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Patient Information:
18198, Donor
DOB: [redacted]
Sex: M
MR#:
Patient#: FT-PT8801264

Partner Information:
Not Tested

Physician:
Kuan, James
ATTN: Kuan, James
Denver Sperm Bank
4915 25th Avenue NE, Ste 204W
Seattle, WA 98105
Phone: (206) 588-1484

Laboratory:
Fulgent Therapeutics LLC
CAP#: 8042697
CLIA#: 05D2043189
Laboratory Director:
Lawrence M. Weiss, MD
Report Date: **Aug 20,2024**

Accession:
FT-7071683
Test#: FT-TS14908215
Specimen Type: Blood (EDTA)
Collected: Jul 30,2024

Accession:
N/A

FINAL RESULTS

Carrier for genetic conditions in **multiple** genes.
Genetic counseling is recommended.

TEST PERFORMED

Beacon Preconception Carrier Screening - 515 Genes (without X-linked Disorders)
(515 Gene Panel; gene sequencing with deletion and duplication analysis)

Condition and Gene	Inheritance	18198, Donor	Partner
Alpha thalassemia *HBA2*	AR	⊕ Carrier Whole Gene Deletion (αα/α-)	N/A
Spinal muscular atrophy *SMN1*	AR	⊕ Carrier Absence of SMN1	N/A
Mitochondrial DNA depletion syndrome 3 *DGUOK*	AR	⊕ Carrier c.3G>A (p.?)	N/A
Systemic primary carnitine deficiency *SLC22A5*	AR	⊕ Carrier c.131C>T (p.Ala44Val)	N/A
Wilson disease *ATP7B*	AR	⊕ Carrier c.3662G>A (p.Gly1221Glu)	N/A

INTERPRETATION:

Notes and Recommendations:

- Based on these results, this individual is positive for carrier mutations in 5 genes. Carrier screening for the reproductive partner is recommended to accurately assess the risk for any autosomal recessive conditions. A negative result reduces, but does not eliminate, the chance to be a carrier for any condition included in this screen. Please see the supplemental table for details.
- This carrier screening test does not screen for all possible genetic conditions, nor for all possible mutations in every gene tested. This report does not include variants of uncertain significance; only variants classified as pathogenic or likely pathogenic at the time of testing, and considered relevant for reproductive carrier screening, are reported. Please see the gene specific notes for details. Please note that the classification of variants can change over time.
- Patients may wish to discuss any carrier results with blood relatives, as there is an increased chance that they are also carriers. These results should be interpreted in the context of this individual's clinical findings, biochemical profile, and family history.
- X-linked genes are not routinely analyzed for male carrier screening tests. Gene specific notes and limitations may be present. See below.
- 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)

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ALPHA THALASSEMIA

Patient	18198, Donor	Partner
Result	⊕ Carrier	N/A
Variant Details	***HBA2*** (NM_000517.5) Whole Gene Deletion (αα/α-)	N/A

What is Alpha thalassemia?

Alpha thalassemia is a blood disorder that reduces the production of a protein called hemoglobin. This reduction in the amount of hemoglobin can prevent enough oxygen from reaching the body's tissues. Affected individuals may have anemia, which can cause pale skin, weakness, fatigue, and more serious complications. There are two distinct types of alpha thalassemia: the more severe type is known as Hb Bart syndrome, and the milder form is called HbH disease. Hb Bart syndrome is characterized by hydrops fetalis, a condition in which excess fluid builds up in the body before birth. Additional signs and symptoms can include severe anemia, hepatosplenomegaly (swollen liver and spleen), heart defects, and abnormalities of the urinary system or genitalia. As a result of these serious health problems, most babies with this condition are stillborn or die soon after birth. Hb Bart syndrome can also cause serious complications for women during pregnancy, including preeclampsia, premature delivery, and abnormal bleeding. HbH disease causes mild to moderate anemia, hepatosplenomegaly, and jaundice. Some affected individuals also have bone changes such as overgrowth of the upper jaw and an unusually prominent forehead.

See the table below for a description of the diseases associated with different combinations of HBA1 and HBA2 mutations.

Carrier Status of Partner 1	Carrier Status of Partner 2	Risk for a child with HbH disease (- -/-α)	Risk for a child with Hb Bart syndrome (- -/- -)
alpha thalassemia trait cis (αα/- -)	alpha thalassemia trait trans (α-/α-) or (-α/-α)	50%	No risk
	alpha thalassemia trait cis (αα/- -)	Residual Risk	25%
	silent carrier (αα/α-) or (αα/-α)	25%	Residual Risk
alpha thalassemia trait trans (α-/α-) or (-α/-α)	alpha thalassemia trait trans (α-/α-) or (-α/-α)	Residual Risk	Residual Risk
	silent carrier (αα/α-) or (αα/-α)	Residual Risk	Residual Risk
silent carrier (αα/α-) or (αα/-α)	silent carrier (αα/α-) or (αα/-α)	Residual Risk	Residual Risk

Note that carriers for single heterozygous deletions of HBA1 or HBA2 are commonly referred to as silent carriers (PubMed: 20301608).

What is my risk of having an affected child?

Generally, each person has two copies of the *HBA1* gene and two copies of the *HBA2* gene, or four copies (alleles) in total. The different forms of alpha thalassemia result from the loss of some or all of these alleles: Hb Bart syndrome results from the loss of all four alleles, while HbH disease results from the loss of three alleles. Alpha thalassemia is inherited in an autosomal recessive manner, which means that if one parent is a carrier for a loss of two alleles on one chromosome and a second parent is a carrier of a loss of one or more alleles on one chromosome, there is a 1 in 4 (25%) risk of having an affected child.

What kind of medical management is available?

There is currently no cure for alpha thalassemia. Medical management of Hb Bart syndrome is limited but may include blood transfusions or a stem cell transplant. For HbH disease, management can vary based on the severity of symptoms. Mild forms may have little effect on daily life, and management for such cases can include supplementation of iron or folic acid. Management for more severe cases usually requires regular transfusions. Untreated, the prognosis for HbH disease is poor, with a shortened lifespan of up to age 5 years. However, when treated, individuals with HbH disease may have a lifespan that approaches normal.

What mutation was detected?

The detected variant was a whole gene deletion (αα/α-) in the HBA2 gene (NM_000517.5). The detected mutation was a whole

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gene deletion in one of four alleles comprising the alpha globin locus. These results are consistent with having three functional copies of alpha-globin (αα/α-). If your partner is also a carrier for alpha thalassemia, there is an increased risk to have a child with HbH disease, but not hydrops fetalis. Genetic counseling is recommended. Individuals with a single alpha globin gene defect (αα/α-) are carriers and clinically asymptomatic. Deletions of HBA2 are common in many human populations (PubMed: 20301608, 25390741). The laboratory classifies this variant as pathogenic.

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SPINAL MUSCULAR ATROPHY

Patient	18198, Donor	Partner
Result	⊕ Carrier	N/A
Variant Details	***SMN1*** (NM_022874.2) Absence of SMN1	N/A

What is Spinal muscular atrophy?

Spinal muscular atrophy (SMA) affects nerve cells called motor neurons in the spinal cord and brainstem. The progressive degeneration of these nerve cells results in muscle weakness and atrophy (degeneration of muscle mass). Common symptoms include limited growth and poor weight gain, restrictive lung disease, and skeletal abnormalities, such as scoliosis or contractures of the joints.

What is my risk of having an affected child?

SMA is inherited in an autosomal recessive manner. This means that when both parents are carriers for the condition, there is a 25% (1 in 4) risk of having an affected child.

What kind of medical management is available?

There is currently no cure for SMA, but management can include supportive, rehabilitative, and palliative care to manage symptoms and prevent complications. Treatment is designed to address the primary and secondary effects of muscle weakness and may include dietary management and placement of a gastronomy tube, breathing exercises or machines to help with breathing problems, and surgical repair for scoliosis. Exercises and stretches help with mobility and assistive equipment such as wheelchairs or splints may also be necessary. There have also been recent drug therapies developed to treat SMA such as Spinraza and Zolgensma. For SMA, it is important to provide proactive care and deliver timely intervention.

What mutation was detected?

The detected heterozygous mutation was an absence of SMN1. While the exact nature of the mutation detected cannot be determined, these results are consistent with absence of the SMN1 gene. Biallelic absence of the SMN1 gene is a common cause of autosomal recessive spinal muscular atrophy (PubMed: 20301526). The laboratory classifies this variant as pathogenic.

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MITOCHONDRIAL DNA DEPLETION SYNDROME 3

Patient	18198, Donor	Partner
Result	⊕ Carrier	N/A
Variant Details	***DGUOK*** (NM_080916.3) c.3G>A (p.?)	N/A

What is Mitochondrial DNA depletion syndrome 3?

Mitochondrial DNA depletion syndrome 3 (or deoxyguanosine kinase deficiency) can present either as a neonatal multisystem condition or as an isolated liver disease that begins in later infancy or early childhood. The neonatal multisystem form is more common and is characterized by liver disease and neurologic manifestations, including low muscle tone, nystagmus, and developmental delay, typically within weeks of birth. The isolated liver disease primarily involves jaundice, cholestasis, hepatomegaly, and elevated transaminases but may also have some kidney involvement and potentially mild low muscle tone.

What is my risk of having an affected child?

DGUOK-related Mitochondrial DNA depletion syndrome 3 is inherited in an autosomal recessive manner. If the patient and the partner are both carriers, the risk for an affected child is 1 in 4 (25%).

What kind of medical management is available?

Although there is no cure for mitochondrial DNA depletion syndrome 3 (or deoxyguanosine kinase deficiency), symptomatic care may improve quality of life. Those with feeding issues will benefit from feeding therapy and potentially tube feeding. Complications from liver disease can be managed through specialized diet, and some individuals may benefit from liver transplantation. Occupational therapy may help address motor dysfunction and communication issues. While those with isolated disease may have near-normal lifespans with appropriate management, individuals with the neonatal multisystem form typically do not survive past early childhood.

What mutation was detected?

The detected heterozygous variant was NM_080916.3:c.3G>A (p.?). This variant is predicted to abolish the initiator methionine. The first downstream in-frame methionine is located at codon 18. At least one variant upstream of this alternative start codon has been determined to be pathogenic, suggesting the region between original and alternative codons is critical to protein function (PubMed: 21534344, 28496820). This variant has been previously reported in either the homozygous state, unknown phase with a pathogenic variant, or in the unknown state in multiple individuals affected with mitochondrial DNA depletion syndrome or progressive liver failure (PubMed: 16908739, 21107780, 32278775, 32793533, 33486010). In addition, this variant has been identified in the homozygous state in an individual with deoxyguanosine kinase deficiency (PubMed: 37775787). Other initiation codon variants (c.1A>G and c.2T>C) that lead to the same molecular consequence, have been previously determined to be pathogenic in the literature and/or in Clinvar (PubMed: 16908739, 32278775, 32793533, 16263314; ClinVar: 2203104, 2734235). The laboratory classifies this variant as likely pathogenic.

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SYSTEMIC PRIMARY CARNITINE DEFICIENCY

Patient	18198, Donor	Partner
Result	⊕ Carrier	N/A
Variant Details	***SLC22A5*** (NM_003060.4) c.131C>T (p.Ala44Val)	N/A

What is Systemic primary carnitine deficiency?

Systemic primary carnitine deficiency (SPCD) is a disorder that varies in symptom and disease severity. In infancy, this disease presents between three months and two years of age with episodes of hypoglycemia, poor feeding, irritability, lethargy, liver disease, and elevated blood ammonia triggered by fasting or common illnesses. In childhood, between two and four years of age, this disease presents as a muscle disease affecting the heart and skeletal muscles. In pregnancy and adulthood, this disease can decrease stamina or worsen cardiac arrhythmias.

What is my risk of having an affected child?

Systemic primary carnitine deficiency is inherited in an autosomal recessive manner. If the patient and the partner are both carriers, the risk for an affected child is 1 in 4 (25%).

What kind of medical management is available?

Infantile and childhood presentations of disease can be fatal, if untreated. The long-term prognosis is favorable, as long as affected individuals remain on carnitine supplements. Affected patients who discontinue carnitine supplements are at risk of hypoglycemia or sudden death from arrhythmia.

What mutation was detected?

The detected heterozygous variant was NM_003060.4:c.131C>T (p.Ala44Val). SLC22A5 c.131C>T (p.Ala44Val) results in a non-conservative amino acid change in the encoded protein sequence. Four of five in-silico tools predict a benign effect of the variant on protein function. The variant allele was found at a frequency of 2.9e-05 in 245048 control chromosomes (gnomAD). The available data on variant occurrences in the general population are insufficient to allow any conclusion about variant significance. c.131C>T has been reported in the literature in individuals affected with Systemic Primary Carnitine Deficiency (PubMed: 23653224, 28711408, 28841266). These data indicate that the variant may be associated with disease. At least one publication reports experimental evidence evaluating an impact on protein function, showing that the variant protein displayed 8.22% of transport activity relative to wild-type OCTN2 (PubMed: 28841266). The following publications have been ascertained in the context of this evaluation (PubMed: 28711408, 23653224, 23963628, 27896095, 34637945). Five ClinVar submitters (evaluation after 2014) have reported the variant with conflicting assessments: 2 submitters classified the variant as likely pathogenic, and 3 submitters classified it as uncertain significance. The laboratory classifies this variant as likely pathogenic.

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WILSON DISEASE

Patient	18198, Donor	Partner
Result	⊕ Carrier	N/A
Variant Details	***ATP7B*** (NM_000053.4) c.3662G>A (p.Gly1221Glu)	N/A

What is Wilson disease?

Wilson disease is a disorder that affects the liver's ability to remove excess copper in the body. Normally, a healthy amount of copper is absorbed by the body through diet. When the liver is impaired and cannot properly excrete copper, the buildup over time can lead to symptoms such as liver disease, psychiatric disturbance, and neurologic disorder. Ages of onset vary, with symptoms most commonly reported between the ages of 5 to 35.

What is my risk of having an affected child?

Wilson disease is inherited in an autosomal recessive manner. This means that when both parents are carriers for the condition, there is a 1 in 4 (25%) risk of having an affected child.

What kind of medical management is available?

Medical management is specific to each individual and should be discussed with your doctor. To treat Wilson disease, lifelong management is required. Administration of chelating agents (medications) to remove excess copper and reducing dietary intake of copper may help to prevent disease progression. Liver transplantation may also be required in severe cases of liver damage. Biannual surveillance is strongly recommended to assess the progression of the disease and/or treatment. Routine monitoring should include blood tests, physical examinations, and liver function tests.

What mutation was detected?

The detected heterozygous variant was NM_000053.4:c.3662G>A (p.Gly1221Glu). This variant has been previously reported in the unknown phase with a pathogenic or a likely pathogenic variant as well as in the heterozygous state, without an identified second variant, in multiple individuals with Wilson disease (PubMed: 23518715, 16088907). This variant is classified as "Pathogenic" in ClinVar, with multiple submitters in agreement (Variation ID: 552337). Structural modeling study characterized this variant as a part of the functional site (PubMed: 22692182). The laboratory classifies this variant as likely pathogenic.

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GENES TESTED:

Beacon Preconception Carrier Screening - 515 Genes (without X-linked Disorders) - 515 Genes

This analysis was run using the Beacon Preconception Carrier Screening - 515 Genes (without X-linked Disorders) gene list. 515 genes were tested with 99.5% of targets sequenced at >20x coverage. For more gene-specific information and assistance with residual risk calculation, see the SUPPLEMENTAL TABLE.

AAAS, ABCA12, ABCA3, ABCA4, ABCB11, ABCB4, ABCC2, ABCC8, ACAD9, ACADM, ACADVL, ACAT1, ACOX1, ACSF3, ADA, ADAMTS2, ADAMTSL4, ADGRG1, ADGRV1, AGA, AGL, AGPS, AGXT, AHI1, AIPL1, AIRE, ALDH3A2, ALDH7A1, ALDOB, ALG1, ALG6, ALMS1, ALPL, AMN, AMT, ANO10, AP1S1, AQP2, ARG1, ARL6, ARSA, ARSB, ASL, ASNS, ASPA, ASS1, ATM, ATP6V1B1, ATP7B, ATP8B1, BBS1, BBS10, BBS12, BBS2, BBS4, BBS5, BBS7, BBS9, BCKDHA, BCKDHB, BCS1L, BLM, BLOC1S3, BLOC1S6, BMP1, BRIP1, BSND, CAD, CANT1, CAPN3, CASQ2, CBS, CC2D1A, CC2D2A, CCDC103, CCDC39, CCDC88C, CD3D, CD3E, CD40, CD59, CDH23, CEP152, CEP290, CERKL, CFTR, CHAT, CHRNE, CHRNG, CIITA, CLCN1, CLN3, CLN5, CLN6, CLN8, CLRN1, CNGB3, COL11A2, COL17A1, COL27A1, COL4A3, COL4A4, COL7A1, COX15, CPS1, CPT1A, CPT2, CRB1, CRTAP, CRYL1, CTNS, CTSA, CTSC, CTSD, CTSK, CYBA, CYP11A1, CYP11B1, CYP11B2, CYP17A1, CYP19A1, CYP1B1, CYP21A2, CYP27A1, CYP27B1, CYP7B1, DBT, DCAF17, DCLRE1C, DDX11, DGAT1, DGUOK, DHCR7, DHDDS, DLD, DLL3, DNAH11, DNAH5, DNAI1, DNAI2, DNMT3B, DOK7, DUOX2, DYNC2H1, DYSF, EIF2AK3, EIF2B1, EIF2B2, EIF2B3, EIF2B4, EIF2B5, ELP1, EPG5, ERCC2, ERCC6, ERCC8, ESCO2, ETFA, ETFB, ETFDH, ETHE1, EVC, EVC2, EXOSC3, EYS, FAH, FAM161A, FANCA, FANCC, FANCD2, FANCE, FANCG, FANCI, FANCL, FBP1, FBXO7, FH, FKBP10, FKRP, FKTN, FMO3, FOXN1, FOXRED1, FRAS1, FREM2, FUCA1, G6PC, G6PC3, GAA, GALC, GALE, GALK1, GALNS, GALNT3, GALT, GAMT, GATM, GBA, GBE1, GCDH, GCH1, GDF5, GFM1, GHR, GJB2, GJB6, GLB1, GLDC, GLE1, GNE, GNPAT, GNPTAB, GNPTG, GNS, GORAB, GRHPR, GRIP1, GSS, GUCY2D, GUSB, HADH, HADHA, HADHB, HAMP, HAX1, HBA1, HBA2, HBB, HEXA, HEXB, HGSNAT, HJV, HLCS, HMGCL, HMOX1, HOGA1, HPD, HPS1, HPS3, HPS4, HPS5, HPS6, HSD17B3, HSD17B4, HSD3B2, HYAL1, HYLS1, IDUA, IGHMBP2, IKBKB, IL7R, INVS, ITGA6, ITGB3, ITGB4, IVD, JAK3, KCNJ1, KCNJ11, LAMA2, LAMA3, LAMB3, LAMC2, LARGE1, LCA5, LDLR, LDLRAP1, LHX3, LIFR, LIG4, LIPA, LMBRD1, LOXHD1, LPL, LRAT, LRP2, LRPPRC, LYST, MAK, MAN2B1, MANBA, MCEE, MCOLN1, MCPH1, MECR, MED17, MESP2, MFSD8, MKKS, MKS1, MLC1, MLYCD, MMAA, MMAB, MMACHC, MMADHC, MOCS1, MOCS2, MPI, MPL, MPV17, MRE11, MTHFR, MTR, MTRR, MTTP, MUSK, MUT, MVK, MYO15A, MYO7A, NAGA, NAGLU, NAGS, NBN, NCF2, NDRG1, NDUFAF2, NDUFAF5, NDUFS4, NDUFS6, NDUFS7, NDUFV1, NEB, NEU1, NGLY1, NPC1, NPC2, NPHP1, NPHS1, NPHS2, NR2E3, NSMCE3, NTRK1, OAT, OCA2, OPA3, OSTM1, OTOA, OTOF, P3H1, PAH, PANK2, PC, PCBD1, PCCA, PCCB, PCDH15, PCNT, PDHB, PEPD, PET100, PEX1, PEX10, PEX12, PEX13, PEX16, PEX2, PEX26, PEX5, PEX6, PEX7, PFKM, PGM3, PHGDH, PHKB, PHKG2, PHYH, PIGN, PJVK, PKHD1, PLA2G6, PLEKHG5, PLOD1, PMM2, PNPO, POLG, POLH, POMGNT1, POMT1, POMT2, POR, POU1F1, PPT1, PRCD, PRDM5, PRF1, PROP1, PSAP, PTPRC, PTS, PUS1, PYGM, QDPR, RAB23, RAG1, RAG2, RAPSN, RARS2, RDH12, RLBP1, RMRP, RNASEH2A, RNASEH2B, RNASEH2C, RPE65, RPGRIP1L, RTEL1, RXYLT1, RYR1, SACS, SAMD9, SAMHD1, SCO2, SEC23B, SEPSECS, SGCA, SGCB, SGCD, SGCG, SGSH, SKIV2L, SLC12A1, SLC12A3, SLC12A6, SLC17A5, SLC19A2, SLC19A3, SLC1A4, SLC22A5, SLC25A13, SLC25A15, SLC25A20, SLC26A2, SLC26A3, SLC26A4, SLC27A4, SLC35A3, SLC37A4, SLC38A8, SLC39A4, SLC45A2, SLC4A11, SLC5A5, SLC7A7, SMARCAL1, SMN1, SMPD1, SNAP29, SPG11, SPR, SRD5A2, ST3GAL5, STAR, STX11, STXBP2, SUMF1, SUOX, SURF1, SYNE4, TANGO2, TAT, TBCD, TBCE, TCIRG1, TCN2, TECPR2, TERT, TF, TFR2, TG, TGM1, TH, TK2, TMC1, TMEM216, TMEM67, TMPRSS3, TPO, TPP1, TREX1, TRIM32, TRIM37, TRMU, TSEN54, TSFM, TSHB, TSHR, TTC37, TTPA, TULP1, TYMP, TYR, TYRP1, UBR1, UNC13D, USH1C, USH2A, VDR, VLDLR, VPS11, VPS13A, VPS13B, VPS45, VPS53, VRK1, VSX2, WISP3, WNT10A, WRN, XPA, XPC, ZBTB24, ZFYVE26, ZNF469

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, 99.56% and 99.52% 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 are analyzed using Fulgent Germline proprietary pipeline for this specimen. Bioinformatics: The Fulgent Germline v2019.2 pipeline was used to analyze this specimen.

LIMITATIONS:

General 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 future pregnancies, and negative results do not rule out the genetic risk to a pregnancy. 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 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 other than specified genes. 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 include one whole gene (buccal swab specimens and whole blood specimens) and are two or more contiguous exons in size (whole blood specimens only); 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.

Gene Specific Notes and Limitations

ALG1: Due to the interference by highly homologous regions, our current testing method has less sensitivity to detect variants in exons 6-13 of the ALG1 gene (NM_019109.4). *CEP290:* Copy number analysis for exons 8-13 and exons 39-42 may have reduced sensitivity in the CEP290 gene. Confirmation of these exons are limited to individuals with a positive personal history of CEP290-related conditions and/or individuals carrying a pathogenic/likely pathogenic sequence variant. *CFTR:* Analysis of the intron 8 polymorphic region (e.g. IVS8-5T allele) is only performed if the p.Arg117His (R117H) mutation is detected. Single exon deletion/duplication analysis is limited to deletions of previously reported exons: 1, 2, 3, 11, 19, 20, 21. Analysis of the intron 8 polymorphic region (e.g. IVS8-5T allele) is only performed if the p.Arg117His (R117H) mutation is detected. Single exon deletion/duplication analysis is limited to deletions of previously reported exons: 1, 2, 3, 11, 19, 20, 21. CFTR variants primarily associated with CFTR-related isolated congenital bilateral absence of the vas deferens and CFTR-related pancreatitis are not included in this analysis. CFTR variants with insufficient evidence of being cystic fibrosis mutations will not be reported either. *CRYL1:* As mutations in the CRYL1 gene are not known to be associated with any clinical condition, sequence variants in this gene are not analyzed. However, to increase copy number detection sensitivity for large deletions including this gene and a neighboring on gene on the panel (GJB6, also known as connexin 30), this gene was evaluated for copy number variation. *CYP11B1:* The current testing method is not able to reliably detect certain pathogenic variants in this gene due to the interference by highly homologous regions. This analysis is not designed to detect or rule-out copy-neutral chimeric CYP11B1/CYP11B2 gene. *CYP11B2:* The current testing method is not able to reliably detect certain pathogenic variants in this gene due to the interference by highly homologous regions. This analysis is not designed to detect or rule-out copy-neutral chimeric CYP11B1/CYP11B2 gene. *CYP21A2:* Significant pseudogene interference and/or reciprocal exchanges between the CYP21A2 gene and its pseudogene, CYP21A1P, have been known to occur and may impact results. As such, the relevance of variants reported in this gene must be interpreted clinically in the context of the clinical findings, biochemical profile, and family history of each patient. LR-PCR is not routinely ordered for NM_000500.9:c.955C>T (p.Gln319Ter). Individuals with c.955C>T (p.Gln319Ter) will be reported as a Possible Carrier indicating that the precise nature of the variant has not been determined by LR-PCR and that the variant may occur in the CYP21A2 wild-type gene or in the CYP21A1P pseudogene. The confirmation test is recommended if the second reproductive partner is tested positive for variants associated with classic CAH. *DDX11:* Due to the interference by highly homologous regions, our current testing method has less sensitivity to detect variants in the DDX11 gene. *DUOX2:* The current testing method is not able to reliably detect variants in exons 6-8 of the DUOX2 gene (NM_014080.5) due to significant interference by the highly homologous gene, DUOX1. *FANCD2:* Due to pseudogene interference, copy-number-variants within exon 14-17 of the FANCD2 gene (NM _033084.4) are not evaluated and detection of single-nucleotide variants and small insertions/deletions in this region is not guaranteed. *GALT:* In general, the D2 "Duarte" allele is not reported if detected, but can be reported upon request. While this allele can cause positive newborn screening results, it is not known to cause clinical symptoms in any state. See GeneReviews for more information: https://www.ncbi.nlm.nih.gov/books/NBK1518/ *GBA:* Significant pseudogene interference and/or reciprocal exchanges between the GBA gene and its pseudogene, GBAP1, have been known to occur and may impact results. As such, the relevance of variants reported in this gene must be interpreted clinically in the context of this individual's clinical findings, biochemical profile, and family history. The current testing method cannot detect copy-neutral rearrangements between the pseudogene and the functional gene, which have been reported in very rare cases of Gaucher disease (PubMed: 21704274). *HBA1:* Significant interference

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from highly homologous regions in exons 1-2 of the HBA1 gene has been recognized to occur, potentially impeding the assay's technical capability to detect pathogenic alterations during sequencing analyses. *HBA2:* Significant interference from highly homologous regions in exons 1-2 of the HBA2 gene has been recognized to occur, potentially impeding the assay's technical capability to detect pathogenic alterations during sequencing analyses. *HSD17B4:* Copy number analysis for exons 4-6 may have reduced sensitivity in the HSD17B4 gene. Confirmation of these exons are limited to individuals with a positive personal history of D-bifunctional protein deficiency and Perrault syndrome and/or individuals carrying a pathogenic/likely pathogenic sequence variant. *LMBRD1:* Copy number analysis for exons 9-12 may have reduced sensitivity in the LMBRD1 gene. Confirmation of these exons are limited to individuals with a positive personal history of combined methylmalonic aciduria and homocystinuria and/or individuals carrying a pathogenic/likely pathogenic sequence variant. *MTHFR:* As recommended by ACMG, the two common polymorphisms in the MTHFR gene - c.1286A>C (p.Glu429Ala, also known as c.1298A>C) and c.665C>T (p.Ala222Val, also known as c.677C>T) - are not reported in this test due to lack of sufficient clinical utility to merit testing (PubMed: 23288205). *NEB:* This gene contains a 32-kb triplicate region (exons 82-105) which is not amenable to sequencing and deletion/duplication analysis. *NPHS2:* If detected, the variant NM_014625.3:c.686G>A (p.Arg229Gln) will not be reported as this variant is not significantly associated with disease when homozygous or in the compound heterozygous state with variants in exons 1-6 of NPHS2. *OTOA:* Due to pseudogene interference, our current testing method is not able to reliably detect variants in exons 20-28 (NM_144672.3) in the OTOA gene. *SMN1:* The current testing method detects sequencing variants in exon 7 and copy number variations in exons 7-8 of the SMN1 gene (NM_022874.2). Sequencing and deletion/duplication analysis are not performed on any other region in this gene. About 5%-8% of the population have two copies of SMN1 on a single chromosome and a deletion on the other chromosome, known as a [2+0] configuration (PubMed: 20301526). The current testing method cannot directly detect carriers with a [2+0] SMN1 configuration but can detect linkage between the silent carrier allele and certain population-specific single nucleotide changes. As a result, a negative result for carrier testing greatly reduces but does not eliminate the chance that a person is a carrier. Only abnormal results will be reported. *TERT:* The TERT promoter region is analyzed for both sequencing and copy number variants. *TYR:* Due to the interference by highly homologous regions, our current testing method has less sensitivity to detect variants in exons 4-5 of the TYR gene (NM_000372.5). *VPS45:* LoF is not a known disease mechanism *WRN:* Due to the interference by highly homologous regions within the WRN gene, our current testing method has less sensitivity to detect variants in exons 10-11 of WRN (NM_000553.6).

SIGNATURE:

Yan Meng, Ph.D., CGMB, FACMG on **8/20/2024**
Laboratory Director, Fulgent

DISCLAIMER:

This test was developed and its performance characteristics determined by Fulgent Therapeutics LLC CAP #8042697 CLIA #05D2043189; 4399 Santa Anita Ave., El Monte, CA, 91731. 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 **626-350-0537** or by email at **info@fulgentgenetics.com**. 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.

To view the supplemental table describing the carrier frequencies, detection rates, and residual risks associated with the genes on this test please visit the following link:
Beacon Expanded Carrier Screening Supplemental Table

Patient Information	Specimen Information	Client Information
18198, DONOR **DOB:** [redacted] **AGE:** [redacted] Gender: M Fasting: U Phone: 303.970.5897 Patient ID: 18198 Health ID: 8573035037814593	Specimen: DV145081N Requisition: 0000132 Collected: 07/30/2024 / 10:31 MDT Received: 07/30/2024 / 21:03 MDT Reported: 08/09/2024 / 15:13 MDT	Client #: 70413924 DN99999 DENVER SPERM BANK 1601 E 19TH AVE STE 4500 DENVER, CO 80218-1289

COMMENTS: FASTING:UNKNOWN

Cytogenetic Report

CHROMOSOME ANALYSIS, BLOOD - 14596 Lab:EZ

CHROMOSOME ANALYSIS, BLOOD

Order ID: 24-360379
Specimen Type: Blood
Clinical Indication: GAMETE DONOR

RESULT:
NORMAL MALE KARYOTYPE

INTERPRETATION:
Chromosome analysis revealed normal G-band patterns within the limits of standard cytogenetic analysis.

Please expect the results of any other concurrent study in a separate report.

NOMENCLATURE:
46,XY

ASSAY INFORMATION:

Method: G-Band (Digital Analysis: MetaSyst
Cells Counted: 30
Band Level: 450
Cells Analyzed: 5
Cells Karyotyped: 5

This test does not address genetic disorders that cannot be detected by standard cytogenetic methods or rare events such as low level mosaicism or subtle rearrangements.

Sibel Kantarci, PhD, FACMG (800) NICHOLS-4307, [site SJC

Electronic Signature: 8/9/2024 4:14 PM

PERFORMING SITE:
EZ QUEST DIAGNOSTICS/NICHOLS SJC, 33608 ORTEGA HWY, SAN JUAN CAPISTRANO, CA 92675-2042 Laboratory Director: IRINA MARAMICA,MD,PHD,MBA, CLIA: 05D0643352