Donor 5310

Genetic Testing Summary

Fairfax Cryobank recommends reviewing this genetic testing summary with your healthcare provider to determine suitability.

Last Updated: 08/03/2020

Donor Reported Ancestry: African American, Italian Jewish Ancestry: No

Genetic Test*	Result	Comments/Donor's Residual Risk**
Chromosome analysis (karyotype)	Normal male karyotype	No evidence of clinically significant chromosome abnormalities
Hemoglobin evaluation	Normal hemoglobin fractionation and MCV/MCH results	Reduced risk to be a carrier for sickle cell anemia, beta thalassemia, alpha thalassemia trait (aa/-- and a-/a-) and other hemoglobinopathies
Cystic Fibrosis (CF) carrier screening	Negative by genotyping of 130 mutations in the CFTR gene	1/248
Spinal Muscular Atrophy (SMA) carrier screening	Negative for deletions of exon 7 in the SMN1 gene. Negative for variant c.*3+80T>G	1/455
Standard testing attached- 22 diseases by genotyping	Negative for mutations tested	
Special Testing		
Usher Syndrome Type 2A	Negative by sequencing in the USH2A gene	1/404
Homocystinuria CBS-Related	Negative by sequencing in the CBS gene	1/1328
Limb Girdle Muscular Dystrophy Type 2l (FKRP)	Negative by sequencing in the FKRP gene	1/1300

*No single test can screen for all genetic disorders. A negative screening result significantly reduces, but cannot eliminate, the risk for these conditions in a pregnancy.

**Donor residual risk is the chance the donor is still a carrier after testing negative.

Recombine

CarrierMap™

Ordering Practice:

Practice Code: [redacted]
Fairfax Cryobank - [redacted]
[redacted]
Physician:
Report Generated: 2015-08-07

Donor 5310

DOB: [redacted]
Gender: Male
Ethnicity: African
Procedure ID: 26914
Kit Barcode: [redacted]
Method: Genotyping
Specimen: Blood, #28372
Specimen Collection: 2015-07-30
Specimen Received: 2015-07-31
Specimen Analyzed: 2015-08-07

Partner Not Tested

SUMMARY OF RESULTS

NO MUTATIONS IDENTIFIED

Donor 5310 was not identified to carry any of the mutations tested.

All mutations analyzed were not detected, reducing but not eliminating your chance to be a carrier for the associated genetic diseases. A list of all the diseases and mutations you were screened for is included later in this report. The test does not screen for every possible genetic disease.

For disease information, please visit www.recombine.com/diseases. To speak with a Genetic Counselor, call 855.OUR.GENES.

♂ Male

Panel: Fairfax Cryobank Panel , Diseases Tested: 21, Mutations Tested: 381, Genes Tested: 22, Null Calls: 0

Assay performed by Reprogenetics
CLIA ID: 31D1054821
Lab Technician Bo Chu

Reviewed by Pere Colls, PhD, HCLD, Lab Director

Methods and Limitations

Genotyping: Genotyping is performed using the Illumina Infinium Custom HD Genotyping assay to identify mutations in >200 genes. The assay is not validated for homozygous mutations, and it is possible that individuals affected with disease may not be accurately genotyped.

Spinal Muscular Atrophy: Spinal Muscular Atrophy is tested for via an Identity-by-State shared haplotype comparison algorithm. Detection is limited to haplotypes within our library of known carriers of the most common mutation (deletion of Exon 7).

Limitations: In some cases, genetic variations other than that which is being assayed may interfere with mutation detection, resulting in false-negative or false-positive results. Additional sources of error include, but are not limited to: sample contamination, sample mix-up, bone marrow transplantation, blood transfusions, and technical errors.

The test does not test for all forms of genetic disease, birth defects, and intellectual disability. All results should be interpreted in the context of family history; additional evaluation may be indicated based on a history of these conditions. Additional testing may be necessary to determine mutation phase in individuals identified to carry more than one mutation in the same gene. All mutations included within the genes assayed may not be detected, and additional testing may be appropriate for some individuals.

Diseases & Mutations Assayed

High Impact | Treatment Benefits | X-Linked | Moderate Impact

H	T	X	M	Disease	#	Mutations
●	○	○	○	Alpha Thalassemia	10	♂ Genotyping | SEA deletion, 11.1kb deletion, c.207C>A (p.N69K), c.223G>C (p.D75G), c.2T>C (p.M1T), c.207C>G (p.N69K), c.340_351delCTCCCCGCCGAG (p.L114_ E117del), c.377T>C (p.L126P), c.427T>C (p.X143Qext32), c.*+94A>G
●	●	○	○	Beta Thalassemia	83	♂ Genotyping | c.17_18delCT, c.20delA (p.E7Gfs), c.217insA (p.S73Kfs), c.223+702_444+342del620insAAGTAGA, c.230delC, c.25_26delAA, c.315+1G>A, c.315+2T>C, c.316-197C>T, c.316-146T>G, c.315+745C>G, c.316-1G>A, c.316-1G>C, c.316-2A>G, c.316-3C>A, c.316-3C>G, c.4delG (p.V2Cfs), c.51delC (p.K18Rfs), c.93-21G>A, c.92+1G>A, c.92+5G>A, c.92+5G>C, c.92+5G>T, c.92+6T>C, c.93-1G>A, c.93-1G>T, c.-50A>C, c.a-78g, c.a-79g, c.a-81g, c.A52T (p.K18X), c.c-137g, c.c-138t, c.c-151t, c.C118T (p.Q40X), c.G169C (p.G57R), c.G295A (p.V99M), c.G34A (p.V12I), c.G415C (p.A139P), c.G47A (p.W16X), c.G48A (p.W16X), c.t-80a, c.T2C (p.M1T), c.T75A (p.G25G), c.444+111A>G, c.g-29a, c.68_74delAAGTTGG, c.G92C (p.R31T), c.27_28insG, c.92+1G>T, c.92+1G>C, c.93-15T>G, c.93-1G>C, c.112delT, c.G113A (p.W38X), c.G114A (p.W38X), c.126delC, c.444+113A>G, c.250delG, c.225delC, c.383_385delAGG (p.Q128_A129delQAinsP), c.321_322insG (p.N109fs), c.316-1G>T, c.316-2A>C, c.316-106C>T, c.287_288insA (p.L97fs), c.271G>T (p.E91X), c.203_204delTG (p.V68Afs), c.154delC (p.P52fs), c.135delC (p.F46fs), c.92+2T>A, c.92+2T>C, c.90C>T (p.G30G), c.59A>G (p.N20S), c.46delT (p.W16Gfs), c.45_46insG (p.L16fs), c.36delT (p.T13fs), c.2T>G (p.M1R), c.1A>G (p.M1V), c.c-137t, c.c-136g, c.c-142t, c.c-140t
●	○	○	○	Bloom Syndrome	24	♂ Genotyping | c.2207_2212delATCTGAinsTAGATTC (p.Y736Lfs), c.2407insT, c.557_559delCAA (p.S186X), c.1284G>A (p.W428X), c.1701G>A (p.W567X), c.1933C>T (p.Q645X), c.C2528T (p.T843I), c.C2695T (p.R899X), c.G3107T (p.C1036F), c.2923delC (p.Q975K), c.3558+1G>T, c.3875-2A>G, c.2074+2T>A, c.2343_2344dupGA (p.781EfsX), c.380delC (p.127Tfs), c.3564delC (p.1188Dfs), c.4008delG (p.1336Rfs), c.C947G (p.S316X), c.2193+1_2193+9del9, c.C1642T (p.Q548X), c.3143delA (p.1048NfsX), c.356_357delTA (p.Cys120Hisfs), c.4076+1delG, c.C3281A (p.S1094X)
●	○	○	○	Canavan Disease	7	♂ Genotyping | c.433-2A>G, c.A854C (p.E285A), c.C914A (p.A305E), c.A71G (p.E24G), c.C654A (p.C218X), c.T2C (p.M1T), c.G79A (p.G27R)

High Impact | Treatment Benefits | X-Linked | Moderate Impact

H	T	X	M	Disease	#	Mutations
●	●	○	○	Cystic Fibrosis	130	♂ Genotyping | c.1029delC, 1153_1154insAT, c.1519_1521delATC (p.507delI), c.1521_1523delCTT (p.508delF), c.1545_1546delTA (p.Y515Xfs), c.1585-1G>A, c.164+12T>C, c.1680-886A>G, c.1680-1G>A, c.1766+1G>A, c.1766+1G>T, c.1766+5G>T, c.1818del84, c.1911delG, c.1923delCTCAAAACTinsA, c.1973delGAAATTCAATCCTinsAGAAA, c.2052delA (p.K684fs), c.2052insA (p.Q685fs), c.2051_2052delAAinsG (p.K684SfsX38), c.2174insA, c.261delTT, c.2657+5G>A, c.273+1G>A, c.273+3A>C, c.274-1G>A, c.2988+1G>A, c.3039delC, c.3140-26A>G, c.325delTATinsG, c.3527delC, c.3535delACCA, c.3691delT, c.3717+12191C>T, c.3744delA, c.3773_3774insT (p.L1258fs), c.442delA, c.489+1G>T, c.531delT, c.579+1G>T, c.579+5G>A (IVS4+5G>A), c.803delA (p.N268fs), c.805_806delAT (p.I269fs), c.933_935delCTT (p.311delF), c.A1645C (p.S549R), c.A2128T (p.K710X), c.C1000T (p.R334W), c.C1013T (p.T338I), c.C1364A (p.A455E), c.C1477T (p.Q493X), c.C1572A (p.C524X), c.C1654T (p.Q552X), c.C1657T (p.R553X), c.C1721A (p.P574H), c.C2125T (p.R709X), c.C223T (p.R75X), c.C2668T (p.Q890X), c.C3196T (p.R1066C), c.C3276G (p.Y1092X), c.C3472T (p.R1158X), c.C3484T (p.R1162X), c.C349T (p.R117C), c.C3587G (p.S1196X), c.C3712T (p.Q1238X), c.C3764A (p.S1255X), c.C3909G (p.N1303K), c.G1040A (p.R347H), c.G1040C (p.R347P), c.G1438T (p.G480C), c.G1624T (p.G542X), c.G1646A (p.S549N), c.G1646T (p.S549I), c.G1652A (p.G551D), c.G1675A (p.A559T), c.G1679C (p.R560T), c.G178T (p.E60X), c.G1865A (p.G622D), c.G254A (p.G85E), c.G271A (p.G91R), c.G274T (p.E92X), c.G3209A (p.R1070Q), c.G3266A (p.W1089X), c.G3454C (p.D1152H), c.G350A (p.R117H), c.G3611A (p.W1204X), c.G3752A (p.S1251N), c.G3846A (p.W1282X), c.G3848T (p.R1283M), c.G532A (p.G178R), c.G988T (p.G330X), c.T1090C (p.S364P), c.T3302A (p.M1101K), c.T617G (p.L206W), c.C14T (p.P5L), c.G19T (p.E7X), c.G171A (p.W57X), c.313delA (p.I105fs), c.G328C (p.D110H), c.580-1G>T, c.G1055A (p.R352Q), c.C1075A (p.Q359K), c.C1079A (p.T360K), c.T1647G (p.S549R), c.1976delA (p.N659fs), c.C2290T (p.R764X), c.2737_2738insG (p.Y913X), c.3067_3072delATAGTG (p.I1023_V1024delT), c.3536_3539delCCAA (p.T1179fs), c.3659delC (p.T1220fs), c.G3808A (p.D1270N), c.G4056C (p.Q1352H), c.C4364G (p.S1455X), c.C4003T (p.L1335F), c.G2538A (p.W846X), c.C200T (p.P67L), c.C4426T (p.Q1476X), c.1116+1G>A, c.1986_1989delAACT (p.T663R), c.2089_2090insA (p.R697Kfs), c.2215delG (p.V739Y), c.T263G (p.L196X), c.3022delG (p.V1008S), c.3908dupA (p.N1303Kfs), c.C658T (p.Q220X), c.C868T (p.Q290X), c.1526delG (p.G509fs), c.2908+1085-3367+260del7201, c.C11A (p.S4X), c.A3700G (p.I1234V), c.A416T (p.H139L), c.T366A (p.Y122X)
●	○	○	○	Familial Dysautonomia	4	♂ Genotyping | c.2204+6T>C, c.C2741T (p.P914L), c.G2087C (p.R696P), c.C2128T (p.Q710X)
●	○	○	○	Familial Hyperinsulinism: Type 1: ABCC8 Related	10	♂ Genotyping | c.3989-9G>A, c.4159_4161delTTC (p.1387delF), c.C4258T (p.R1420C), c.C4477T (p.R1493W), c.G2147T (p.G716V), c.G4055C (p.R1352P), c.T560A (p.V187D), c.4516G>A (p.E1506K), c.C2506T (p.Q836X), c.579+2T>A
●	●	○	○	Fanconi Anemia: Type C	8	♂ Genotyping | c.456+4A>T, c.67delG, c.C37T (p.Q13X), c.C553T (p.R185X), c.T1661C (p.L554P), c.C1642T (p.R548X), c.G66A (p.W22X), c.G65A (p.W22X)
●	●	○	○	Gaucher Disease	6	♂ Genotyping | c.84_85insG, c.A1226G (p.N409S), c.A1343T (p.D448V), c.C1504T (p.R502C), c.G1297T (p.V433L), c.G1604A (p.R535H)
●	●	○	○	Glycogen Storage Disease: Type IA	13	♂ Genotyping | c.376_377insTA, c.79delC, c.979_981delTTC (p.327delF), c.C1039T (p.Q347X), c.C247T (p.R83C), c.C724T (p.Q242X), c.G248A (p.R83H), c.G562C (p.G188R), c.G648T, c.G809T (p.G270V), c.A113T (p.D38V), c.975delG (p.L326fs), c.724delC
●	○	○	○	Joubert Syndrome	1	♂ Genotyping | c.G35T (p.R12L)

High Impact Treatment Benefits X-Linked Moderate Impact

H	T	X	M	Disease	#	Mutations
●	●	○	○	Maple Syrup Urine Disease: Type 1B	6	♂ Genotyping | c.G1114T (p.E372X), c.G548C (p.R183P), c.G832A (p.G278S), c.C970T (p.R324X), c.G487T (p.E163X), c.C853T (p.R285X)
●	●	○	○	Maple Syrup Urine Disease: Type 3	8	♂ Genotyping | c.104_105insA, c.G685T (p.G229C), c.A214G (p.K72E), c.A1081G (p.M361V), c.G1123A (p.E375K), c.T1178C (p.I393T), c.C1463T (p.P488L), c.A1483G (p.R495G)
●	○	○	○	Mucolipidosis: Type IV	4	♂ Genotyping | c.406-2A>G, c.G1084T (p.D362Y), c.C304T (p.R102X), c.244delC (p.L82fsX)
●	○	○	○	Nemaline Myopathy: NEB Related	1	♂ Genotyping | c.7434_7536del2502bp
●	○	○	○	Niemann-Pick Disease: Type A	6	♂ Genotyping | c.996delC, c.G1493T (p.R498L), c.T911C (p.L304P), c.C1267T (p.H423Y), c.G1734C (p.K578N), c.1493G>A (p.R498H)
●	○	○	○	Spinal Muscular Atrophy: SMN1 Linked	19	♂ Genotyping | DEL EXON 7, c.22_23insA, c.43C>T (p.Q15X), c.91_92insT, c.305G>A (p.W102X), c.400G>A (p.E134K), c.439_443delGAAGT, c.558delA, c.585_586insT, c.683T>A (p.L228X), c.734C>T (p.P245L), c.768_778dupTGCTGATGCTT, c.815A>G (p.Y272C), c.821C>T (p.T274I), c.823G>A (p.G275S), c.834+2T>G, c.835-18_835-12delCCTTTAT, c.835G>T, c.836G>T
●	○	○	○	Tay-Sachs Disease	30	♂ Genotyping | c.1073+1G>A, c.1277_1278insTATC, c.1421+1G>C, c.805+1G>A, c.C532T (p.R178C), c.G533A (p.R178H), c.G805A (p.G269S), c.C1510T (p.R504C), c.G1496A (p.R499H), c.G509A (p.R170Q), c.A1003T (p.I335F), c.910_912delTTC (p.305delF), c.G749A (p.G250D), c.T632C (p.F211S), c.C629T (p.S210F), c.613delC, c.A611G (p.H204R), c.G598A (p.V200M), c.A590C (p.K197T), c.571-1G>T, c.C540G (p.Y180X), c.T538C (p.Y180H), c.G533T (p.R178L), c.C508T (p.R170W), c.C409T (p.R137X), c.T380G (p.L127R), c.346+1G>C, c.T116G (p.L39R), c.G78A (p.W26X), c.A1G (p.M1V)
●	○	○	○	Usher Syndrome: Type 1F	6	♂ Genotyping | c.C733T (p.R245X), c.2067C>A (p.Y684X), c.C7T (p.R3X), c.C1942T (p.R648X), c.2800C>T (p.R934X), c.4272delA (p.L1425fs)
●	○	○	○	Usher Syndrome: Type 3	4	♂ Genotyping | c.T144G (p.N48K), c.T359A (p.M120K), c.300T>G (p.Y176X), c.C634T (p.Q212X)
●	○	○	○	Walker-Warburg Syndrome	1	♂ Genotyping | c.1167insA (p.F390fs)

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CARRIER SCREENING REPORT

Patient	Sample	Referring Doctor
Patient Name: Donor 5310 **Date of Birth:** [redacted] **Reference #:** [redacted] **Indication:** Carrier Testing **Test Type:** NGS single gene full sequencing test	**Specimen Type:** Blood **Lab #:** [redacted] **Date Collected:** 12/12/2017 **Date Received:** 12/13/2017 **Final Report:** 12/26/2017	[redacted] **Fairfax Cryobank, Inc.** [redacted] [redacted] [redacted] Fax: [redacted]

RESULT SUMMARY

NGS single gene full sequencing test

Results: No clinically significant variant(s) detected

Gene(s) analyzed: *USH2A* and *CBS*

Interpretation: Screening for the presence of pathogenic variants in the *USH2A* and *CBS* genes which are associated with Usher syndrome, type IIA and homocystinuria (*CBS*-related), respectively, was performed by next generation sequencing and possibly genotyping for select variants on DNA extracted from this patient's sample. No clinically significant variants were detected during this analysis. This negative result does not rule out the possibility that a pathogenic variant in the genes examined is present.

Genetic counseling is recommended.

This technology may not detect all small insertion/deletions and is not diagnostic for large duplications/deletions and structural genomic variation. The coding DNA sequence of the gene plus at least five base pairs flanking splice sites were sequenced and analyzed relative to the hg19 assembly. A mutation(s) deep in intronic sequences or in untranslated regions of the gene except a portion described above or a pathogenic variant(s) in other genes not included in this test could be present in this patient. The analytical sensitivity of this test is estimated at 99% for single base substitutions and 97% overall. All potentially pathogenic variants were subjected to Sanger sequencing or genotyping by allele specific primer extension analysis for confirmation of the result. Any benign variants identified during this analysis were not reported.

Please note that this carrier screening test masks likely benign variants and variants of uncertain significance (VUS) if there are any. Only known pathogenic variants or likely pathogenic variants which are expected to result in deleterious effects on protein function are reported. If reporting of likely benign variants and VUS is desired in this patient, please contact the laboratory (tel. 212-241-2537) to request an amended report.

Comments: This test was developed and its performance characteristics were determined by Mount Sinai Genomics, Inc. It is considered acceptable for patient testing. It has not been cleared or approved by the FDA. The FDA has determined that such clearance or approval is not necessary.

Patient:	Donor 5310	DOB:	[redacted]	Lab #:	[redacted]

This type of mutation analysis generally provides highly accurate genotype information for point mutations and single nucleotide polymorphisms. Despite this level of accuracy, it should be kept in mind that there are many potential sources of diagnostic error, including misidentification of samples, polymorphisms, mosaicism or other rare genetic variants that interfere with analysis. In addition, families should understand the limitations of the testing and that rare diagnostic errors may occur for the reasons described.

For Disease Specific Standards and Guidelines:
https://www.acmg.net/
Additional disease-specific references available upon request.

This case has been reviewed and electronically signed by Guiqing Cai, Ph.D, Assistant Laboratory Director

Laboratory Medical Consultant: George A. Diaz, M.D., Ph.D.

Table of Residual Risks by Ethnicity

Please note: This table displays residual risks after a negative result for each of the genes and corresponding disorders. **If a patient is reported to be a carrier of a disease, their residual risk is 1 and this table does not apply for that disease.**

Disease (Inheritance)	Gene	Ethnicity	Carrier Frequency	Detection Rate	Residual Risk	Analytical Detection Rate
Homocystinuria (*CBS*-Related) (AR)	*CBS*	Caucasian	1 in 52	74%	1 in 197	>95%
NM_000071.2		Worldwide	1 in 293	78%	1 in 1328	>95%
		Qatari	1 in 21	86%	1 in 144	>95%
Usher Syndrome, Type IIA (AR)	*USH2A*	Caucasian	1 in 73	77%	1 in 314	88%
NM_206933.2		Worldwide	1 in 126	69%	1 in 404	>95%
		Sephardic Jewish – Iraqi and Iranian	1 in 36	71%	1 in 122	75%

AR: Autosomal Recessive

Patient Information
Name: Donor 5310
Date of Birth: [redacted]
Sema4 ID: [redacted]
Client ID: [redacted]
Indication: Carrier Testing

Specimen Information
Specimen Type: Purified DNA
Date Collected: 07/13/2020
Date Received: 07/17/2020
Final Report: 07/31/2020

Referring Provider
[redacted]
Fairfax Cryobank, Inc.
[redacted]

Custom Carrier Screen (ECS)

Number of genes tested: 2

SUMMARY OF RESULTS AND RECOMMENDATIONS

Negative

Negative for all genes tested: *FKRP, and SMN1*
To view a full list of genes and diseases tested
please see Table 1 in this report

AR=Autosomal recessive; XL=X-linked

Recommendations

- Consideration of residual risk by ethnicity after a negative carrier screen is recommended for the other diseases on the panel, especially in the case of a positive family history for a specific disorder.

Test description

This patient was tested for the genes listed above using one or more of the following methodologies: target capture and short-read sequencing, long-range PCR followed by short-read sequencing, targeted genotyping, and/or copy number analysis. Please note that negative results reduce but do not eliminate the possibility that this individual is a carrier for one or more of the disorders tested. Please view the Table of Residual Risks Based on Ethnicity at the end of this report or at **go.sema4.com/residualrisk** for gene transcripts, sequencing exceptions, specific detection rates, and residual risk estimates after a negative screening result. With individuals of mixed ethnicity, it is recommended to use the highest residual risk estimate. Only known pathogenic or likely pathogenic variants are reported. This carrier screening test does not report likely benign variants and variants of uncertain significance (VUS). If reporting of likely benign variants and VUS are desired in this patient, please contact the laboratory at 800-298-6470, option 2 to request an amended report.

Anastasia Larmore, Ph.D., Assistant Laboratory Director
Laboratory Medical Consultant: George A. Diaz, M.D., Ph.D.

Genes and diseases tested

For specific detection rates and residual risk by ethnicity, please visit **go.sema4.com/residualrisk**

Table 1: List of genes and diseases tested with detailed results

Disease	Gene	Inheritance Pattern	Status	Detailed Summary
⊖ **Negative**				
Limb-Girdle Muscular Dystrophy, Type 2I	*FKRP*	AR	Reduced Risk (see table below)	
Spinal Muscular Atrophy	*SMN1*	AR	Reduced Risk (see table below)	*SMN1* copy number: 2 *SMN2* copy number: 2 c.*3+80T>G: Negative

AR=Autosomal recessive; XL=X-linked

Table 2: Residual Risk by ethnicity for negative results

Disease (Inheritance)	Gene	Ethnicity	Carrier Frequency	Detection Rate	Residual Risk	Analytical Detection Rate
Limb-Girdle Muscular Dystrophy, Type 2I (AR) NM_024301.4	*FKRP*	African	1 in 452	86%	1 in 3,300	99%
		Ashkenazi Jewish	1 in 184	87%	1 in 1,400	
		East Asian	1 in 196	57%	1 in 460	
		Finnish	1 in 229	99%	1 in 22,800	
		European (Non-Finnish)	1 in 176	86%	1 in 1,300	
		Native American	1 in 239	16%	1 in 280	
		South Asian	1 in 2190	45%	1 in 4,000	
		Worldwide	1 in 220	75%	1 in 880	
		Norwegian	1 in 116	99%	1 in 11,500	

Spinal Muscular Atrophy (AR)
NM_000344.3 — *SMN1*

Ethnicity	Carrier Frequency	Detection rate	Residual risk after negative result*	Detection rate with *SMN1* c.*3+80T>G	Residual risk c.*3+80T>G negative	Residual risk c.*3+80T>G positive	Residual Risk with ≥3 Copies of *SMN1*
African American	1 in 85	71%	1 in 160	91%	1 in 455	1 in 49	1 in 4,300
Ashkenazi Jewish	1 in 76	90%	1 in 672	93%	1 in 978	1 in 10	1 in 4,800
East Asian	1 in 53	94%	1 in 864	95%	1 in 901	1 in 12	1 in 4,900
European (Non-Finnish)	1 in 48	95%	1 in 803	95%	1 in 894	1 in 23	1 in 4,900
Native American	1 in 63	91%	1 in 609	94%	1 in 930	1 in 47	1 in 4,800
South Asian	1 in 103	87%	1 in 637	87%	1 in 637	1 in 608	1 in 4,700
Sephardic Jewish	1 in 34	96%	1 in 696	97%	1 in 884	1 in 12	1 in 4,900

*Residual risk with two copies *SMN1* detected using dosage sensitive methods. The presence of three or more copies of *SMN1* reduces the risk of being an *SMN1* carrier between 5-10 fold, depending on ethnicity.

* Carrier detection by HEXA enzyme analysis has a detection rate of approximately 98% (Applies to *HEXA* gene testing only).
ǂ Carrier frequencies include milder and reduced penetrance forms of the disease. Therefore, carrier frequencies may appear higher than reported in the literature (Applies to *BTD, F9, GJB2, GJB1, GLA,* and *MEFV* gene testing only).
‡ Please note that *GJB2* testing includes testing for the two upstream deletions, del(GJB6-D13S1830) and del(GJB6-D13S1854) (PMID:11807148 and 15994881) (Applies to *GJB2* gene testing only).
AR: Autosomal recessive; N/A: Not available; XL: X-linked

Test methods and comments

Genomic DNA isolated from this patient was analyzed by one or more of the followingmethodologies, as applicable:

Next Generation Sequencing (NGS) (Analytical Detection Rate >95%)

NGS was performed on a panel of genes for the purpose of identifying pathogenic or likelypathogenic variants.

Agilent SureSelect^{TM}QXT technology was used with a custom capture library to target theexonic regions and intron/exon splice junctions of the relevant genes, as well as a numberof UTR, intronic or promoter regions that contain previously reported mutations. Sampleswere pooled and sequenced on the Illumina HiSeq 2500 platform in the Rapid Run mode or theIllumina NovaSeq platform in the Xp workflow, using 100 bp paired-end reads. Thesequencing data was analyzed using a custom bioinformatics algorithm designed andvalidated in house.

The coding exons and splice junctions of the known protein-coding RefSeq genes wereassessed for the average depth of coverage (minimum of 20X) and data quality thresholdvalues. Most exons not meeting a minimum of >20X read depth across the exon are furtheranalyzed by Sanger sequencing. Please note that several genomic regions presentdifficulties in mapping or obtaining read depth >20X. The exons contained within theseregions are noted within Table 1 (as "Exceptions") and will not be reflexed to Sangersequencing if the mapping quality or coverage is poor. Any variants identified duringtesting in these regions are confirmed by a second method and reported if determined to bepathogenic or likely pathogenic. However, as there is a possibility of false negativeresults within these regions, detection rates and residual risks for these genes have beencalculated with the presumption that variants in these exons will not be detected, unlessincluded in the MassARRAY® genotyping platform.

This test will detect variants within the exons and the intron-exon boundaries of thetarget regions. Variants outside these regions may not be detected, including, but notlimited to, UTRs, promoters, and deep intronic areas, or regions that fall into theExceptions mentioned above. This technology may not detect all small insertion/deletionsand is not diagnostic for repeat expansions and structural genomic variation. In addition,a mutation(s) in a gene not included on the panel could be present in this patient.

Variant interpretation and classification was performed based on the American College ofMedical Genetics Standards and Guidelines for the Interpretation of Sequence Variants(Richards et al, 2015). All potentially pathogenic variants may be confirmed by either aspecific genotyping assay or Sanger sequencing, if indicated. Any benign variants, likelybenign variants or variants of uncertain significance identified during this analysis willnot be reported.

Copy Number Variant Analysis (Analytical Detection Rate >95%)

Large duplications and deletions were called from the relative read depths on anexon-by-exon basis using a custom exome hidden Markov model (XHMM) algorithm. Deletions orduplications determined to be pathogenic or likely pathogenic were confirmed by either acustom arrayCGH platform, quantitative PCR, or MLPA (depending on CNV size and gene content). While this algorithm is designed to pick updeletions and duplications of 2 or more exons in length, potentially pathogenicsingle-exon CNVs will be confirmed and reported, if detected.

Exon Array (Confirmation method) (Accuracy >99%)

The customized oligonucleotide microarray (Oxford Gene Technology) is a highly-targetedexon-focused array capable of detecting medically relevant microdeletions andmicroduplications at a much higher resolution than traditional aCGH methods. Each arraymatrix has approximately 180,000 60-mer oligonucleotide probes that cover the entiregenome. This platform is designed based on human genome NCBI Build 37 (hg19) and the CGHprobes are enriched to target the exonic regions of the genes in this panel.

Quantitative PCR (Confirmation method) (Accuracy >99%)

The relative quantification PCR is utilized on a Roche Universal Library Probe (UPL)system, which relates the PCR signal of the target region in one group to another. To testfor genomic imbalances, both sample DNA and reference DNA is amplified with primer/probesets that specific to the target region and a control region with known genomic copynumber. Relative genomic copy numbers are calculated based on the standard ΔΔCt formula.

Long-Range PCR (Analytical Detection Rate >99%)

Long-range PCR was performed to generate locus-specific amplicons for *CYP21A2* , *HBA1*and *HBA2*and *GBA* . The PCR products were then prepared for short-read NGS sequencing and sequenced.Sequenced reads were mapped back to the original genomic locus and run through thebioinformatics pipeline. If indicated, copy number from MLPA was correlated with thesequencing output to analyze the results. For *CYP21A2* , a certain percentage of healthy individuals carry a duplication of the *CYP21A2*gene, which has no clinical consequences. In cases where two copies of a gene are locatedon the same chromosome in tandem, only the second copy will be amplified and assessed forpotentially pathogenic variants, due to size limitations of the PCR reaction. However,because these alleles contain at least two copies of the *CYP21A2*gene in tandem, it is expected that this patient has at least one functional gene in thetandem allele and this patient is therefore less likely to be a carrier. When anindividual carries both a duplication allele and a pathogenic variant, or multiplepathogenic variants, the current analysis may not be able to determine the phase(cisrans configuration) of the *CYP21A2*alleles identified. Family studies may be required in certain scenarios where phasing isrequired to determine the carrier status.

Residual Risk Calculations

Carrier frequencies and detection rates for each ethnicity were calculated through thecombination of internal curations of >28,000 variants and genomic frequency data from>138,000 individuals across seven ethnic groups in the gnomAD database. Additionalvariants in HGMD and novel deleterious variants were also incorporated into thecalculation. Residual risk values are calculated using a Bayesian analysis combining the *a priori* risk of being a pathogenic mutation carrier (carrier frequency) and the detection rate.They are provided only as a guide for assessing approximate risk given a negative result,and values will vary based on the exact ethnic background of an individual. This reportdoes not represent medical advice but should be interpreted by a genetic counselor,medical geneticist or physician skilled in genetic result interpretation and the relevantmedical literature.

Sanger Sequencing (Confirmation method) (Accuracy >99%)

Sanger sequencing, as indicated, was performed using BigDye Terminator chemistry with theABI 3730 DNA analyzer with target specific amplicons. It also may be used to supplementspecific guaranteed target regions that fail NGS sequencing due to poor quality or lowdepth of coverage (<20 reads) or as a confirmatory method for NGS positive results. Falsenegative results may occur if rare variants interfere with amplification or annealing.

SELECTED REFERENCES

Carrier Screening

Grody W et al. ACMG position statement on prenatal/preconception expanded carrierscreening. *Genet Med.*2013 15:482-3.

Variant Classification:

Richards S et al. Standards and guidelines for the interpretation of sequence variants: ajoint consensus recommendation of the American College of Medical Genetics and Genomicsand the Association for Molecular Pathology. *Genet Med.*2015 May;17(5):405-24

Additional disease-specific references available upon request.