Ambry Genetics®

Final Report: 8/5/2022

Ordered By
Medical Professional: Unknown, Unknown, MD
Client: Ambry

Additional Authorized Recipient:
Thai, Julia

Patient Name: **9570_9571, Test DEF**
Accession #: **00-247040**
AP2 Order #: 2033397
Birthdate: 09/29/2014
MRN #: N/A
Indication: Internal Testing

Specimen #: N/A
Specimen: Blood EDTA
Sex at Birth: F
Collected: 07/22/2022
Received: 07/26/2022

NeuropathySelect™: Analyses of 81 Genes Associated with Hereditary Neuropathy

SUMMARY

NEGATIVE: No Clinically Significant Variants Detected

RESULTS

Pathogenic Mutation(s): None Detected
Variant(s) of Uncertain Significance: None Detected
Gross Deletion(s)/Duplication(s): None Detected

INTERPRETATION

- No pathogenic mutations, variants of uncertain significance, or gross deletions or duplications were detected.
- **Risk Estimate:** low likelihood of variants in the genes analyzed contributing to this individual's clinical history.
- Genetic counseling is a recommended option for all individuals undergoing genetic testing.

Order Summary: The following products were included in the test order for this individual. Please Note: tests on hold and those that have been cancelled (including reflex testing steps cancelled due to a positive result in a preceding test) are excluded. For additional information, please contact Ambry Genetics.

- NeuropathySelect™ (Product Code 9570)
- PMP22 deletion/duplication (Product Code 9571)

Electronically Signed By Wendy Alcaraz, PhD, DABMGG, CGMBS, on 8/5/2022 at 9:21:11 AM

All content hereafter is supplemental information to the preceding report.

Genes Analyzed

(81 total): ***AARS, AIFM1, APOA1, ATL1, ATL3, ATP7A, BICD2, BSCL2, CHCHD10, DCTN1, DNAJB2, DNM2, DNMT1, DST, DYNC1H1, EGR2, FAM134B, FBXO38, FGD4, FIG4, FUS, GAN, GARS, GDAP1, GJB1, GNB4, GSN, HARS, HINT1, HSPB1, HSPB8, IGHMBP2, IKBKAP, INF2, KIF1A, LITAF, LMNA, LRSAM1, MARS, MFN2, MORC2, MPZ, MTMR2, NDRG1, NEFH, NEFL, NGF, NTRK1, OPTN, PDK3, PLEKHG5, PMP22, PRDM12, PRPS1, PRX, RAB7A, REEP1, SBF2, SCN10A, SCN11A, SCN9A, SETX, SH3TC2, SIGMAR1, SLC25A46, SLC52A2, SLC52A3, SLC5A7, SPG11, SPTLC1, SPTLC2, TARDBP, TFG, TRPV4, TTR, UBA1, VAPB, VCP, VRK1, WNK1 and YARS.***

Metrics and Coverage

Complete coverage data for this proband is available for download through AmbryPort or can be e-mailed by request.

All genes analyzed achieved 100% coverage at ≥10X for all nucleotides in the coding regions.

Assay Information

General Information: Peripheral neuropathies (PNP), also known as polyneuropathy disorders affecting a variety of peripheral nerve cells and fibers (motor, sensory, and autonomic), are a relatively common diverse group of diseases with an estimated prevalence of 5-8%. Clinically, this group of disorders presents most frequently with distal symmetric sensorimotor neuropathy. Heterogeneous clinical symptoms are observed depending upon involvement of sensory, motor, or autonomic nerve fiber impairment. Symptoms of PNP may include hypalgesia, heat and cold allodynia, dysesthesia, sensory ataxia, paresis, muscle atrophy, hypotonia, hypohidrosis and/or anhidrosis, bladder dysfunction, indigestion, cardiac arrhythmias and tachycardia, gastroparesis, urogenital dysfunction, and periodic pain. Hereditary forms of peripheral neuropathy include but are not limited to hereditary motor and sensory neuropathy, often referred to as Charcot-Marie-Tooth disease, distal hereditary motor neuronopathies, and small fiber neuropathies. Over 100 genes have been identified as associated with hereditary peripheral neuropathies with autosomal dominant, autosomal recessive, X-linked, and mitochondrial inheritance patterns observed.

Methodology: Genomic deoxyribonucleic acid (gDNA) is isolated from the patient's whole blood. Samples are prepared using the IDT xGen Exome Research Panel v1.0 (Integrated DNA Technologies). Each DNA sample is sheared, adaptor ligated, PCR-amplified and incubated with the exome baits. Captured DNA is eluted, and PCR amplified. Final quantified libraries are seeded onto an Illumina flow cell and sequenced using paired-end, 150 cycle chemistry on the Illumina NovaSeq. Initial data processing, base calling, alignments and variant calls are generated by various bioinformatics tools. Data is annotated with the Ambry Variant Analyzer tool (AVA), including, but not limited to, the following information: nucleotide and amino acid conservation, biochemical nature of amino acid substitutions, population frequency, and predicted functional impact. The following sites are used to search for previously described gene mutations and polymorphisms: The Human Gene Mutation Database (HGMD), the Online Mendelian Inheritance in Man (OMIM), the genome aggregation database (gnomAD), HapMap data, and online search engines (e.g., PubMed). Variants are filtered further by the bioinformatics pipeline based on likelihood of pathogenicity (Farwell, 2015). For example, alterations in the following categories are typically filtered out unless otherwise protected: non-coding changes, synonymous changes, and alterations with a high population allele frequency (>1%). Additional manual screening is performed by licensed genetic counselors using criteria obtained from Ambry's General Variant Classification Scheme (https://www.ambrygen.com/science/variant-classification) to further filter alterations that are unlikely to be classified as disease-causing. The exome is targeted and sequenced, but analysis is limited to only the set of genes listed in this report. All reportable findings undergo manual review by molecular geneticists using integrated genomics software (IGV) and undergo confirmation either by automated fluorescence dideoxy (aka "Sanger") sequencing or via coverage and alternate read ratios above established confidence thresholds (heterozygous calls with 40-65% variant allele frequency and >35x coverage, hemizygous and homozygous calls with 100% variant allele frequency and >35x coverage). In most cases, phase cannot be determined.

Analytical range: The **NeuropathySelect™** test targets detection of DNA sequence variants in 81 genes by Next-Generation sequencing of coding domains (excluding *INF2* c.1250-1560 in coding exon 7 and *PRDM12* c.1042-1077 in coding exon 5) and plus at least 6 bases into the 5' and 3' ends of all the introns. For *PDK3*, only the status of the c.473G>A alteration is analyzed and reported. For *LMNA*, only the status of the c.892C>T alteration is analyzed and reported. Approximately 75% of the bases are expected to have quality scores of Q30 or higher, which translates to an expected base-calling error rate of 1:1000, or an expected base-calling accuracy of 99.9%. Additionally, 97% and 98% of the exome will be covered at ≥20x and ≥10x, respectively. Coverage is sufficient to detect >98% and up to 99.7% of disease-causing mutations (LaDuca H, et al. (2017) *PLoS ONE* 12(2):e0170843).

Test Limitations and Disclaimer: This test was developed, and its performance characteristics were determined, by Ambry Genetics. It has not been cleared or approved by the US Food and Drug Administration. The FDA does not require this test to go through premarket FDA review. It should not be regarded as investigational or for research. This test should be interpreted in context with other clinical findings. This report does not represent medical advice. Any questions, suggestions, or concerns regarding interpretation of results should be referred to a genetic counselor, medical geneticist, or physician skilled in evaluating the relevant medical literature. This laboratory is certified under the Clinical Laboratory Improvement Amendments (CLIA) as qualified to perform high complexity clinical laboratory testing. The following types of mutations are detectable: nucleotide substitutions, small deletions, small insertions, and small indels. The overall coverage of each gene varies and each individual may have slightly different coverage yield. This assay is not intended to systematically detect and analyze gross deletions/duplications, gross rearrangements, deep intronic variations, long repeat sequences, portions of genes with highly homologous pseudogenes, repeat expansions, mutations involved in tri-allelic inheritance, mitochondrial genome mutations, epigenetic effects, oligogenic inheritance, or other unknown abnormalities. A negative result from the analysis cannot rule out the possibility that the tested individual carries a rare unexamined mutation or mutation in the undetectable region. Although molecular tests are highly accurate, rare diagnostic errors may occur. Possible diagnostic errors include sample mix-up, erroneous paternity identification, technical errors, clerical errors, and genotyping errors. Genotyping errors can result from trace contamination of PCR reactions, from rare genetic variants that may interfere with analysis, or from other sources. The clinical sensitivity of the test may vary widely according to the specific clinical and family history. Mutations in other genes or regions not analyzed by this panel can also give rise to similar clinical conditions.

Resources: The following references are used in variant analysis and classification when applicable for observed genetic alterations.

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Ambry Genetics®

Understanding Your Negative NeuropathySelect™ Genetic Test Result

INFORMATION FOR PATIENTS

Result	**NEGATIVE**	The result of your genetic testing did not find any mutations (changes) in genes that cause polyneuropathy (peripheral and/or autonomic) and/or hereditary transthyretin-mediated (hATTR) amyloidosis. Not all patients with polyneuropathy and/or hATTR amyloidosis have a mutation in a gene in this test. You may have a mutation in a gene that was not included in this test. If someone in your family has a specific mutation in one of the genes in this test, it is likely that you do not carry that mutation.
Diagnosis	**NO CHANGE**	This testing does not change your clinical diagnosis. If you have been diagnosed with polyneuropathy and/or amyloidosis, that remains the same.
Further Testing	**DISCUSS**	More genetic testing may be right for you. Please talk about this with your healthcare providers.
Medical Management Options	**PATIENTS WITH POLYNEUROPATHY AND/ OR AMYLOIDOSIS**	Treatment options may include: medications, supportive therapies, supportive devices, surgery and organ transplantation. Talk to your healthcare providers about which may be right for you.
Screening Options	**PATIENTS WITH A FAMILY HISTORY OF POLYNEUROPATHY AND/ OR AMYLOIDOSIS, BUT NO PERSONAL SIGNS**	Options for screening and early detection include: physical exams, imaging studies, laboratory tests, biopsies and specialized analysis (electromyography, nerve conduction velocity testing, etc.). Talk to your healthcare providers about whether these options are right for you.
Next Steps	**DISCUSS**	Even though your genetic testing was negative, some causes of polyneuropathy and/or amyloidosis can run in families. All close blood-related family members of someone with polyneuropathy and/or amyloidosis (like parents, brothers, sisters, children) should talk with their healthcare providers about screening
Reach Out	**RESOURCES**	• Amyloidosis Foundation amyloidosisresearchfoundation.org • National Society of Genetic Counselors nsgc.org • Canadian Association of Genetic Counsellors cagc-accg.ca • Genetic Information Nondiscrimination Act (GINA) ginahelp.org

Polyneuropathy and/or Amyloidosis in the Family

Even though your genetic testing was negative, some causes of polyneuropathy and/or amyloidosis can run in families. All close family members of someone with an inherited polyneuropathy and/or amyloidosis (like parents, brothers, sisters, children) should talk with their healthcare providers about screening.

Please discuss this information with your healthcare providers. The field of genetics is continuously changing, so updates related to your genetic testing results and/or, medical management options recommendations, and/or potential treatments may be available over time. This information is not meant to replace a discussion with your healthcare provider and should not be considered or taken as medical advice.