



Patient Name: SAMPLE	Provider Name: SAMPLE	Sample Type: Buccal
Patient Date of Birth: SAMPLE	Provider Phone: SAMPLE	Sample Collected: 07/15/2022
Patient Sex: Female	Provider Fax: SAMPLE	Sample Received: 07/18/2022
Patient ICD-10 code(s): F88, G93.40, F82, F80.1	Provider Institution: SAMPLE	Order Date: 07/15/2022
Accession Number: SAMPLE		Report Date: 09/26/2022

Test Result Summary: Pathogenic

Whole Exome Sequencing (WES) identified one pathogenic sequence variant. This variant is consistent with Phelan-McDermid syndrome.

Test performed: Trio (mother: SAMPLE; father: SAMPLE)

Patient phenotype utilized for analysis: Developmental delay, nonverbal, motor delay, poor coordination, learning issue, heart galloping, neonatal hypertension in her lung, persistent fetal lung and heart resolved, sleeping issue, and other phenotypes noted in the provided medical and family history.

Secondary Findings: Opt-In V3.1; Negative

Sequence Variant Results

Gene (Transcript)	DNA change	Protein Change	Classification	Zygosity	Parental Inheritance	Associated Syndrome (inheritance)
<i>SHANK3</i> (NM_033517.1)	c.SAMPLE	p.SAMPLE	Pathogenic	Heterozygous	<i>De novo</i>	Phelan-McDermid Syndrome (Autosomal dominant)

Clinical Interpretation and Discussion

***SHANK3* gene, NM_033517.1, c.SAMPLE, p.SAMPLE, pathogenic**

A pathogenic sequence variant in one copy (heterozygous) of the *SHANK3* gene was found. This sequence variant is consistent with Phelan-McDermid syndrome and is expected to result in an increased risk of the associated symptoms.

Genetic variants affecting one copy of the *SHANK3* gene cause autosomal dominant Phelan-McDermid syndrome (PMS; PMIDs: 20301377, 23758760, 25784960, 33949759, 34246244, 34325981). PMS is characterized by a wide range of symptoms including global developmental delay, moderate-to-profound intellectual disability, severely impaired or absent speech, hypotonia, brain malformations, sleep disturbances, and neurobehavioral differences (such as autism spectrum disorder or impulsivity). Seizures have been reported to affect up to half of individuals, with the most common seizure types including absence and grand mal (PMIDs: 20301377, 34664257). Mild dysmorphic facial features may include dolichocephaly,



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large or prominent ears, epicanthal folds, long eyelashes, supraorbital fullness, full cheeks, and a short or bulbous nose (PMID: 20301377). Developmental regression affecting speech, motor, and/or self-help skills has been reported. One study suggested that up to 43% of individuals experience a loss of skills, commonly with onset in mid-childhood (PMID: 28346892). There is also an increased risk of psychiatric conditions in adolescents and young adults with PMS, including bipolar disorder, anxiety, mood disorders, catatonia, and possibly schizophrenia (PMIDs: 20385823, 26824576, 31879555, 32050889). **There are published expert opinions on medical management for PMS (PMIDs: 20301377, 25784960).**

Loss of function of one gene copy (haploinsufficiency) of *SHANK3* has been suggested as the mechanism of pathogenicity for PMS (PMID: 20301377). The majority of affected individuals have a deletion involving at least the *SHANK3* gene, although approximately 3% of individuals have a *SHANK3* sequence variant (PMIDs: 23758760, 29719671, 30537371, 34737294). Individuals with PMS due to large deletions involving *SHANK3* are sometimes reported to have more severe clinical presentations, likely due to the inclusion of other genes within the deletion region (PMIDs: 23758760, 33949759, 34325981).

Most pathogenic *SHANK3* variants are reported to be *de novo* (not inherited); however, inheritance of a pathogenic *SHANK3* variant from a mildly affected parent (PMID: 29263841) or an unaffected parent with germline mosaicism has been reported (PMIDs: 15930901, 20301377, 29719671, 34737294). This reported *SHANK3* variant was not identified in the parental samples provided, indicating it is likely *de novo*, however, germline mosaicism cannot be ruled out.

This frameshift variant has been shown by functional studies to result in a truncated protein, lacking several crucial domains involved in mGluR and actin binding (Homer, AbPI, cortactin) and in the synaptic targeting and postsynaptic assembly of *SHANK3* multimer. When over-expressed in rat hippocampal neuronal cells, no synaptic localization was observed, consistent with previous mislocalization studies (PMIDs: 17173049, 21606927).

This variant has been reported as pathogenic by multiple submitters in ClinVar, a database that curates genetic information from individuals with clinical features (Accession: VCV000208759.18; PMID: 26582918). It has also been reported in multiple individuals with PMS, including at least four cases of *de novo* events (PMIDs: 17173049, 29719671, 30763456, 32050889, 33256793).

In summary, this finding is expected to increase the risk for symptoms of Phelan-McDermid syndrome. **There are published expert opinions on medical management (PMIDs: 20301377, 25784960).** Evaluation with a medical geneticist or discussion with a genetic counselor may be considered to determine if additional testing or assessments would be useful.

Secondary Findings

When performing genome or exome-scale sequencing, the American College of Medical Genetics (ACMG) recommends the interpretation and reporting of known (pathogenic) and likely pathogenic sequence variants in 78 genes (ACMG SF v3.1; PMID: 35802134). These particular genes have been selected because the discovery of a pathogenic/likely pathogenic sequence variant would immediately prompt significant medical management changes for the person being, some of which may be life-saving. Sequence variants in these genes are often unrelated to the reason for testing and are therefore called "secondary findings." Please note that a subset of these genes may be interpreted as part of NextStep^{Dx} PLUS WES based on their relationship to the specific patient's phenotype.

If one opts in for analysis of secondary findings, the additional ACMG recommended genes will be interpreted and reported. Only known pathogenic variants and likely pathogenic variants for secondary findings would be reported. The absence of any particular secondary finding reported in this test does not rule out the presence of a causative variant in any of these genes and does not guarantee that the individual tested will not develop any of the conditions or medical issues related to these genes. This is due in part to the variability of coverage: some exons have better coverage than others and average or typical coverage may not be achieved for each individual for each exon. Bionano Laboratories analyzes and reports on secondary



findings for the proband only. If a secondary finding is identified, parental follow up testing may be ordered. Please contact a Bionano Laboratories genetic counselor to discuss this testing at 801-931-6191.

References

The references in this report can be found by searching the PubMed IDs (PMID) from the PubMed home page: <http://www.ncbi.nlm.nih.gov/pubmed/>

For more information on how to use PubMed, see the following tutorial: <http://www.nlm.nih.gov/bsd/disted/pubmedtutorial/cover.html>

PubMed IDs referenced in this report:

ACMG Board of Directors. 2012. PMID: 22863877.
Miller, et al. 2021. PMID: 34012068.
Phelan K, Rogers RC, Boccuto L. 2018. PMID: 20301377
Soorya L, et al. 2013. PMID: 23758760
Kolevzon A, et al. 2014. PMID: 25784960
Ricciardello Z, Tomaiuolo P, Persico AM. 2021. PMID: 33949759
Burdeus-Olavarrieta M, et al. 2021. PMID: 34246244
Frank Y, et al. 2021. PMID: 34325981
Jain L, et al. 2022. PMID: 34664257
Reierson G, et al. 2017. PMID: 28346892
Gauthier J, et al. 2010. PMID: 20385823
Egger JIM, et al. 2016. PMID: 26824576
Kolevzon Z, et al. 2019. PMID: 31879555
Kohlenberg TM, et al. 2020. PMID: 32050889
De Rubeis S, et al. 2018. PMID: 29719671
Li Y, et al. 2018. PMID: 30537371
Loureiro LO, et al. 2021. PMID: 34737294
Tabet A-C, et al. 2017. PMID: 29263841
Landrum MJ, et al. 2016. PMID: 26582918
Durand CM, et al. 2007. PMID: 17173049
Durand CM, et al. 2012. PMID: 21606927
Zhou WZ, et al. 2019. PMID: 30763456
Xu N, et al. 2020. PMID: 33256793
The following references provide information about the utilization of whole exome sequencing:
Yang Y, et al. 2014. PMID: 25326635
ACMG Board of Directors. 2013. PMID: 23970068.

Genetic Counseling and Family Resources

A genetic test called whole exome sequencing (WES) was completed. WES looks for sequence variants, or spelling changes, in the DNA code across thousands of genes. These genetic changes may be responsible for a person's developmental and/or medical symptoms. Since everyone has sequence variants, the genetics community is continuously learning about what variants in which genes cause symptoms and which variants do not (called benign variants).

Bionano Laboratories genetic counselors are available by phone to speak with providers or the family about this test result. You can schedule a time to speak with a Bionano Laboratories genetic counselor by calling 801-931-6191.

A genetic counselor can help review what these results mean for an individual and family members, background on genetics, and discuss additional resources or next steps that may be helpful. Additionally, the genetic counselor may review medical,



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developmental, and family history of the person tested. This will help the genetic counselor better answer questions about how a specific result may impact the family. To best prepare for a genetic counseling session, it may be helpful for a family to create a list of questions. Additionally, it may help to review some information on basic genetic concepts such as what are genes and chromosomes. The following resource(s) may be helpful for families to review:

MedlinePlus: This website can be used to find information about genetic topics, genes, specific genetic conditions, and broad topics like autism spectrum disorder. It also provides links to other websites for more in-depth information about genetic conditions, patient support and advocacy resources, and relevant clinical trials. <https://medlineplus.gov/genetics>

MyGene2: This is a resource for connecting families with rare conditions to other families, clinicians, and researchers. <http://mygene2.org/MyGene2/>

Phelan-McDermid Syndrome Foundation: This organization supports families with relatives who have PMS. <http://www.pmsf.org>

Bionano Laboratories' Clinical Interpretation Process

Sequence variants identified by NextStep^{Dx} PLUS WES are analyzed with a comprehensive interpretation process. Guidelines from the American College of Medical Genetics and Genomics (ACMG) are applied to variants reported (PMID: 25741868). The interpretation process involves collaboration between experts including laboratory directors, variant analysts, and certified genetic counselors. This team completes an up-to-date review of the medical literature, patient databases (PMIDs: 26582918, 12754702) and control datasets (PMIDs: 24174537, 32461654). This process provides comprehensive information about the current evidence for genes and specific sequence variants within them. A phenotype-driven analysis of the submitted specimen is undertaken, focusing on genes associated with the provided clinical and family history information. The relevant phenotypes for this individual are listed on page 1 and a list of the genes analyzed is available upon request. The relevant evidence from this interpretation process is summarized in each report.

Methodology and Limitations

Genes tested: 20,096

Genes associated with patient phenotype: 2,920 genes, 99.26% at 20x, 99.52% at 10x.

Methodology: Genomic DNA was isolated from the submitted specimen indicated above (if cellular material was submitted). DNA was barcoded, and enriched for the coding exons of targeted genes using hybrid capture technology. Prepared DNA libraries were then sequenced using a Next Generation Sequencing technology. Following alignment to the human genome reference sequence (assembly GRCh37), variants were detected in regions of at least 10x coverage. Specific specimen coverage is provided at the beginning of this section. Variants were interpreted manually using locus specific databases, literature searches, and other molecular biological principles. To minimize false positive results, any variants that do not meet internal quality standards are confirmed by Sanger sequencing. Variants classified as pathogenic, likely pathogenic, unknown significance, or risk allele which are located in the coding regions and nearby intronic regions (+/- 20bp) of the genes listed above are reported. Variants outside these intervals may be reported but are typically not guaranteed. When a single pathogenic or likely pathogenic variant is identified in a clinically relevant gene with autosomal recessive inheritance, the laboratory will attempt to ensure 100% coverage of coding sequences either through NGS or Sanger sequencing technologies ("fill-in"). Deletion/duplication analysis has not been performed. New York patients: diagnostic findings are confirmed by Sanger, MLPA, or qPCR; exception SNV variants in genes for which confirmation of NGS results has been performed ≥ 10 times may not be confirmed if identified with high quality by NGS. Bioinformatics: The Fulgent Germline v2019.2 pipeline was used to analyze this specimen.

Limitations: All laboratory tests have limitations. These results assume that the specimen received in the laboratory belongs to the named individual and that no mixup or co-mingling of specimens has occurred. Positive results do not imply that there are no other pathogenic alterations in the patient's genome, and negative results do not rule out a genetic cause for the indication for testing. This assay assumes



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that any stated familial relationships are accurate. This assay is not designed or validated for the detection of somatic mosaicism or somatic mutations. This assay will not detect certain types of genomic aberrations which may cause disease such as, but not limited to, translocations or inversions. Result interpretation assumes that the human reference sequences are correct at the queried loci. Official gene names change over time. Fulgent uses the most up to date gene names based on HUGO Gene Nomenclature Committee (<https://www.genenames.org>) recommendations. If the gene name on report does not match that of ordered gene, please contact the laboratory and details can be provided. Result interpretation is based on the collected information available at the time of reporting; additional information may exist in the future which will not be represented. DNA alterations in regulatory regions or deep intronic regions (greater than 20bp from an exon for exome or 200bp for genome) will not be detected by this test. All sequencing technologies have limitations. There are technical limitations on the ability of DNA sequencing to detect small insertions and deletions. Our laboratory uses a sensitive detection algorithm, however these types of alterations are not detected as reliably as single nucleotide variants. Rarely, due to systematic chemical or computational issues, or human error, DNA variants may be missed. Repeat expansions (eg. trinucleotides or hexanucleotides) are not reliably detected by NGS, and pathogenic expansions will not be detected in disease-related genes, even if those genes are sequenced. Unless otherwise indicated, no other assay has been performed to evaluate the submitted specimen for repeat expansions. Although next generation sequencing technologies and our bioinformatics analysis significantly reduce the contribution of pseudogene sequences or other highly-homologous sequences, sometimes, these may still interfere with the technical ability of the assay to identify pathogenic variant alleles sequencing analyses.

Disclaimer: This test was developed and its performance characteristics determined by Fulgent Genetics CAP # 8042697 CLIA# 05D2043189; 4978 Santa Anita Ave., Suite 205, Temple City, CA 91780. It has not been cleared or approved by the FDA. The laboratory is regulated under CLIA as qualified to perform high-complexity testing. This test is used for clinical purposes. It should not be regarded as investigational or for research. Since genetic variation, as well as systematic and technical factors, can affect the accuracy of testing, the results of testing should always be interpreted in the context of clinical and familial data. It is recommended that patients receive appropriate genetic counseling to explain the implications of the test result, including its residual risks, uncertainties and reproductive or medical options. Fulgent CLIA Laboratory Director: Hanlin Gao, M.D.

Lineagen, Inc.'s (DBA Bionano Laboratories) reporting service is certified under CLIA (CLIA# 46D2042721). Bionano Laboratories, 2677 E Parleys Way Salt Lake City, Utah 84109, USA. T: 801-931-6200 F: 801-931-6201 www.bionanolaboratories.com

Lineagen CLIA Laboratory Director: Moises Serrano, Ph.D., DABMGG

Report Reviewed By: SAMPLE