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Clinical Utility of a Sponsored Gene Panel Testing Program for Pediatric Epilepsy and CLN2 Disease Diagnosis: Results from 14,589 Tests

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Introduction

- Epilepsy is a common childhood neurological disorder[1]
- More than 50% of pediatric-onset seizures have a genetic basis. However, many epilepsies are still diagnosed based on seizure semiology (+/- EEG) and not with molecular genetic testing[2]
- Epilepsy gene panels may uncover the etiology of pediatric seizures and expedite the time to treatment[2]
- Genetic testing may: impact clinical management (e.g., choice of anti-epileptic drugs, targeted therapy), shorten the diagnostic journey, prevent unnecessary testing, lead to clinical trial enrollment opportunities, or facilitate genetic counseling/family planning
- Behind the Seizure® (BTS, www.invitae.com/en/behindtheseizure/) is a sponsored, no-cost-to-patient testing program which provides a panel of 180+ genes associated with both syndromic and non-syndromic causes of epilepsy (Invitae Epilepsy Panel) and includes the option to add on preliminary-evidence genes
- CLN2 disease, one form of neuronal ceroid lipofuscinosis (NCL), is caused by deficient TPP1 activity and commonly presents non-specifically with seizures and a history of language development delay at 2–4 years of age[3]
- CLN2 disease diagnoses occur on average at 5 years old: a full 2 years after average age of seizure onset and after significant neurodegeneration has occurred[3,4]
- Our objective was to determine whether the BTS program can decrease the age of diagnosis for patients with CLN2 disease

Methods

- Data from BTS program tests conducted between December 2016 and April 2021 are presented
- Individuals were eligible for testing through BTS if they were aged 24–60 months with unprovoked seizure onset at/after 24 months (Dec 2016 to Feb 2019) or, following program expansion, aged 0–60 months (Feb 2019 to Jan 2020) or 0–108 months (Jan 2020 to Apr 2021) with unprovoked seizures onset at any age
- Variants were classified according to ACMG standards[5]:
  - Pathogenic (PATH), Likely Pathogenic (LPATH), Variant of Uncertain Significance (VUS), Benign (BEN), Likely Benign (LBEN)
- Outcome groups: data were divided into 3 groups by outcome

Outcome Group	Description
No MDx	No molecular diagnosis identified
All MDx	Any molecular diagnosis in a gene included in the Invitae Epilepsy Panel
CLN2 MDx	Molecular diagnosis of CLN2 disease (biallelic *TPP1* variants, PATH or LPATH)

BTS Data Collected
▪ Patient Age ▪ Physician Suspicion of Genetic Basis ▪ Medical History

- Suspicion of genetic basis of epilepsy and medical history were optional on the requisition form
  - All proportions calculated based on these data considered only orders where "Y" or "N" was selected— a blank item was not taken to be a negative

Results

- Between December 2016 and April 2021, a total of 14,589 tests were conducted through the BTS program: the molecular diagnostic yield was 13.3% overall (n=1946; 142 genes) and 0.16% for *TPP1* (n=24) **(Table 1)**

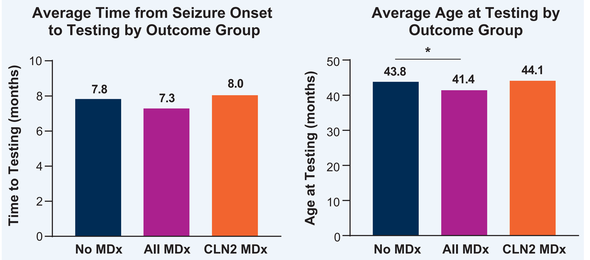
Table 1. Frequent Molecular Diagnoses

Gene	Inheritance	Conditions	Number of Diagnoses[a]
PRRT2	AD	Episodic kinesigenic dyskinesia (EKD); Benign familial infantile seizures (BFIS); Familial infantile convulsions with paroxysmal choreoathetosis (ICCA)	291
SCN1A	AD	Febrile seizures; Genetic epilepsy with febrile seizures plus (GEFS+); Dravet syndrome; Intractable childhood epilepsy with generalized tonic-clonic seizures (ICE-GTC); Familial hemiplegic migraine 3 (FHM3)	251
KCNQ2	AD	Benign familial neonatal seizures (BFNS); Early infantile epileptic encephalopathy (EIEE)	187
SCN2A	AD	Benign familial neonatal-infantile seizures (BFNIS); Early infantile epileptic encephalopathy (EIEE); Episodic ataxia; Intellectual disability (ID); Autism spectrum disorder (ASD)	59
MECP2	XL	X-linked Rett syndrome/atypical Rett syndrome; X-linked MECP2 duplication syndrome	58
PCDH19	XL	Early infantile epileptic encephalopathy (EIEE)	56
UBE3A	AD	Angelman syndrome	52
DEPDC5	AD	Familial focal epilepsy with variable foci (FFEVF); Nocturnal frontal lobe epilepsy (ADNFLE)	50
TSC2	AD	Tuberous sclerosis complex	49
NPRL3	AD	Familial focal epilepsy with variable foci (FFEVF)	48
SLC2A1	AD/AR	Glucose transporter type 1 deficiency syndrome (Glut1 DS)	44
CACNA1A	AD	Early infantile epileptic encephalopathy (EIEE); Episodic ataxia type 2 (EA2); Familial hemiplegic migraine type 1 (FHM1); Spinocerebellar ataxia 6 (SCA6)	42
GABRB3	AD	Early infantile epileptic encephalopathy (EIEE)	42
CDKL5	XL	Early infantile epileptic encephalopathy (EIEE)/West syndrome; Atypical Rett syndrome; Angelman-like syndrome	38
STXBP1	AD	Early infantile epileptic encephalopathy (EIEE)	38
GRIN2A	AD	Epileptic encephalopathies, typically presenting as an epilepsy-aphasia syndromes (EAS)	33
SYNGAP1	AD	Intellectual disability (ID); Early infantile epileptic encephalopathy (EIEE)	33
TSC1	AD	Tuberous sclerosis complex	33
CHD2	AD	Childhood-onset epileptic encephalopathy	30
GABRG2	AD	Childhood absence epilepsy (CAE); Generalized epilepsy with febrile seizures plus (GEFS+); Familial febrile seizures; Epileptic encephalopathy	27
KCNT1	AD	Nocturnal frontal lobe epilepsy (ADNFLE); Early infantile epileptic encephalopathy (EIEE)	27
SCN8A	AD	Early infantile epileptic encephalopathy (EIEE)	25
TPP1	AR	Neuronal ceroid lipofuscinosis type 2 (CLN2)	24
SLC6A1	AD	Myoclonic-atonic epilepsy (MAE)	21
SCN1B	AD/AR	Generalized epilepsy with febrile seizures; Early infantile epileptic encephalopathy (EIEE)	16
ATP1A3	AD	Dystonia; Cerebellar ataxia, areflexia, pes cavus, optic atrophy and sensorineural hearing loss (CAPOS) syndrome; Alternating hemiplegia of childhood (AHC)	15
ATP1A2	AD	Familial hemiplegic migraine (FHM); Alternating hemiplegia of childhood (AHC)	13
WDR45	XL	Beta-propeller protein-associated neurodegeneratiton (BPAN); Early infantile epileptic encephalopathy (EIEE); Rett syndrome	13
ZEB2	AD	Mowat-Wilson syndrome (MOWS)	13
HNRNPU	AD	Early infantile epileptic encephalopathy (EIEE); Intellectual disability (ID)	12
KCNB1	AD	Early infantile epileptic encephalopathy (EIEE)	12
MBD5	AD	Intellectual disability (ID)	12
IQSEC2	XL	Intellectual disability (ID)	11
GPHN	AR	Molybdenum cofactor deficiency; GPHN-realted spectrum disorder including seizures, autism, and intellectual disability	10
STX1B	AD	Generalized epilepsy with febrile seizures plus (GEFS+)	10
ALDH7A1	AR	Pyridoxine responsive epilepsy	9
ARX	XL	Early infantile epileptic encephalopathy/West syndrome; Lissencephaly with ambiguous genitalia (XLAG)	9
FOXG1	AD	Congenital/atypical Rett syndrome	9
GLDC	AR	Glycine encephalopathy (GCE)	9
KCNA2	AD/AR	Early infantile epileptic encephalopathy (EIEE); Spastic paraplegia and ataxia	9
KCNH5	AD	Early infantile epileptic encephalopathy (EIEE)	9
KCNQ3	AD/AR	Beningn familial neonatal seizures (BFNS); Early infantile epileptic encephalopathy (EIEE)	9
SLC6A8	XL	Creatine transporter deficiency (CTD)	9
GABRA1	AD	Early infantile epileptic encephalopathy (EIEE); Childhood absence epilepsy (CAE); Juvenile myoclonic epilepsy	8
NEXMIF	XL	Mental retardation	8
SPTAN1	AD	Early infantile epileptic encephalopathy (EIEE)	8
TBC1D24	AD/AR	Early infantile epileptic encephalopathy (EIEE); DOORS syndrome; Familial infantile myoclonic epilepsy; Progressive myoclonic epilepsy; Nonsyndromic hearing loss	8
KANSL1	AD	Koolen-De Vries syndrome (KDVS)	7
PAFAH1B1	AD	PAFAH1B1-related lissencephaly/subcortical band heterotopia	7
SCN9A	AD/AR	Genetic epilepsy with febrile seizures (GEFS+); Primary erythromelalgia; Small fiber neuropathy (SFNP); Paroxysmal extreme pain disorder (PEXPD); Congenital indifference to pain (CIP)/Hereditary sensory and autonomic neuropathy (HSAN)	7
SMC1A	XL	Cornelia de Lange syndrome (CDLS); Early infantile epileptic encephalopathy (EIEE); Holoprosencephaly (HPE)	7
DYRK1A	AD	Intellectual disability (ID)	6
EHMT1	AD	Kleefstra syndrome	6

Table shows genes identified in >5 MDx cases
[a]Individuals with a molecular diagnosis associated with more than one gene are counted for each gene

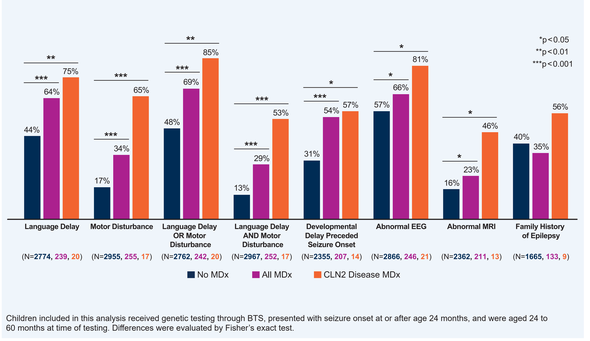
- In the subset of individuals tested through BTS who were aged 24–60 months with seizure onset at or after 24 months (n=4165), the molecular diagnostic yield was 7.8% overall (n=324) and 0.6% for TPP1 (n=23)
- The average time from seizure onset to testing was slightly shorter for All MDx groups than the No MDx and CLN2 MDx groups, however differences were not statistically significant **(Figure 1)**
- Average age at testing was slightly, but statistically significantly, lower for the All MDx group than for the No MDx group; difference in age at testing between the No MDx and CLN2 MDx groups was not statistically significant **(Figure 1)**
- In the CLN2 MDx group, the average age at testing was 44.1 months: diagnosis of CLN2 disease in these individuals occurred approximately 1 year earlier than the reported average[3]

Figure 1. Time to Testing and Age at Testing



Children included in this analysis received genetic testing through BTS, presented with seizure onset at or after age 24 months, and were aged 24 to 60 months at time of testing. *Indicates p<0.05 by t-test.

Figure 2. Clinical Presentation by Outcome Group



Children included in this analysis received genetic testing through BTS, presented with seizure onset at or after age 24 months, and were aged 24 to 60 months at time of testing. Differences were evaluated by Fisher's exact test.

- Individuals in the All MDx and CLN2 MDx groups more often had language delay and/or motor disturbance, developmental delays preceding seizure onset, and abnormal MRI or EEG compared to the No MDx group **(Figure 2)**
- Family history of epilepsy was not identified as a good predictor of molecular genetic testing outcome (40% for No MDx, 35% for All MDx, 56% for CLN2 MDx) **(Figure 2)**

Conclusions

- Over the time period December 2016 to April 2021, a total of 14,589 tests were performed through the BTS program and 1946 individuals received a positive molecular diagnosis (overall diagnostic yield: 13.3%), including 24 children with biallelic *TPP1* pathogenic variants who were diagnosed with CLN2 disease
- The average age of diagnosis of CLN2 disease through the BTS program was approximately 1 year earlier than the natural history published average[3]
  - Among those diagnosed with CLN2 disease, the average time between seizure onset and genetic testing was 8 months, compared with 22.7 months from symptom onset to diagnosis reported in the natural history study
- Early diagnosis has significant clinical impact with respect to disease severity and can enable the earlier initiation of treatment that may slow clinical deterioration
- Increased rates of molecular diagnosis in patients with language delay and/or motor disturbance supports early genetic testing in these patients presenting with seizures
- Family history of epilepsy was not found to be a good predictor of genetic testing outcome

References

1. Camfield P, Camfield C. Incidence, prevalence and aetiology of seizures and epilepsy in children. *Epileptic Disord.* 2015;17(2):117-23.

2. Wang J et al. Epilepsy-associated genes. *Seizure* 2017;44:11-20.

3. Nickel M et al. Disease characteristics and progression in patients with late-infantile neuronal ceroid lipofuscinosis type 2 (CLN2) disease: an observational cohort study. *Lancet Child Adolesc Health* 2018;2(8):582–90.

4. Schulz A et al. Neuronal ceroid lipofuscinosis-2 (CLN2) natural history and path to diagnosis: international experts' current experience and recommendations on CLN2 disease, a type of Batten disease, resulting from TPP1 enzyme deficiency. *Eur J Paediatr Neurol.* 2015;19:S119.

5. Richards S et al. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet Med.* 2015;17(5):405.

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