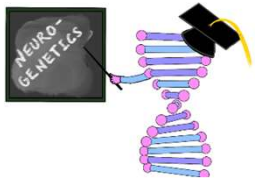
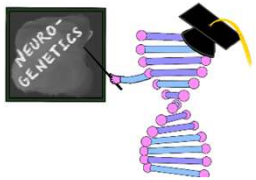
Somatic Mosaicism

Module 12



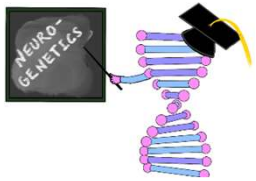
Learning Objectives

- Describe how somatic variants arise in the brain
- Explain how brain somatic variants contribute to neurological disease in children
- Compare genetic testing modalities used for detection of somatic variants
- Appraise the evidence that variants that activate the mTOR pathway cause brain malformations and epilepsy
- List other mosaic variants that cause neurological disease in children



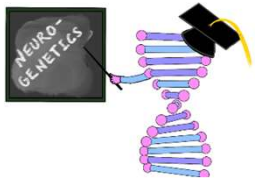
Chief Complaint

- 4.5 y/o boy with:
  - Refractory focal epilepsy
  - Electrographic status epilepticus of sleep
  - Worsening cognition and behavior
  - Chronic left hemibody weakness



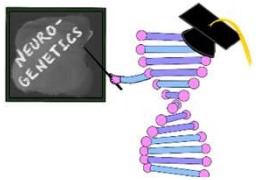
Differential Diagnosis - Interactive

Gene/ Condition	Biochemical Marker	Treatment/ Unique features



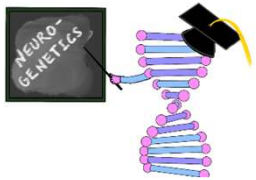
Differential Diagnosis

- Non-lesional focal epilepsy
- Focal cortical dysplasia
- Hemimegalencephaly
- Sturge Weber Syndrome
- Rasmussen Syndrome
- Stroke and post-stroke epilepsy
- Epilepsy from other focal structural abnormality (e.g. tumor, porencephaly, schizencephaly)
- Landau Kleffner Syndrome



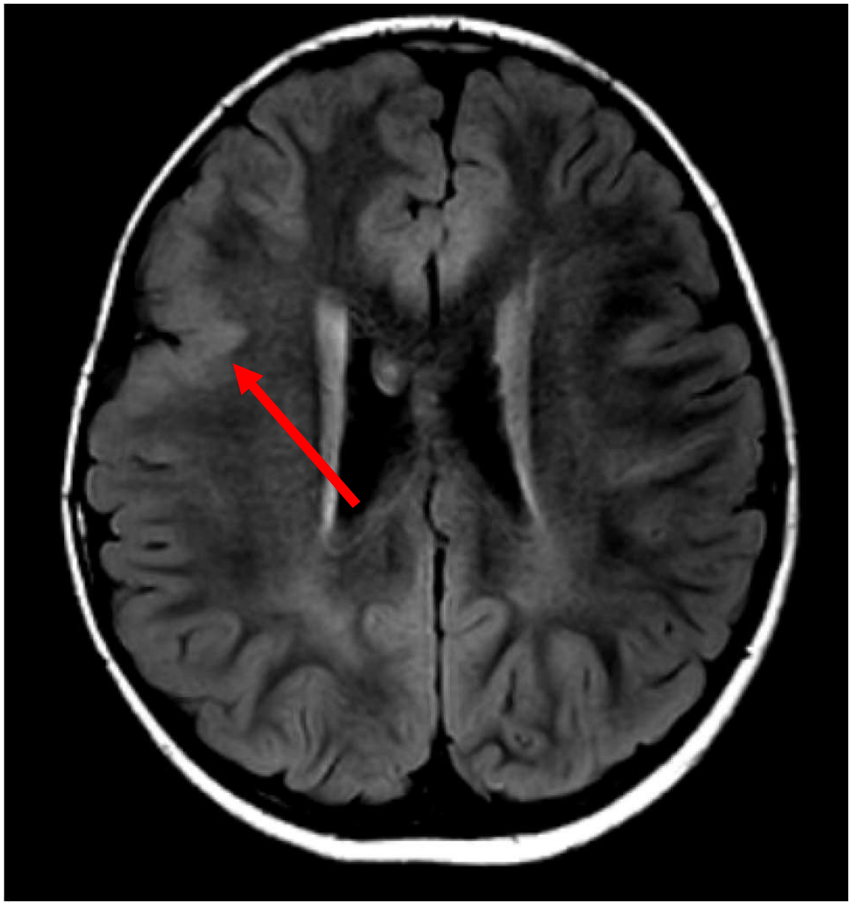
HPI and Exam

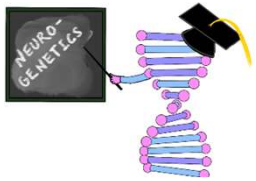
- Relatively unremarkable history other than strong right-handedness, dragging of left leg, and heterochromia iridum
- Seizure onset at age 3 with typical semiology
  - During sleep: Stiffening of arms/body, have pulling of his left face
  - Left eye deviation and rolling of the body to the left side
- No family history of epilepsy or neurodevelopmental disorders
- Physical exam: Doesn't use left side as proficiently as right



Work-Up

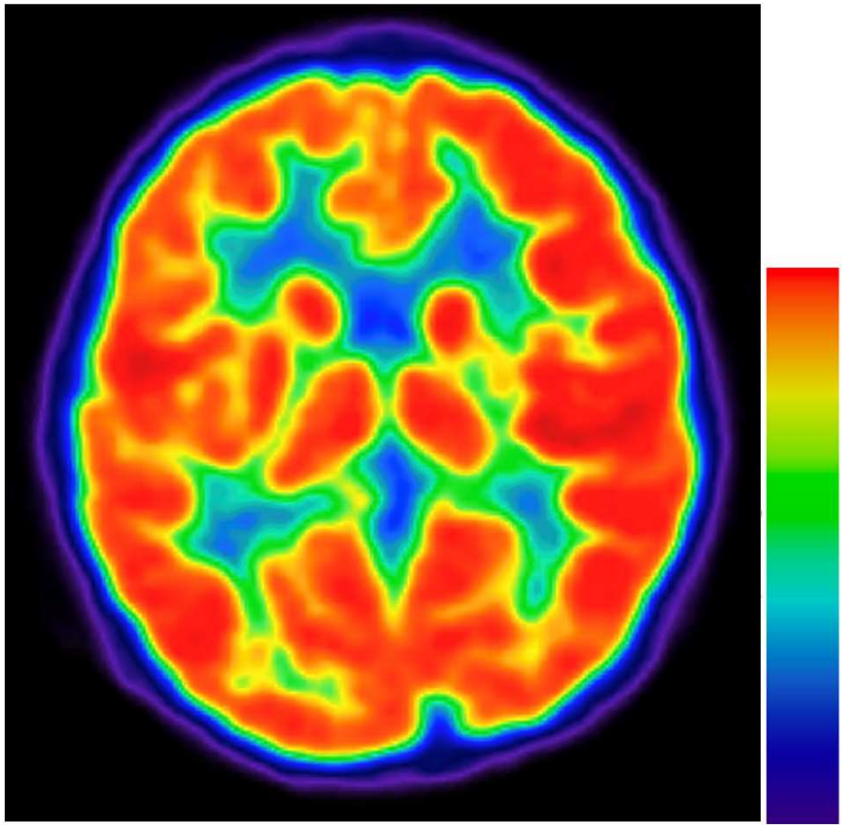
- Brain MRI (age 4)
  - FLAIR hyperintensities in the **right** anterior temporal tip, insula, mid and posterior hippocampus, and **lateral frontal region**

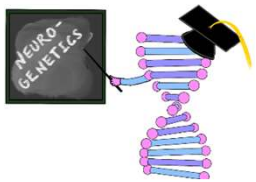




Work-Up Continued

- PET scan (age 4)
  - Diffuse hypometabolism in the **right frontal lobe** and superior temporal gyrus extending to the striatum.
- Lumbar puncture unremarkable (4.5 yrs)
- Epilepsy gene panel negative





Genetic Testing Results

- Epilepsy gene panel negative from saliva

Test Performed
Sequence analysis and deletion/duplication testing of the 125 genes listed in the results section below.
- Invitae Epilepsy Panel

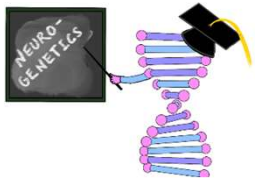
Reason for Testing
Diagnostic test for a personal history of disease

Summary

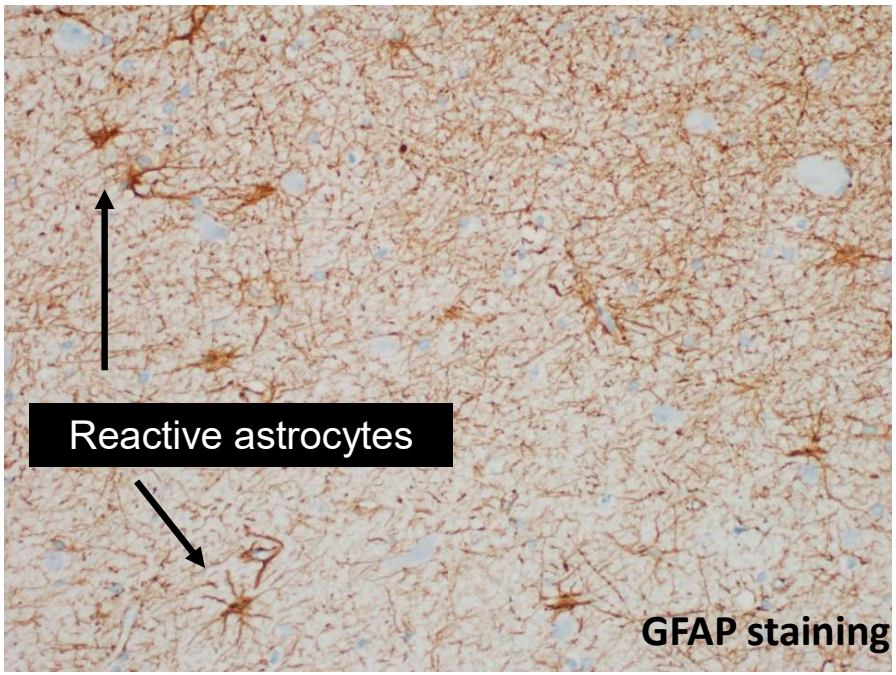
Negative result. No Pathogenic sequence variants or deletions/duplications identified.

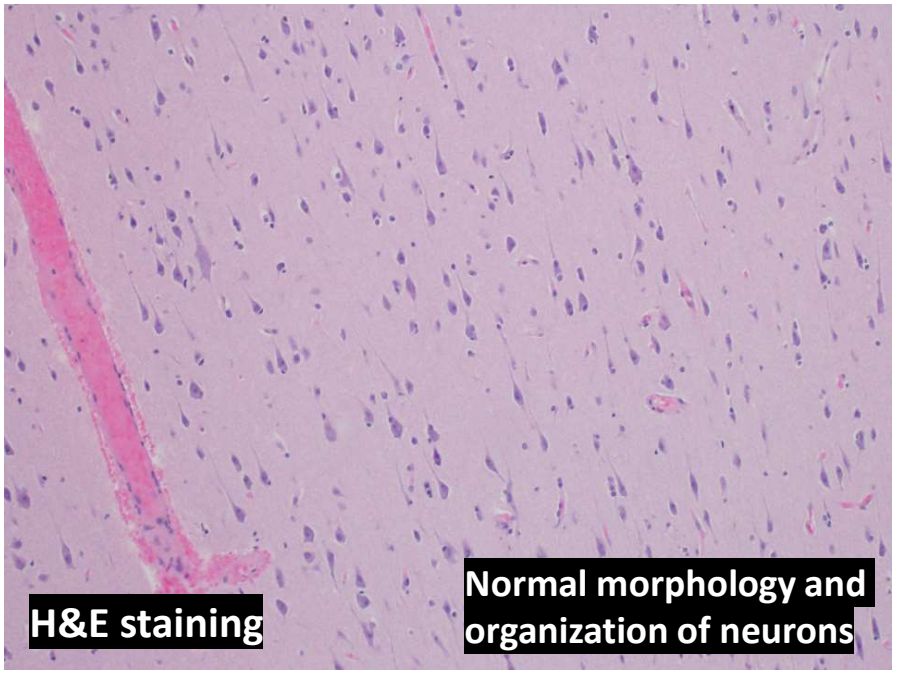
Clinical Summary

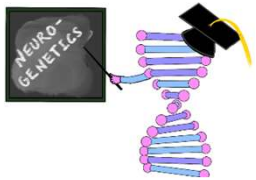
- This negative test result does not eliminate the possibility that this individual's condition has a genetic component. Clinical follow up of this individual and their family members may still be warranted.
- These results should be interpreted within the context of additional laboratory results, family history, and clinical findings. Genetic counseling is recommended to discuss the implications of this result. For access to a network of genetic providers, please contact Invitae at clientservices@invitae.com, or visit www.nsgc.org or tagc.med.sc.edu/professional_organizations.asp.



Right Temporal Brain Biopsy



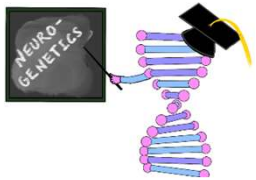




Mild reactive gliosis with minimal perivascular lymphohistiocytic inflammation with no evidence of dysplasia

Next-Gen Sequencing from Brain Tissue (from 2017)

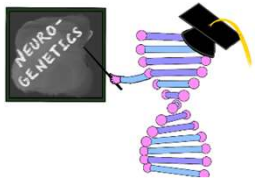
- MTOR exon 53 single nucleotide substitution
- DNA Change: c.7255G>A (for NM_004958.3)
- Amino acid change: E2419K mutation in mTOR gene
- Variant allele frequency of 10%



Interactive Exercise

MTOR E2419K, variant allele frequency of 10% in brain tissue

- Team A: What does the gene code for and where is it expressed?
- Team B: What neurological disorders do variants in MTOR cause?
- Team C: Is the E2419K variant a gain or loss of function?
- Team D: Does the identified variant explain the clinical presentation?

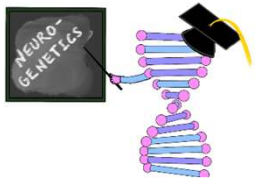


Testing Lab Interpretation

"While the specific MTOR detected in this specimen (E2419K) has not been described in epileptogenic malformations, this mutation is well-described as a somatic mutation in **cancers**: colorectal cancer, clear cell renal cell carcinoma and urothelial carcinoma.

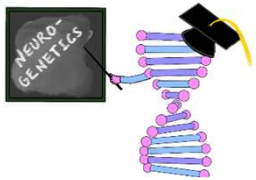
This mutation affects the kinase domain of MTOR and has been shown to result in **constitutive activation** of MTOR and sensitivity to rapamycin, in vitro.

In a phase 1 clinical trial, a patient with urothelial carcinoma harboring the MTOR E2419K mutation was identified as an **exceptional responder to everolimus** and pazopanib. Everolimus has also been shown to be an effective treatment for TSC-associated epilepsy."



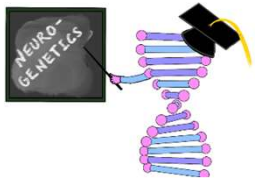
Testing of Brain Tissue for Somatic Variants

- Somatic Overgrowth Panel
- Megalencephaly panel
- Can use:
  - Skin biopsy (ectodermal skin cells more likely to reflect pathogenic variant present in brain than mesodermal blood cells)
  - Fresh tissue
  - formalin-fixed slides OR formalin-fixed tissue block

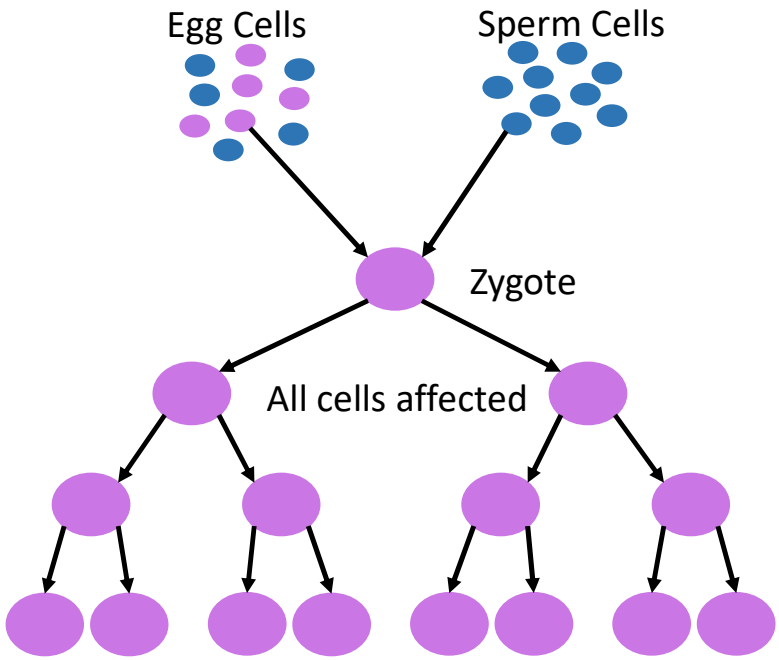


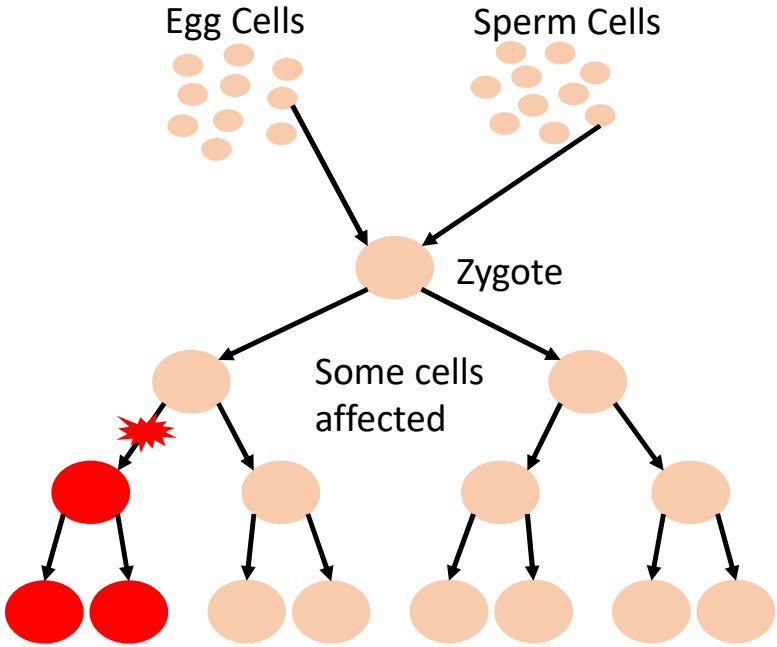
Limitations of Next-Gen Sequencing for Brain Samples

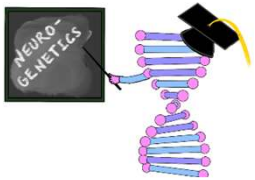
- High depth of sequencing is needed to detect lower variant allele frequencies
- High depth of sequencing is available with targeted gene panels but not clinically available for exome and genome sequencing yet
- Sequencing has higher yield in brain tissue, compared to blood, saliva, or skin, and brain tissue is hard to access



Germline vs. Somatic Mosaicism

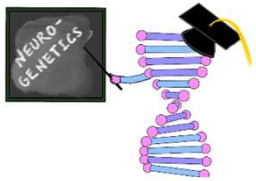




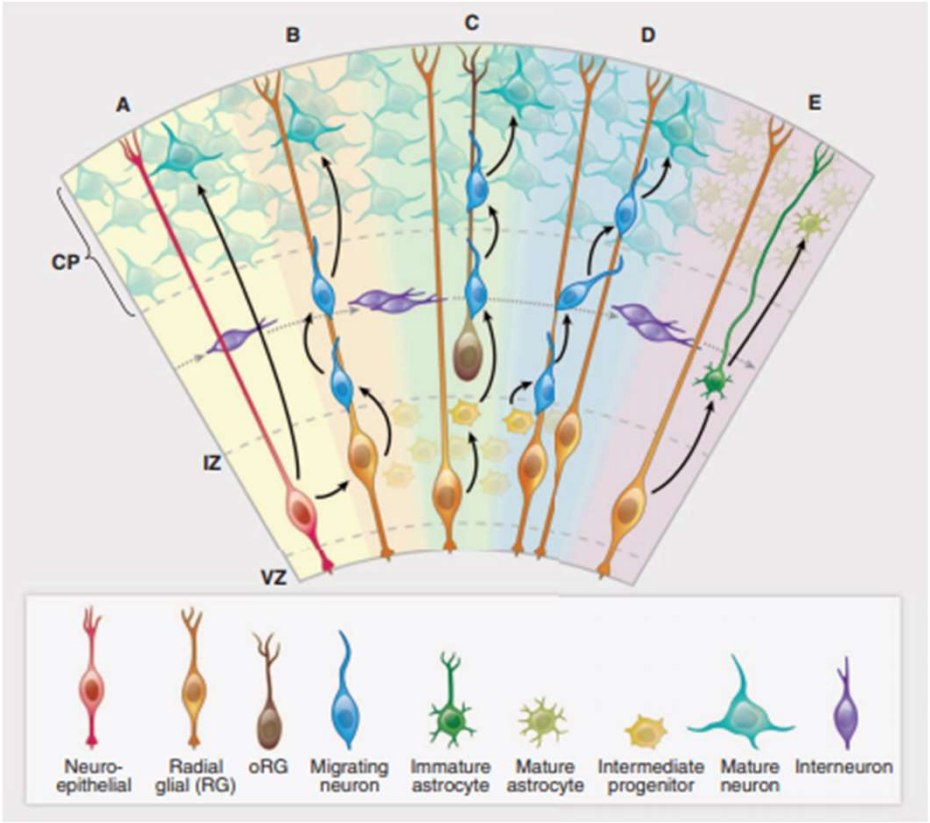


Germline vs. Somatic

	WHAT IS TESTED?	INHERITANCE
INHERITED	Blood or saliva	Can be inherited/passed on through a family
SOMATIC	Tissue in the body thought to be affected (frequently skin biopsy, can be other organ such as brain*)	Not inherited; arose after conception, not present in germ cells and only present in affected tissue



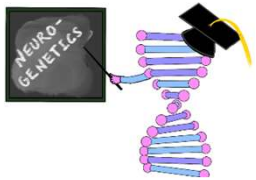
Somatic Variant Can Occur at Various Stages of Corticogenesis



Neural stem cells

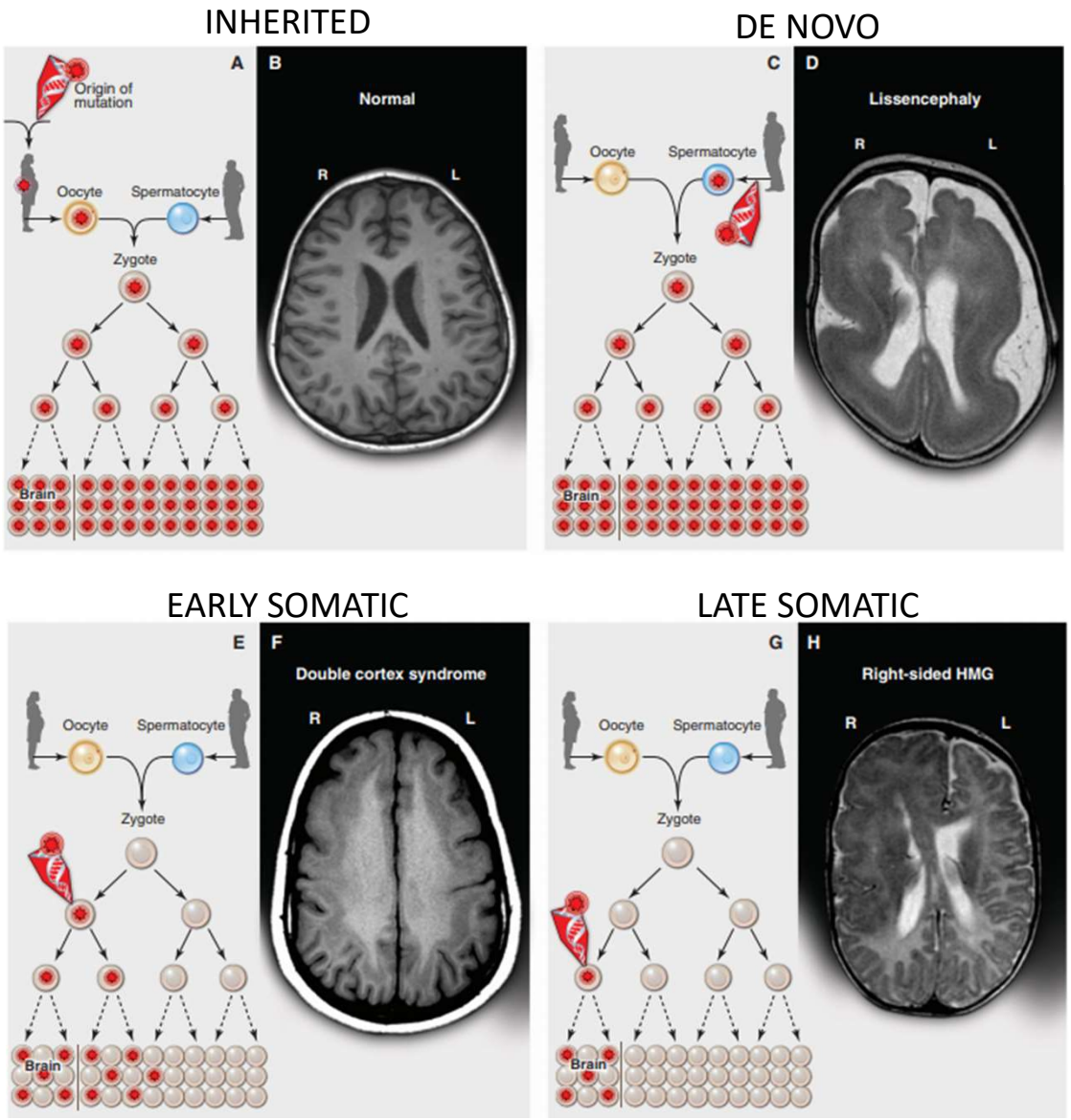
Neurogenesis

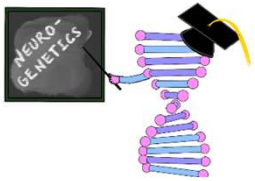
Astrogliogenesis



Poduri et al, 2013.

Different types of mutations can cause neurological disease

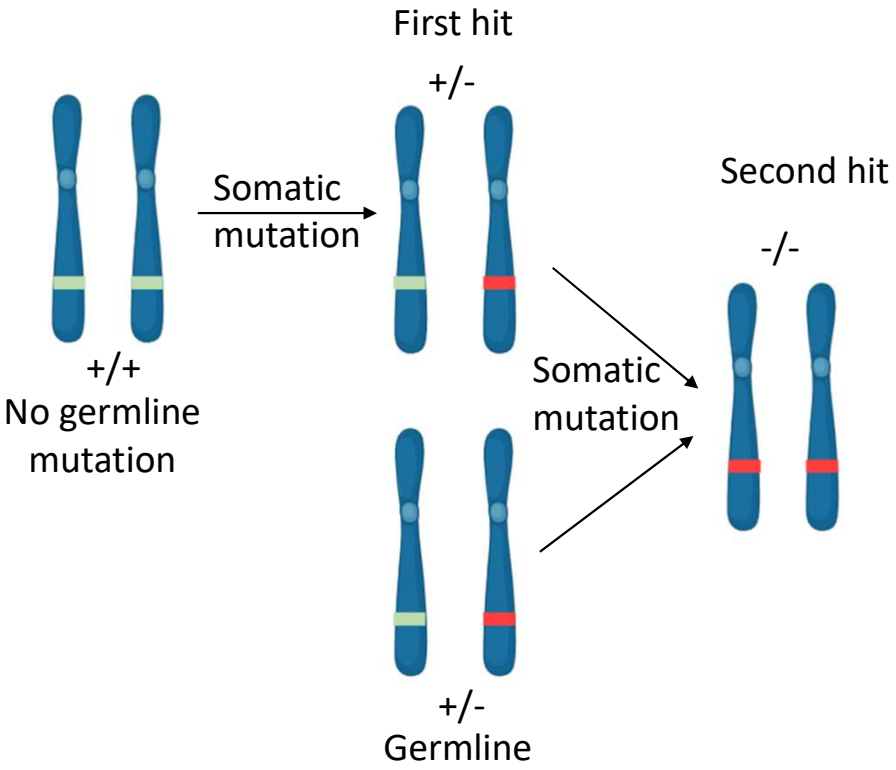


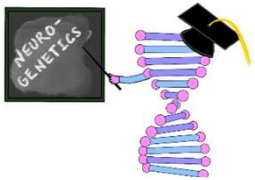


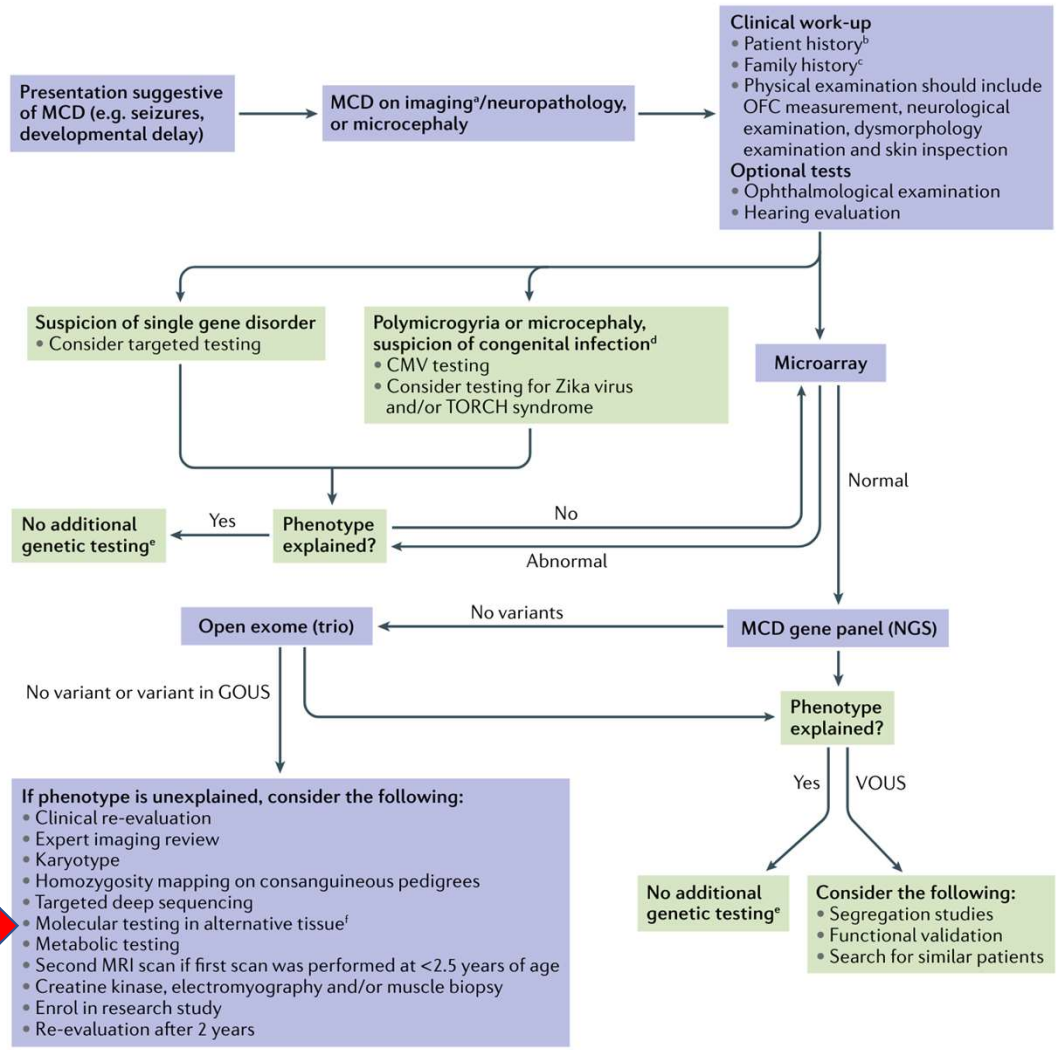
Poduri et al, 2013.

“Two-Hit” Hypothesis

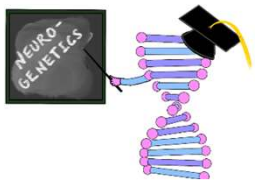
- Proposed in 1971 for tumorigenesis
- Patients inheriting an RB1 mutation develop retinoblastoma
- Loss of heterozygosity
- Can also explain focal disease in germline heterozygous genetic malformations of cortical development (TSC, DEPDC5, NPRL2, NPRL3)







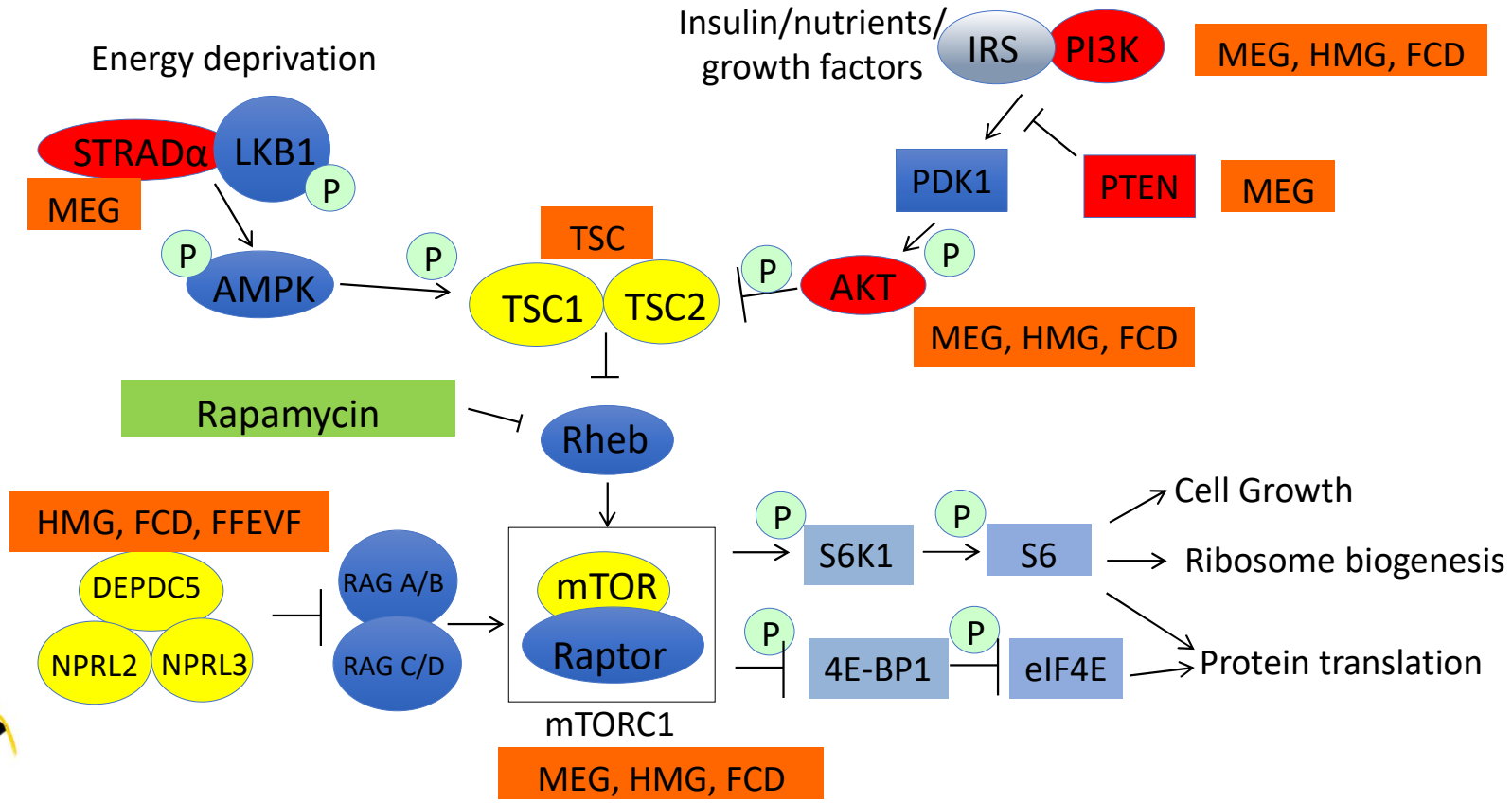
Molecular testing in alternative tissue

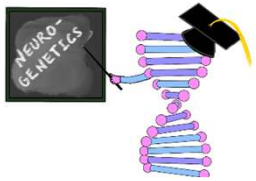


Diagnostic workflow for malformations of cortical development

Oegema, R. et al. Nat Rev Neurol, 2020

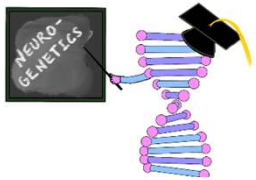
mTOR Pathway in Cortical Malformations and Epilepsy





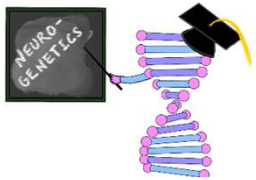
Treatment of Patient with HMEG/MTOR Variant

- Everolimus started and after a year on this medication, seizures finally abated.
- Now patient is seizure-free > 2 years, was able to wean to only 1 additional anti-seizure medication and had remission of ESES.
- Had evaluation for epilepsy surgery with sEEG, but right around this time he improved on everolimus, so has not needed surgery.
- Cognition and behavior improved as well.



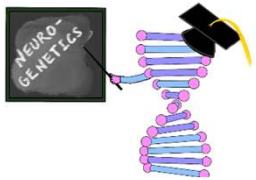
Non-mTOR Genes and Somatic Variants

- SLC35A2 – NLFE, FCD I, mMCD, MOGHE
- Multiple monogenic epilepsies that are typically germline can present with somatic mosaicism (CDKL5, GABRA1, GABRG2, GRIN2B, KCNQ2, MECP2, PCDH19, SCN1A, and SCN2A)
- Double cortex (subcortical band heterotopia): heterozygous DCX in females with random X-inactivation, and early somatic mosaicism in males
- RAS-RAF-MAPK pathway-Hippocampal sclerosis and MCD
- SHH – Hypothalamic hamartoma
- GNAQ-Sturge-Weber syndrome
- Trisomy 1q – PMG, epilepsy



Suggested Reading

- D'Gama, A.M., Poduri, A., 2023. Brain somatic mosaicism in epilepsy: Bringing results back to the clinic. Neurobiology of Disease 106104. https://doi.org/10.1016/j.nbd.2023.106104
- Jourdon et al., 2020. The role of somatic mosaicism in brain disease. Current Opinion in Genetics & Development 65, 84–90. https://doi.org/10.1016/j.gde.2020.05.002
- Lai et al., 2022. Somatic variants in diverse genes leads to a spectrum of focal cortical malformations. Brain 145, 2704–2720. https://doi.org/10.1093/brain/awac117
- Winawer et al., 2018. Somatic SLC35A2 variants in the brain are associated with intractable neocortical epilepsy. Annals of Neurology 83, 1133–1146. https://doi.org/10.1002/ana.25243
- Jamuar et al., 2014. Somatic Mutations in Cerebral Cortical Malformations. New England Journal of Medicine 371, 733–743. https://doi.org/10.1056/NEJMoa1314432



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- Kristin Baranano (Hopkins)
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