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Symphony Conductors Lose the Baton: Role of Fast-Spiking Interneurons in Orchestrating DS

Impaired Theta-Gamma Coupling Indicates Inhibitory Dysfunction and Seizure Risk in a Dravet Syndrome Mouse Model

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Dravet syndrome (DS) is an epileptic encephalopathy that still lacks biomarkers for epileptogenesis and its treatment. Dysfunction of NaV1.1 sodium channels, which are chiefly expressed in inhibitory interneurons, explains the epileptic phenotype. Understanding the network effects of these cellular deficits may help predict epileptogenesis. Here, we studied theta–gamma coupling as a potential marker for altered inhibitory functioning and epileptogenesis in a DS mouse model. We found that cortical theta–gamma coupling was reduced in both male and female juvenile DS mice and persisted only if spontaneous seizures occurred. Theta–gamma coupling was partly restored by cannabidiol. Locally disrupting NaV1.1 expression in the hippocampus or cortex yielded early attenuation of theta–gamma coupling, which in the hippocampus associated with fast ripples, and which was replicated in a computational model when voltage-gated sodium currents were impaired in basket cells. Our results indicate attenuated theta–gamma coupling as a promising early indicator of inhibitory dysfunction and seizure risk in DS.

Commentary

Dravet syndrome (DS) is a severe type of genetic childhood epilepsy. Although seizures appear early in life in patients with DS and are associated with SUDEP, overall there is good life expectancy. Dravet syndrome is receiving renewed focus for novel interventions with the advent of precision medicines such as antisense oligonucleotide (ASO) technology.[1,2] However, reliable clinical and translatable preclinical biomarkers to guide prognostication and quantification of treatment efficacy other than seizure severity or SUDEP are still lacking. GABAergic dysfunction could provide that biomarker. Significant components of the syndrome relate to developmental delays, behavioral/sleep dysfunction, cognitive deficits, and sensory integration disorders, all of which rely on the GABAergic system.

The orchestration of fine-tuning complex cortical outputs to incoming signals is one of the primary functions of interneurons which underlies all cortical higher functions, including learning, memory, and cognition. The role of parvalbumin interneuron (PV-IN) dysfunction in DS pathogenesis and seizures is well-documented,[3] and the concept of specifically targeting PV-INs for therapy is not new. More importantly, recent studies have emphasized the role of PV fast-spiking INs in cortical function and critical homeostatic mechanisms[4,5] that could reliably be quantified using continuous electroencephalograms (cEEG) both in the laboratory and clinic. IN activity is known to organize excitatory neural activity between local networks especially for high-frequency rhythms such as gamma oscillations. Fast-spiking PV-IN plays a critical role in synchronizing cortical gamma oscillations between spatially distant sites.[4,6] The PV-INs are electrically coupled, but the gap junction coupling via Connexin 36 between functionally distinct IN subtypes give PV fast-spiking INs a distinct role as symphony conductors between distant brain regions.[7] GABAergic hypotheses for the emergence of seizures[8] are also evolving. Given that the GABAergic system continues its circuit-level maturation long after birth into adolescence,[9] elucidating the temporal window for future interventional therapies for DS where a significant pathology related to INs is established becomes critical.

Theta–gamma coupling is a term used to quantitate the phenomenon of cross-frequency coupling wherein high-frequency gamma (30-50 Hz) oscillations are modulated by low-frequency theta (4-8 Hz) oscillations. Both decreased synaptic inhibition and increased synaptic excitation onto PV-INs disrupt cortical gamma oscillation synchrony and gamma–theta coupling.[5,6,10] In this study, the authors showed that decreased theta–gamma coupling preceded and associated with seizure activity in a DS mouse model, which was replicated by brain region-specific ablation of NaV1.1, a sodium channel affected in the majority of patients with DS.[11] NaV1.1 expression has been shown to have a predominantly cell type–specific expression pattern in the developing neocortex and is clustered to the initial axon segments of PV-INs. Computational modeling of sodium channel dysfunction in inhibitory INs alone yielded a similar decrease in theta–gamma coupling, highlighting its significant role in the emergence of the biomarker in

this study. More interestingly, the biomarker only persisted in animals with the spontaneous seizures.

Identification of effective biomarkers will be critical for translating promising therapies for DS to the clinic. On the immediate horizon is the lure of the clinical use of precision medicine such as ASO therapy for severe monogenic epilepsies but particularly DS given the progress made by Stoke Therapeutics with promising preclinical results in a mouse model of DS,[1] and NaV1.1 activation using selective targeting by peptides derived from spider venom.[12] The recent protocols with ASO therapeutics for spinal muscular atrophy, with initial repeated dosing titrated over weeks with maintenance dosing every few months for life, show efficacy for $\sim 60\%$ of the patients. Tailoring similar titration approaches for DS will require individualization for each patient by both developmental age and severity. Early life seizures that are common in DS can independently drive pathogenesis in the immature brain. Transitioning from precision medicines for spinal cord to central nervous system disorders related to monogenic syndromes will also depend on the known cell type–specific expression patterns of Nav1.1 in addition to the developmental maturation stage of the GABAergic system which may require the targeting of functionally distinct neuronal subtypes. It is therefore critical that biomarkers of prognostication and evaluation of treatment efficacy in DS are independent of the seizure burden alone. Notably, here the authors show that the biomarker of decreased theta–gamma coupling in juvenile DS mice makes its onset independent of seizure activity.

Moving forward, it seems logical to pursue identification of similar theta–gamma coupling biomarkers in patients with DS using cEEGs recorded overnight for sleep dysfunction, during different behavioral states (wake vs sleep), and for increases in seizure severities or the onset of new seizure phenotypes. Continuous electroencephalograms recorded during visual or motor tasks are commonly used for standard assessment of severity of cognitive deficits and sensory integration disorders in children and could be an additional avenue to identify similar EEG biomarkers during task engagement. The clinical EEG data will likely come from a cohort of much younger patients given the early life onset of seizures in DS compared to the ages represented by the data acquired and reported in this study. Although recording stable cEEGs from neonatal DS mice remains challenging, the preclinical research focus needs to target developmental ages where underlying circuit maturation more closely represents the symptomatic patient population.

The largest readily available resources of data sets that can be leveraged for rare disorders with epilepsy are cEEGs and neuroimaging. In the recent "Curing the Epilepsies" NIH conference held early in 2021, these 2 were identified as untapped resources that could help identify biomarker and phenotypes dictating severity in preparation for future interventional clinical trials for the rare neurological disorders community. Automated algorithms built into EEG acquisition software could help identify and quantify the complete or partial reversal of translationally validated EEG biomarkers in real time. When partnered with new technologies such as remote ambulatory cEEG, this approach may allow the evidence-based fine tuning of individualized titrations of future interventions in DS.

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References

1. Han Z, Chen C, Christiansen A, et al. Antisense oligonucleotides increase Scn1a expression and reduce seizures and SUDEP incidence in a mouse model of Dravet syndrome. *Sci Transl Med*. 2020;12(558):eaaz6100. doi:10.1126/scitranslmed.aaz6100
2. Stoke Therapeutics. *Stoke Therapeutics Presents Data Showing Single Dose of ASO Therapy Restores Normal Protein Levels in Animal Model of Genetic Epilepsy – Stoke Therapeutics (Press Release)*; 2018.
3. Tran CH, Vaiana M, Nakuci J, et al. Interneuron desynchronization precedes seizures in a mouse model of Dravet syndrome. *J Neurosci*. 2020;40(13):2764-2775. doi:10.1523/JNEUROSCI.2370-19.2020
4. Cho KKA, Davidson TJ, Bouvier G, Marshall JD, Schnitzer MJ, Sohal VS. Cross-hemispheric gamma synchrony between prefrontal parvalbumin interneurons supports behavioral adaptation during rule shift learning. *Nat Neurosci*. 2020;23(7):892-902. doi:10.1038/s41593-020-0647-1
5. Sullivan BJ, Ammanuel S, Kipnis PA, Araki Y, Huganir RL, Kadam SD. Low-dose perampanel rescues cortical gamma dysregulation associated with parvalbumin interneuron GluA2 upregulation in epileptic syngap1+/− mice. *Biol Psych Neuro*. 2020;87(9):829-842. doi:10.1016/j.biopsych.2019.12.025
6. Fuchs EC, Doheny H, Faulkner H, et al. Genetically altered AMPA-type glutamate receptor kinetics in interneurons disrupt long-range synchrony of gamma oscillation. *Proc Natl Acad Sci U S A*. 2001;98(6):3571-3576. doi:10.1073/pnas.051631898
7. Kraft AW, Mitra A, Rosenthal ZP, et al. Electrically coupled inhibitory interneurons constrain long-range connectivity of cortical networks. *Neuroimage*. 2020;215:116810. doi:10.1016/j.neuroimage.2020.116810
8. Magloire V, Mercier MS, Kullmann DM, Pavlov I. GABAergic interneurons in seizures: investigating causality with optogenetics. *Neuroscientist*. 2018;25(4):344-358. doi:10.1177/1073858418805002
9. Kilb W. Development of the GABAergic system from birth to adolescence. *Neuroscientist*. 2012;18(6):613-630. doi:10.1177/1073858411422114
10. Wulff P, Ponomarenko AA, Bartos M, et al. Hippocampal theta rhythm and its coupling with gamma oscillations require fast inhibition onto parvalbumin-positive interneurons. *Proc Natl Acad Sci U S A*. 2009;106(9):3561-3566. doi:10.1073/pnas.0813176106
11. Jansen NA, Perez C, Schenke M, et al. Impaired θ-γ coupling indicates inhibitory dysfunction and seizure risk in a Dravet syndrome mouse model. *J Neurosci*. 2021;41(3):524-537. doi:10.1523/JNEUROSCI.2132-20.2020
12. Richards KL, Milligan CJ, Richardson RJ, et al. Selective NaV1.1 activation rescues Dravet syndrome mice from seizures and premature death. *Proc Natl Acad Sci U S A*. 2018;115(34):E8077-E8085. doi:10.1073/pnas.1804764115