Current Literature

In Clinical Science

Finding the Missing Pieces: The Microdeletion Burden in GGE

Burden Analysis of Rare Microdeletions Suggests a Strong Impact of Neurodevelopmental Genes in Genetic Generalised Epilepsies.

Lal D, Ruppert AK, Trucks H, Schulz H, de Kovel CG, Kasteleijn-Nolst Trenité D, Sonsma AC, Koeleman BP, Lindhout D, Weber YG, Lerche H, Kapser C, Schankin CJ, Kunz WS, Surges R, Elger CE, Gaus V, Schmitz B, Helbig I, Muhle H, Stephani U, Klein KM, Rosenow F, Neubauer BA, Reinthaler EM, Zimprich F, Feucht M, Möller RS, Hjalgrim H, De Jonghe P, Suls A, Lieb W, Franke A, Strauch K, Gieger C, Schurmann C, Schminke U, Nürnberg P; EPICURE Consortium, Sander T. *PLoS Genet* 2015;11:e1005226. doi:10.1371/journal.pgen.1005226.

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 ($P = 1.8 \times 10^{-7}$; 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 ($P = 2.6 \times 10^{-17}$) relative to controls. Microdeletions affecting either a gene previously implicated in neurodevelopmental disorders ($P = 8.0 \times 10^{-18}$, OR = 4.6) or an evolutionarily conserved brain-expressed gene related to autism spectrum disorder ($P = 1.3 \times 10^{-12}$, 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.

Commentary

The genetic generalized epilepsies (GGE) are the most common group of genetic epilepsies and include several distinct epilepsy syndromes such as juvenile myoclonic epilepsy, childhood absence epilepsy, and epilepsy with generalized tonic-clonic seizures, among others (1). Epidemiologic studies show an increased risk to first-degree relatives of individuals with GGE (2), and twin studies confirm an increased concordance rate for monozygotic compared with dizygotic twins (3). Classic studies such as these long ago confirmed a strong genetic contribution to the etiology of GGE, but the specific genetic or genomic variants that act as risk factors for or causes of GGE remained elusive for many decades.

A major advance in understanding the genetics of GGE came with the introduction of new technologies to scan the human genome for copy number variants (CNVs). CNVs are deletions or duplications of a stretch of chromosomal DNA and can vary in size from a few kilobases to many megabases. While many CNVs are benign variants found in healthy individuals, some increase risk for or cause disease. Array comparative genomic hybridization and single nucleotide polymorphism array technologies can be used for genome-wide detection of CNVs, and both technologies are widely used in research and clinical labs.

Previous studies to identify CNVs in epilepsy identified three specific deletions that are risk factors for GGE: a 1.5-Mb deletion at 15q13.3, a 600-kb deletion at 15q11.2, and a 800-kb deletion at 16p13.11 (4–7). These CNVs occur (and reoccur) at regions of the genome that are referred to as “hotspots” because they are susceptible to deletion and duplication. Each of these three hotspot deletions is found in 0.5 to 1% of patients with GGE. Of importance, each CNV is also a risk factor

for other conditions, including intellectual disability, autism spectrum disorder, and schizophrenia (8), which can be comorbidities with epilepsy.

A recent study by Lal and colleagues looked at the overall burden of rare microdeletions in a large cohort of patients with GGE compared with a control cohort. They specifically focused on deletions that are large (>400 kb) and rare (present in <1% of the study group) and included both hotspot and non-hotspot deletions in their analysis.

First, the authors compared the overall burden (or frequency) of large, rare deletions in the cases compared with a control cohort. Compared with 4% of controls, 7.3% of individuals with GGE carried at least one microdeletion, and the median size of microdeletions in cases was larger than those in control individuals. This excess of deletions in affected individuals supports the hypothesis that CNVs—specifically deletions—play a significant role in the genetic etiology of GGE.

Notably, more than one-third of the deletions in the GGE cohort were one of seven recurrent microdeletions, each of which is known to be associated with one or more neurodevelopmental disorders (8). Compared with controls, there was an impressive 7.5-fold excess of these recurrent deletions. Not surprising, deletions of 15q11.2, 15q13.3, and 16p13.11 accounted for 30 of 38 hotspot deletions; the overall frequency of these three deletions (2.2% of the cohort) was similar to that found in previous studies. In addition, deletions of 1q21.1 and 16p11.2 were present in one patient each, and deletions of 22q11.2 and 16p12 in three patients each, with the three 22q11 deletions representing a statistically significant enrichment.

Many CNVs contain one or more genes, any of which could be new candidate genes for GGE. The authors, therefore, went on to investigate the genes within the deletions in their cohort. First, they evaluated the genes affected by deletions that were seen in cases but were never observed in the controls. The genes within these deletions were enriched for known epilepsy-associated genes as well as genes associated with other neuropsychiatric disorders, confirming the notion that individual genes within CNVs are likely contributing to the genetic risk. Of interest, one of the epilepsy-only deletions contained the *PCDH7* gene, which has been implicated as a candidate gene by genome-wide association study (9).

Finally, given that there is an increasingly recognized relationship between the genetic etiologies of epilepsy and other neurodevelopmental disorders, the authors tested whether the CNVs in patients with epilepsy were more likely to contain genes that have been implicated in neurodevelopmental disorders than the CNVs found in controls. Indeed, deletions containing at least one neurodevelopmental or autism gene were >4-fold enriched in individuals with GGE, further supporting the role for some genes in many (related) disorders.

This large case-control study clearly highlights the role of large, rare microdeletions in genetic risk for GGE and further emphasizes the shared genetic risk among neurodevelopmental disorders. The results support CNV testing in individuals with GGE, especially those with comorbid features or a family history of related neurodevelopmental disorders. Additional

studies to understand the risk associated with smaller deletions, rare duplications, CNVs affecting conserved, noncoding regions, and eventually more common CNVs will certainly lend additional insight and identify novel candidate genes.

by Heather C. Mefford, MD, PhD

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