HDAC2, HDAC3, HDAC8, HDAC7
Kinases	7	AURKA, AURKC, AURKB, NEK6, PAK1, RPS6KA3, RPS6KA5
Ubiquitin Ligases	9	DZIP3, RNF2, RNF20, UBE2A, UBE2B, USP16, USP21, USP22, MYSM1
DNA Methyl Transferases	3	DNMT3B, DNMT3A, DNMT1

Epigenetic Enzymes Shows Differential Regulation Across 3 Brain Regions

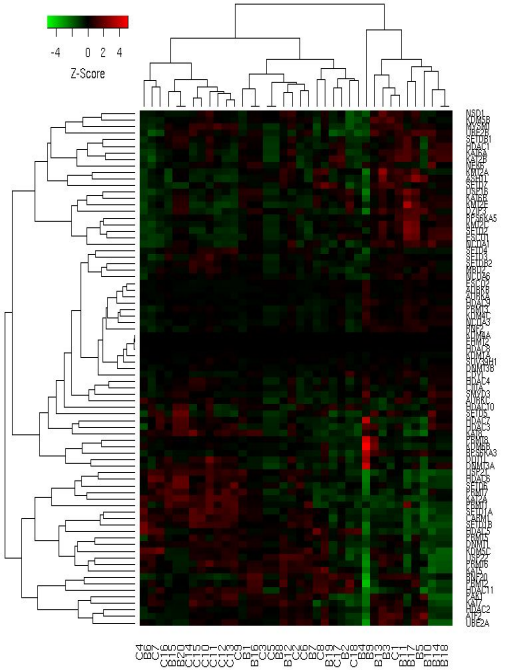
DLPFC region



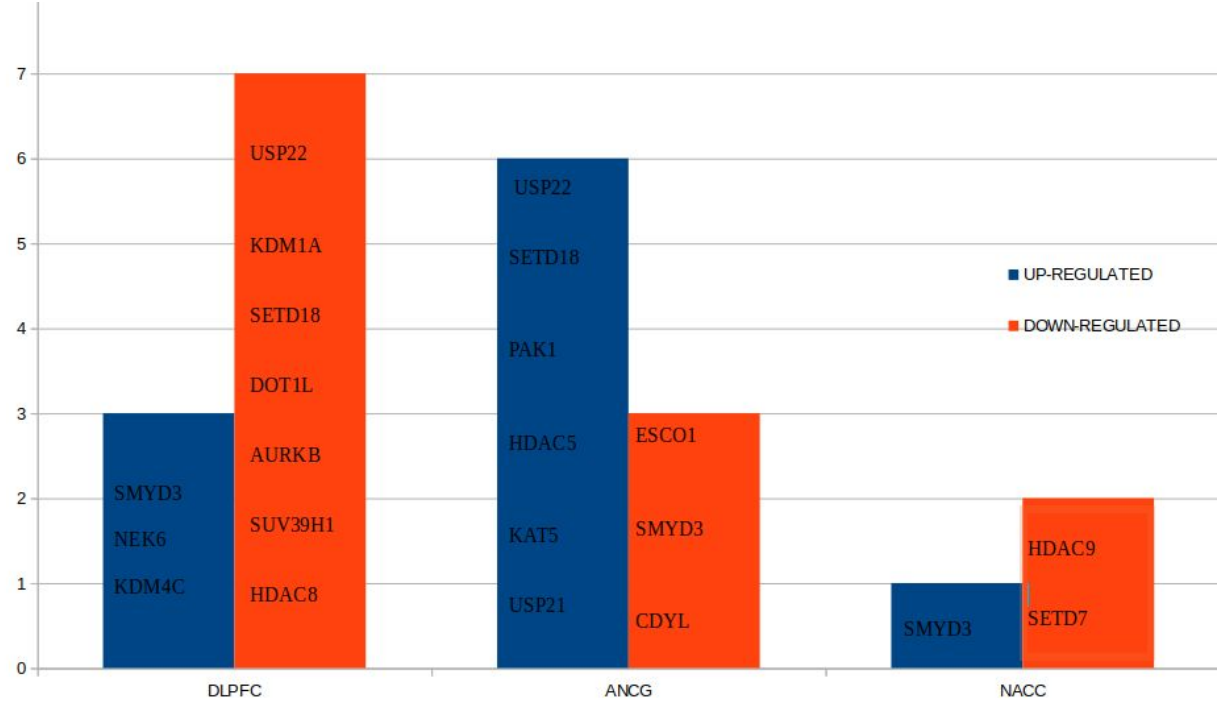
NAcc region



ANcg region



Expression Of Upregulated and Downregulated Epigenetic Enzymes



Epigenetic markers	Up regulated Epigenetic enzymes	Down-regulated Epigenetic enzymes
Readers	SMYD3 NEK3	CDYL
Writers	USP22 SETD1B KAT5 PAK1	DOT1L SUV39H1 SETD1B SETD7 ESC01
Erasers	KDM4C KDM1A HDAC5 USP21 USP22	KDM1A HDAC9 HDAC8

Conclusion

- **SMYD3** encodes a histone methyltransferase which is upregulated in Dlpfc and NAcc regions, is an **active marker of H3K4(me3)** having distinct synaptic profile. Upregulation of SMYD3 alters glutamatergic transmission and plasticity(mood shifts) in Bipolar, Inhibitors of SMYD3 can be a potential therapeutics
- **USP22(Ubiquitin-Specific Peptidase 22)** is downregulated in Dlpfc and upregulated in ANcg region, catalyzes deubiquitination of histones H2BK120. It is responsible for cerebral and retinal degeneration but has not been connected with bipolar yet
- **DOT1L,** a unique histone methyltransferase that targets the histone H3K79me residue for mono-, di- and tri- methylation, activation of Wnt-responsive genes, involving a GSK-3β(pivoted role in pathogenesis of Mood Disorders). Inhibitors for DOT1L(lithium, valproic acid) can be targetted as potential therapeutic
- Genes associated with hypertrophic cardiomyopathy- **MYOZ2,MYLK,CAV3** are downregulated in Ancg region which seems a connection between BAPD and cardiac hypertrophy
- Inhibitors targeting these specific proteins can be used as an effective therapy for treating the disease.

References

- https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2767266/
- https://academic.oup.com/ijnp/article/16/2/289/623541
- https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5190139/
- https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6504680/
- https://www.nature.com/articles/mp2013170
- https://science.sciencemag.org/content/362/6420/eaat8127
- https://genomemedicine.biomedcentral.com/articles/10.1186/s13073-017-0458-5

Thank you!