Using Exome Data to Identify Malignant Hyperthermia Susceptibility Mutations
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ABSTRACT

Background: Malignant hyperthermia (MHS) is a life-threatening, inherited disorder of muscle calcium metabolism, triggered by anesthetics and depolarizing muscle relaxants. An unselected cohort was screened for MHS mutations using exome sequencing. The aim of this study was to pilot a strategy for the RYR1 and CACNA1S genes.

Design: Multidisciplinary analysis of gene variants identified through exome sequencing.

Methods: Exome sequencing was performed on 870 volunteers not ascertained for MHS. Variants in RYR1 and CACNA1S were annotated using an algorithm that filtered results based on mutation type, frequency, and information in mutation databases. Variants were scored on a six-point penetrance scale. Medical histories and pedigrees were reviewed for malignant hyperthermia and related disorders.

Results: The authors identified 70 RYR1 and 53 CACNA1S variants among 870 exomes. Sixty-three RYR1 and 41 CACNA1S variants passed the quality and frequency metrics but the authors excluded synonymous variants. In RYR1, the authors identified 65 missense mutations, one nonsense, two that affected splicing, and one non-frameshift indel. In CACNA1S, 48 missense, one frameshift deletion, one splicing, and one non-frameshift indel were identified. RYR1 variants predicted to be pathogenic for MHS were found in three participants without medical or family histories of MHS. Numerous variants, previously described as pathogenic in mutation databases, were reclassified by the authors as being of unknown pathogenicity.

Conclusions: Exome sequencing can identify asymptomatic patients at risk for MHS, although the interpretation of exome variants can be challenging. The use of exome sequencing in unselected cohorts is an important tool to understand the prevalence and penetrance of MHS, a critical challenge for the field.

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Display 1. Sequencing Coverage of the coding exons was 83% (RYR1) and 93% (CACNA1S).

Box and whisker plots showing base coverage for the RYR1 and CACNA1S genes for a cohort of 870 probands.

Display 2. Quality/ Frequency Filter Algorithm

Variants filtered on genotype quality, coverage and allele frequencies (Class 0-1). Variants assessed for pathogenicity (Class 2-5) on data in the Human Gene Mutation Database (HGMD) and locus-specific databases (LSDBs). MPG = most probable genotype. MAF = minor allele frequency. NHLBI ESP = The National Heart, Lung, and Blood Institute, exome sequencing project.

Display 3. Variant Pathogenicity Classification System

Criteria for assignment of pathogenicity class 1 to 5 for RYR1 and CACNA1S variants based on data available in the HGMD, LSDBs, family history and the European Malignant Hyperthermia Group’s (EMHG) list of diagnostic and non-pathogenic variants. VUS = variant of unknown significance.

Display 4. Frequency Histogram of RYR1 Variants

Frequency histogram of the 69 RYR1 variants with predicted protein changes from the ClinSeq® 870 cohort.

Display 5.

Pathogenicity of ClinSeq® RYR1 Variants Percent of Each Class

Clinical Action:

a. Results disclosure
b. Provide MH management guidelines
c. Refer proband to Specialists for CHCT testing and NHMRA Registry

CONCLUSIONS:

- While the interpretation and assessment of pathogenicity can be challenging, causative mutations can be filtered from ES data
- Clinically relevant mutations can be identified as secondary (so-called incidental) findings in exomes sequenced for clinical care and clinical research
- The application of ES technology to large and diverse cohorts has the potential to accelerate the pace of MHS gene mutation discovery