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# Novel Disease-Gene Discovery in the Epilepsies Using Diagnostic Exome Sequencing

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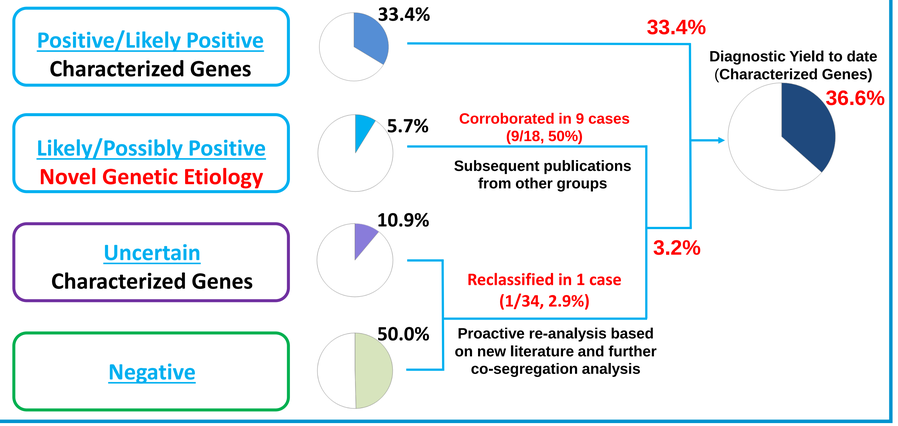
## BACKGROUND

- Although diagnostic exome sequencing (DES) has transformed the diagnosis and management of patients with neurological diseases, the diagnostic yield reported by clinical laboratories performing DES has rarely exceeded 25–30%.[1,2,3]
- Since DES can simultaneously interrogate virtually all coding genes in the genome, it provides unprecedented clinical and research opportunities for novel gene/genetic etiology (gene-disease association) discovery.
- As an integral part of family-centered DES, we have been performing novel genetic etiology analysis of proband-family trios since we introduced DES in October 2011.
- We demonstrate an enhanced diagnostic yield in epilepsy cases, emphasizing the clinical utility of reporting novel findings through DES.

## METHODS

- **Patients/study population:** Proband-family trios from 314 probands with epilepsy referred to Ambry Genetics for diagnostic exome sequencing (DES) are included in this study. Informed consent was obtained from all family members involved in the testing process.
- **Diagnostic exome sequencing and analysis:** Genomic deoxyribonucleic acid (gDNA) was isolated from whole blood from probands and first degree relatives. Samples were prepared using SureSelect Target Enrichment System (Agilent Technologies, Santa Clara, CA) or SeqCap EZ VCRome 2.0 (Roche Nimblegen, Madison, WI). The enriched exome libraries were sequenced using paired-end, 100-cycle chemistry on the Illumina HiSeq 2000 or 2500 (Illumina, San Diego, CA). Data Analysis and interpretation were performed as previously described. [3]
- **Evaluation of the Molecular Basis of Disease (EMBoDy):** We extensively evaluated variants of novel genetic etiology in proband-family trios based on critical and highly stringent assessments at both the gene and variant(s) level .
- **Proactive re-analysis:** We maintain an internal database of characterized genes, which we update on a weekly basis with the latest literature. When a new gene is added to the database, we review and perform clinical correlations on all previous cases that had rare variants detected within the gene.[3]

## DIAGNOSTIC YIELD AND NOVEL GENE RESULTS



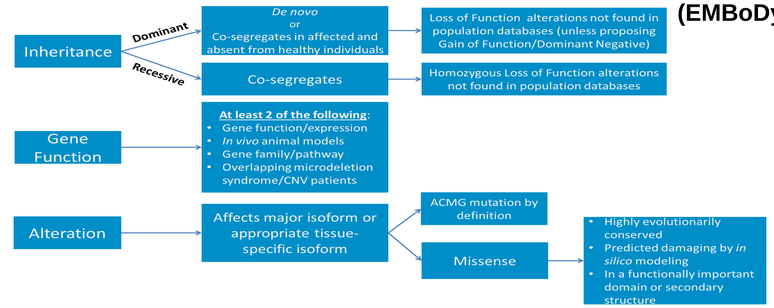

## CASES (N=314)

| EPILEPSY DIAGNOSES | N (%) |
|---|---|
| Epileptic encephalopathies | 89 (28.3) |
| Fever related seizures | 27 (8.6) |
| Focal epilepsies | 41 (13.1) |
| Generalized epilepsies | 25 (8.0) |
| Symptomatic seizures | 3 (1.0) |
| Single unprovoked seizure | 5 (1.6) |
| Benign familial neonatal seizures | 1 (0.3) |
| Unclassifiable epilepsy | 123 (39.2) |

## RESULTS

- Our diagnostic rate in this epilepsy cohort was 33.4%(105/314) among characterized genes.
- 200 cases underwent analysis for novel genetic etiologies and for 9.0% (18/200) a novel genetic etiology was identified.
- Following our reporting, the novel genes in 50% of the cases were corroborated in subsequent, independent publications from other groups, in less than 7 months on average.
- Half of the identified novel genetic etiologies were found in cases with a clinical indication of epileptic encephalopathy.
- Ongoing re-analysis of initially uncertain results has led to reclassification in 1 case thus far.

## EVALUATION OF THE MOLECULAR BASIS OF DISEASE (EMBoDy)




## SELECTED NOVEL GENETIC ETIOLOGIES

| GENE | *COQ4* | *DNM1* | *PURA* | *CLTC* | *OGT* | *SNAP25* | *SON* | *SV2A* |
|---|---|---|---|---|---|---|---|---|
| **TIME BETWEEN REPORTING AND FIRST INDEPENDENT PUBLICATION (in weeks)** | 16.0 | 0.8 | 37.4 | 29.7 | 5.9 | 124.0 | 29.9 | 63.3 |

## TAKE-HOME POINTS

- Novel genetic etiology findings accounted for ~8% of the total diagnostic yield from DES in this epilepsy cohort, underscoring the importance of reporting novel findings.
- The 50% corroboration rate in our cohort thus far (within 7 months on average) demonstrates that overall clinical utility outweighs uncertainty for reporting novel findings.
- Analysis of novel genetic etiologies may be particularly revealing in the epileptic encephalopathies.

## REFERENCES

1. Yang *et al.* JAMA. 2014. 2. Lee *et al.* JAMA. 2014. 3. Farwell *et al.* Genet Med. 2014.

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