DE NOVO MISSENSE VARIANTS IN GNAI1 GENE ARE ASSOCIATED WITH EPILEPTIC ENCEPHALOPATHY

Mingjuan Liu, PhD; Lindsay Rhodes, MS, CGC; Holly Dukter, MD, LGCG; Ethan Goldberg, MD, PhD; Lia Zitano, MS, CGC; Cecile Rupp, MD, FACMG; Junqi Chen, MS, CGC; Klaas J. Winkens, MD; Gabriela Paranzo, MD; Soledad N. Wilson, MD, PhD; Jose Martinez, MD; Amy Duck, MS, CGC; Ingrid M. Vortmenn, MD, FACMG; Rhonda E. Schnur, MD; Kristin Manahan, PhD, FACMG; Jane Jussutu, PhD, FACMG

*GeneDx, Gaithersburg, MD; Department of Pediatrics, Division of Neurology, Children’s Hospital of Philadelphia, Philadelphia, PA; *Spectrum Health Medical Genetics, Grand Rapids, MI; Department of Pediatrics, Section of Genetics, University of Oklahoma Health Sciences Center, Oklahoma City, OK; Department of Neurology, Section of Child Neurology, University of Oklahoma Health Sciences Center, Oklahoma City, OK; Texas Tech Health Science Center, Lubbock and KristenGenome Medical Genetics, Dallas

1Department of Pediatrics and Adolescent Medicine, Division of Genetics, University of South Alabama, Mobile AL

Background

- Epileptic encephalopathies are a genetically heterogeneous group of disorders associated with frequent seizures and developmental delay or regression, and exome sequencing (ES) has led to the identification of novel genes associated with these disorders (McTague et al., 2016)
- Guanine-binding proteins (G proteins) are a family of signal transducing molecules which function as heterotrimeric consisting of alpha, beta and gamma subunits. The alpha subunit binds the guanine nucleotide, is capable of hydrolyzing GTP, and interacts with specific receptor and effector molecules (Sullivan et al., 1986)
- The GNAI1 gene encodes the alpha subunit of subtype 1 inhibitory G protein, a member of the G protein family and the inhibitory GTP-binding regulator of adenylate cyclase (Sullivan et al., 1986; Bray et al., 1987)
- Disruption of GNAI1 was reported to cause long term memory defects in mice (Pineda et al., 2004). One de novo heterozygous variant in GNAI1 affecting a residue of the GDP/GTP binding site (p.Gln204Arg) has been previously identified as a variant of interest in an individual with autism (Turner et al., 2016)
- Multiple missense variants clustering in a region involved in GDP/GTP binding in the GNAI1 gene, which encodes the alpha subunit of subtype 3 inhibitory G protein, have been previously reported to cause auriculocondylar syndrome, likely through a dominant negative effect (Rieder et al., 2012; Romanelli et al., 2015)

Methods

- Exome sequencing was performed on 9,659 patients with developmental delay or intellectual disability and their parents (when available)
- The sequencing methodology and variant interpretation protocol has been previously described (Tanaka et al., 2015)
- Identified sequence changes of interest were confirmed in all members of the family on whom biospecimens were available by di-deoxy DNA sequence analysis

Results

- Four de novo heterozygous missense variants in GNAI1 affecting two residues were identified in five individuals from unrelated proband-parent trios
- The five probands were male, between 3-11 years of age, with developmental delay and intellectual disability
- Four probands were reported with seizures of varying types, including infantile spasms, generalized tonic-clonic, focal onset, and complex partial seizures
- All variants were absent from the ExAC database as well as from our internal ES database of unaffected individuals
- All of our variants occurred at highly conserved residues 40 or 48 (p.Gly40Cys, p.Gly40Arg, p.Gly40Lys, and p.Gly40Thr). They are located within or in the proximity of the conserved GDP/GTP binding motif in the GNAI1 protein (Gi1). The Thr48 residue has been shown to be the guanine nucleotide binding site based on Gi1 crystal structure. Alteration of amino acid at 40 or 48 may disrupt GDP/GTP binding and impact catalytic function of Gi1
- Two Gly40 variants (p.Gly40Cys/c.118G>T and p.Gly40Arg/c.118G>C) variants affected a highly conserved nucleotide position located in the end of exon 1 and are predicted to damage the splice donor site of intron 1, which may alter gene splicing

Table 1. Clinical Presentation of Patients with heterozygous de novo GNAI1 variants

<table>
<thead>
<tr>
<th>Case</th>
<th>GNAI1 Variant</th>
<th>Developmental Delay</th>
<th>Seizures</th>
<th>Autism</th>
<th>Hypotonia</th>
<th>Brain abnormalities</th>
<th>Head Circumference</th>
<th>Short Stature</th>
<th>Dysmorphic Features</th>
<th>Other</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>p.Thr48Lys</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>unknown</td>
<td>microcephaly</td>
<td>-</td>
<td>+</td>
<td>bilateral inguinal hernia and cryptorchidism</td>
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<tr>
<td></td>
<td>(c.143 C&gt;A)</td>
<td></td>
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<td></td>
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<tr>
<td>2</td>
<td>p.Thr48Lys</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>cerebral atrophy</td>
<td>microcephaly</td>
<td>-</td>
<td>none reported</td>
<td>mild hip dysplasia</td>
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<tr>
<td></td>
<td>(c.143 C&gt;A)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>bitemporal malformation of cortical development</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>3</td>
<td>p.Gly40Thr</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>normal MRI</td>
<td>microcephaly</td>
<td>-</td>
<td>none reported</td>
<td>bilateral clubfeet, limb reductions due to amniotic band syndrome</td>
</tr>
<tr>
<td></td>
<td>(c.143 T&gt;C)</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>4</td>
<td>p.Gly40Arg</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>unknown</td>
<td>within normal limits</td>
<td>+</td>
<td></td>
<td>hyperphagia, overweight</td>
</tr>
<tr>
<td></td>
<td>(c.118 G&gt;C)</td>
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</table>

Case #5 (p.Gly40Cys; c.118 G>T) had overlapping clinical features with the other cases but was not complete enough to present.

Conclusions

- All identified variants in the GNAI1 gene occurred at highly conserved codons 40 or 48
- Similar to the previously described GNAQ3 gene, all GNAI1 variants lie in or in close proximity to the GDP/GTP binding site, and might interfere with GDP/GTP binding of Gi1, presumably leading to a dominant negative effect in Gi1
- Germline variants in GNAI1 may be associated with a neurodevelopmental disorder characterized by developmental delay, seizures, and hypotonia

References