Patient Information: Accession: Physician: Report Date: **Dec 04,2020**

Final Report

TEST PERFORMED

Diabetes-Obesity - 56 Genes
(56 Gene Panel; gene sequencing with deletion and duplication analysis)

RESULTS:

No clinically significant sequence or copy-number variants were identified which are sufficient for a molecular diagnosis.

However, one variant of potential clinical relevance is reported.

Clinically significant Variants
None

Additional Variants of Potential Clinical Relevance

Gene Info		Variant Info		
GENE	INHERITANCE	VARIANT	ZYGOSITY	CLASSIFICATION
BBS9 NM_198428.2	Autosomal Recessive	c.966G>A p.Trp322*	Heterozygous	**Likely Pathogenic**

INTERPRETATION:

Notes and Recommendations:

- As requested, this report only includes variants classified as Pathogenic, Likely Pathogenic, or Risk Allele at the time of analysis. If detected, this report does not include variants classified as of uncertain significance.
- Gene specific notes and limitations may be present. See below.
- These results should be interpreted in the context of this individual's clinical findings, biochemical profile, and family history.
- Genetic counseling is recommended. Available genetic counselors and additional resources can be found at the National Society of Genetic Counselors (NSGC; https://www.nsgc.org)

About *BBS9*

Biallelic mutations in *BBS9* have been associated with Bardet-Biedl syndrome-9 (BBS9), characterized by obesity, polydactyly, renal anomalies, retinopathy, and mental retardation (PubMed: 20301537; OMIM: 607968).

The gene product of the *BBS9* gene is a protein called Bardet-Biedl syndrome 9. See OMIM gene entry for *BBS9* (OMIM: 607968) for further information.

96% of the coding sequence of the NM_198428.2 transcript of*BBS9* gene was sequenced to a minimum depth of 20x in the submitted specimen. Neither a second sequencing mutation nor a copy number variant was detected in this gene; however, the presence of mutations in the deep intronic or regulatory regions cannot be ruled out. As the clinical condition(s) associated with mutations in the *BBS9* gene are recessive and only a single heterozygous variant has been detected, this result is interpreted as carrier status only. Further clinical evaluation may be warranted to clarify these findings.

BBS9 NM_198428.2:c.966G>A (p.Trp322*)

Classification: **Likely Pathogenic**

Zygosity and Inheritance	• This heterozygous Likely Pathogenic variant is consistent with this individual being a carrier for an autosomal recessive *BBS9*-related condition.
Variant Type	• Genomic change: Chr7(GRCh37):g.33313518G>A. • This variant is in the dbSNP database: rs928813600 • This variant is predicted to result in a stop gain (nonsense) in exon 9 of the*BBS9* gene.
Variant in Cases	• This nonsense variant introduces a premature stop codon and is expected to result in the loss of function of the protein product of the BBS9 gene, either as the result of protein truncation or of nonsense-mediated mRNA decay. This stop-gain variant occurs at least 50 nucleotides upstream of the penultimate exon and is consistent with the resulting transcript being targeted for nonsense mediated decay (PubMed: 27618451, 11532962, 18066079). or • While this truncating variant has not, to our knowledge, been reported in the literature, truncating variants upstream and/or downstream of this position have been reported to be pathogenic (PubMed: 20177705, 27894351, 16380913, 26766544). • This variant has not been reported as associated with a clinical condition in the Human Gene Mutation Database (HGMD).
Variant in Controls	• This variant has been observed at a frequency of less than 0.01% (3/282274 alleles). • The highest allele frequency that this variant has been observed at in any sub-population with available data is 0.01% in the Other population. • There are no homozygous control individuals for this variant. • The Broad Institute gnomAD database (>120,000 Individuals with no known severe, pediatric onset disease) was used for this analysis.

GENES TESTED:

Diabetes-Obesity - 56 Genes
56 genes tested (97.94% at >20x).

ABCC8, ADRB2, ADRB3, AGRP, ALMS1, ARL6, BBS1, BBS10, BBS12, BBS2, BBS4, BBS5, BBS7, BBS9, BDNF, CARTPT, CEL, CEP290, EIF2AK3, ENPP1, FOXP3, GCK, GHRL, GLIS3, GNAS, HNF1A, HNF1B, HNF4A, INS, KCNJ11, LEP, LEPR, MAGEL2, MC4R, MKKS, MKS1, NEUROD1, NEUROG3, NTRK2, PCSK1, PDX1, POMC, PPARG, PPARGC1B, PTF1A, PYY, RFX6, SDC3, SDCCAG8, SIM1, TRIM32, TTC8, UCP1, UCP3, WDPCP, WFS1

Gene Specific Notes and Limitations

CEL
Due to pseudogene interference and tandem repeats in exon 11 of *CEL*, the current testing method is not able to reliably detect the variable number of tandem repeats (VNTR) and/or certain pathogenic variants in this gene.

METHODS:

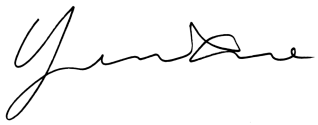
Genomic DNA was isolated from the submitted specimen indicated above (if cellular material was submitted). DNA was barcoded, and enriched for the coding exons of targeted genes using hybrid capture technology. Prepared DNA libraries were then sequenced using a Next Generation Sequencing technology. Following alignment to the human genome reference sequence (assembly GRCh37), variants were detected in regions of at least 10x coverage. For this specimen, 98.88% and 97.94% of coding regions and splicing junctions of genes listed had been sequenced with coverage of at least 10x and 20x, respectively, by NGS or by Sanger sequencing. The remaining regions did not have 10x coverage, and were not evaluated. 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, 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"). All genes listed were evaluated for large deletions and/or duplications. However, single exon deletions or duplications will not be detected in this assay, nor will copy number alterations in regions of genes with significant pseudogenes. Putative deletions or duplications identified by NGS are confirmed by an orthogonal method (qPCR or MLPA), unless exceeding an internally specified and validated quality score, beyond which deletions and duplications are considered real without further confirmation. 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:

These test results and variant interpretation are based on the proper identification of the submitted specimen, accuracy of any stated familial relationships, and use of the correct human reference sequences at the queried loci. In very rare instances, errors may result due to mix-up or co-mingling of specimens. Positive results do not imply that there are no other contributors, genetic or otherwise, to this individual's phenotype, and negative results do not rule out a genetic cause for the indication for testing. 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 available clinical and family history information for this individual, collected published information, and Alamut annotation available at the time of reporting. This assay is designed and validated for detection of germline variants only. It is not designed or validated for the detection of low-level mosaicism or somatic mutations. This assay will not detect certain types of genomic aberrations such as translocations, inversions, or repeat expansions (eg. trinucleotide or hexanucleotide repeat expansion). DNA alterations in regulatory regions or deep intronic regions (greater than 20bp from an exon) may not be detected by this test. Unless otherwise indicated, no additional assays have been performed to evaluate genetic changes in this specimen. 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, computational, or human error, DNA variants may be missed. Although next generation sequencing technologies and our bioinformatics analysis significantly reduce the confounding

contribution of pseudogene sequences or other highly-homologous sequences, sometimes these may still interfere with the technical ability of the assay to identify pathogenic alterations in both sequencing and deletion/duplication analyses. Deletion/duplication analysis can identify alterations of genomic regions which are two or more contiguous exons in size; single exon deletions or duplications may occasionally be identified, but are not routinely detected by this test. When novel DNA duplications are identified, it is not possible to discern the genomic location or orientation of the duplicated segment, hence the effect of the duplication cannot be predicted. Where deletions are detected, it is not always possible to determine whether the predicted product will remain in-frame or not. Unless otherwise indicated, deletion/duplication analysis has not been performed in regions that have been sequenced by Sanger.

SIGNATURE:



Yuan Xue, Ph.D., CGMB, FACMG on **12/04/2020 1:54 PM PST**
Electronically signed

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. For assistance with interpretation of these results, healthcare professionals may contact us directly at **800-516-1676**. 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.