INVITAE

INVITAE DIAGNOSTIC TESTING RESULTS

Patient name:	Milana Bulyga	**Sample type:**	Blood	**Report date:**	01/05/2022
DOB:	10/07/2019	**Sample collection date:**	12/03/2021	**Invitae #:**	RQ3000695
Sex assigned at birth:	Female	**Sample accession date:**	12/16/2021	**Clinical team:**	Qamar Hussain
Gender:		**MRN:**			

Reason for testing

Diagnostic test for a personal history of disease

Test performed

Sequence analysis and deletion/duplication testing of the 458 genes listed in the Genes Analyzed section.
Multiple panels/genes ordered: see Methods for complete list.

RESULT: POSITIVE

Two Pathogenic variants identified in SMN1. SMN1 is associated with autosomal recessive spinal muscular atrophy.

SMN2 copy number: 3.

Additional Variant(s) of Uncertain Significance identified.

GENE	VARIANT	ZYGOSITY	VARIANT CLASSIFICATION
SMN1	Deletion (Entire coding sequence)	homozygous	PATHOGENIC
ITGA7	c.3200G>A (p.Arg1067Gln)	heterozygous	Uncertain Significance
MEGF10	c.2704G>A (p.Val902Ile)	heterozygous	Uncertain Significance
MOCS1	c.520G>A (p.Ala174Thr)	heterozygous	Uncertain Significance
MYO18B	c.4984G>T (p.Val1662Leu)	heterozygous	Uncertain Significance
MYO18B	c.5593C>T (p.His1865Tyr)	heterozygous	Uncertain Significance
PNPLA2	c.236G>A (p.Arg79Gln)	heterozygous	Uncertain Significance
SPEG	c.5635C>T (p.Arg1879Cys)	heterozygous	Uncertain Significance
GALC	c.1685T>C (p.Ile562Thr)	heterozygous	Benign (Pseudodeficiency allele)

About this test

This diagnostic test evaluates 458 gene(s) for variants (genetic changes) that are associated with genetic disorders. Diagnostic genetic testing, when combined with family history and other medical results, may provide information to clarify individual risk, support a clinical diagnosis, and assist with the development of a personalized treatment and management strategy.

Laboratory Director: Kristin Gibson, PhD, FACMG, NYCQ, CGMB, ErCLG
Invitae 1400 16th Street, San Francisco, CA 94103 · clientservices@invitae.com · 800.436.3037

Next steps

- This is a medically important result that should be discussed with a healthcare provider, such as a genetic counselor, to learn more about this result and the appropriate next steps for further evaluation, treatment and/or management. This result should be interpreted within the context of additional laboratory results, family history and clinical findings.
- Consider sharing this result with relatives as they may also be at risk. Details on our Family Variant Testing program can be found at **www.invitae.com/family**.
- One or more variants were identified that are not known to cause disease. See the GALC variant(s) in the Variant Details section for more information.
- Register your test at **www.invitae.com/patients** to download a digital copy of your results. You can also access educational resources about how your results can help inform your health.

Clinical summary

Two Pathogenic variants, Deletion (Entire coding sequence) (homozygous), were identified in SMN1.

- The SMN1 gene is associated with autosomal recessive spinal muscular atrophy (SMA) (MedGen UID: 21913, 101816, 95975, 325364).
- This result is consistent with a diagnosis of spinal muscular atrophy.
- Spinal muscular atrophy (SMA) is a neuromuscular disorder caused by the loss of neurons within the spinal cord, which results in progressive muscle weakness and atrophy (PMID: 10679938, 18572081). Other features of SMA may include muscle fasciculations, tremor, poor weight gain, sleeping difficulties, pneumonia, scoliosis, joint contractures, and congenital heart disease (PMID: 18662980, 7658877). Four clinical SMA subtypes have been distinguished: severe infantile acute SMA type I (also referred to as Wednig-Hoffman disease), infantile chronic SMA type II, juvenile SMA type III (also referred to as Wohlfard-Kugelberg-Welander disease), and adult-onset SMA type IV (PMID: 10679938, 18572081). The age of onset, phenotype, prognosis, and life expectancy display marked inter- and intrafamilial variability, between and also within subtypes (PMID: 6855803). For SMA clinical management guidelines, refer to PMID: 17761659.
- Biological relatives have a chance of being a carrier for or being at risk for autosomal recessive spinal muscular atrophy. Testing should be considered if clinically appropriate. The chance of having a child with autosomal recessive spinal muscular atrophy depends on the carrier state of this individual's partner.

This individual has 3 copies of SMN2 exon 8, typically referred to as exon 7, which is expected to reflect SMN2 copy number.

- While the SMN2 gene alone is not associated with disease, variation in SMN2 copy number can modify the phenotype of SMN1-related SMA (PMID: 15378550). Accumulating evidence indicates that, in individuals with homozygous Pathogenic deletions in SMN1, the presence of two or fewer copies of SMN2 may be associated with a more severe phenotype compared to the presence of three or more copies of SMN2, which may be associated with a milder phenotype (PMID: 8824882, 9199562, 9837824, 11839954). Unaffected individuals with five copies of SMN2 and a homozygous deletion in SMN1 have been reported, suggesting that five copies of SMN2 may compensate for the lack of SMN1 expression (PMID: 15378550).

A Variant of Uncertain Significance, c.3200G>A (p.Arg1067Gln), was identified in ITGA7.

- The ITGA7 gene is associated with autosomal recessive congenital muscular dystrophy due to integrin alpha-7 deficiency (MedGen UID: 413044).
- Not all variants present in a gene cause disease. The clinical significance of the variant(s) identified in this gene is uncertain. Until this uncertainty can be resolved, caution should be exercised before using this result to inform clinical management decisions.
- Familial VUS testing is not offered. Testing family members for this variant will not contribute evidence to allow variant reclassification. Details on our VUS Resolution and Family Variant Testing Programs can be found at https://www.invitae.com/family.

A Variant of Uncertain Significance, c.2704G>A (p.Val902Ile), was identified in MEGF10.

- The MEGF10 gene is associated with autosomal recessive early-onset myopathy, areflexia, respiratory distress and dysphagia (EMARDD) (MedGen UID: 482309).
- Not all variants present in a gene cause disease. The clinical significance of the variant(s) identified in this gene is uncertain. Until this uncertainty can be resolved, caution should be exercised before using this result to inform clinical management decisions.
- Familial VUS testing is not offered. Testing family members for this variant will not contribute evidence to allow variant reclassification. Details on our VUS Resolution and Family Variant Testing Programs can be found at https://www.invitae.com/family.

A Variant of Uncertain Significance, c.520G>A (p.Ala174Thr), was identified in MOCS1.

- The MOCS1 gene is associated with autosomal recessive molybdenum cofactor deficiency (MedGen UID: 381530).
- Not all variants present in a gene cause disease. The clinical significance of the variant(s) identified in this gene is uncertain. Until this uncertainty can be resolved, caution should be exercised before using this result to inform clinical management decisions.
- Familial VUS testing is not offered. Testing family members for this variant will not contribute evidence to allow variant reclassification. Details on our VUS Resolution and Family Variant Testing Programs can be found at https://www.invitae.com/family.

Two Variants of Uncertain Significance, c.4984G>T (p.Val1662Leu) and c.5593C>T (p.His1865Tyr), were identified in MYO18B. The data from this test cannot definitively determine if these variants are on the same or opposite chromosomes.

- The MYO18B gene is associated with autosomal recessive Klippel-Feil syndrome with myopathy and facial dysmorphism (KFS4) (MedGen UID: 894399).

- Not all variants present in a gene cause disease. The clinical significance of the variant(s) identified in this gene is uncertain. Until this uncertainty can be resolved, caution should be exercised before using this result to inform clinical management decisions.
- Familial VUS testing is not offered. Testing family members for these variants will not contribute evidence to allow variant reclassification. Details on our VUS Resolution and Family Variant Testing Programs can be found at https://www.invitae.com/family.

A Variant of Uncertain Significance, c.236G>A (p.Arg79Gln), was identified in PNPLA2.

- The PNPLA2 gene is associated with autosomal recessive neutral lipid storage disease with myopathy (NLSDM) (MedGen UID: 339913).
- Not all variants present in a gene cause disease. The clinical significance of the variant(s) identified in this gene is uncertain. Until this uncertainty can be resolved, caution should be exercised before using this result to inform clinical management decisions.
- Familial VUS testing is not offered. Testing family members for this variant will not contribute evidence to allow variant reclassification. Details on our VUS Resolution and Family Variant Testing Programs can be found at https://www.invitae.com/family.

A Variant of Uncertain Significance, c.5635C>T (p.Arg1879Cys), was identified in SPEG.

- The SPEG gene is associated with autosomal recessive centronuclear myopathy 5 (CNM5) (MedGen UID: 863251). Additionally, the SPEG gene has preliminary evidence supporting a correlation with autosomal recessive dilated cardiomyopathy (PMID: 32925938) and autosomal dominant autism spectrum disorder (PMID: 28191890).
- Not all variants present in a gene cause disease. The clinical significance of the variant(s) identified in this gene is uncertain. Until this uncertainty can be resolved, caution should be exercised before using this result to inform clinical management decisions.
- Familial VUS testing is not offered. Testing family members for this variant will not contribute evidence to allow variant reclassification. Details on our VUS Resolution and Family Variant Testing Programs can be found at https://www.invitae.com/family.

Variant details

SMN1, Deletion (Entire coding sequence), homozygous, PATHOGENIC

- This variant is a gross deletion of the genomic region encompassing exon 8 (conventionally referred to as exon 7) of the SMN1 gene. Due to the sequence similarity between other exons of SMN1 and SMN2, the presence of this variant is used to infer a whole-gene deletion of SMN1.
- This variant is clearly defined as a spinal muscular atrophy (SMA) causative allele (PMID: 11839954, 18572081). It has been reported in the homozygous state in approximately 96.4% of individuals affected with 5q13-linked SMA, and in the compound heterozygous state with a second loss-of-function SMN1 allele in the remaining 3.6% of affected individuals (PMID: 10679938).
- For these reasons, this variant has been classified as Pathogenic.

ITGA7, Exon 25, c.3200G>A (p.Arg1067Gln), heterozygous, Uncertain Significance

- This sequence change replaces arginine, which is basic and polar, with glutamine, which is neutral and polar, at codon 1067 of the ITGA7 protein (p.Arg1067Gln).
- This variant is present in population databases (rs752159189, gnomAD 0.02%).
- This variant has not been reported in the literature in individuals affected with ITGA7-related conditions.
- ClinVar contains an entry for this variant (Variation ID: 451131).
- Algorithms developed to predict the effect of missense changes on protein structure and function (SIFT, PolyPhen-2, Align-GVGD) all suggest that this variant is likely to be disruptive.
- Algorithms developed to predict the effect of sequence changes on RNA splicing suggest that this variant may create or strengthen a splice site.
- In summary, the available evidence is currently insufficient to determine the role of this variant in disease. Therefore, it has been classified as a Variant of Uncertain Significance.

MEGF10, Exon 21, c.2704G>A (p.Val902Ile), heterozygous, Uncertain Significance

- This sequence change replaces valine, which is neutral and non-polar, with isoleucine, which is neutral and non-polar, at codon 902 of the MEGF10 protein (p.Val902Ile).
- This variant is present in population databases (rs138034219, gnomAD 0.1%).

- This variant has not been reported in the literature in individuals affected with MEGF10-related conditions.
- ClinVar contains an entry for this variant (Variation ID: 472737).
- Algorithms developed to predict the effect of missense changes on protein structure and function output the following: SIFT: "Tolerated"; PolyPhen-2: "Benign"; Align-GVGD: "Class C0". The isoleucine amino acid residue is found in multiple mammalian species, which suggests that this missense change does not adversely affect protein function.
- In summary, the available evidence is currently insufficient to determine the role of this variant in disease. Therefore, it has been classified as a Variant of Uncertain Significance.

MOCS1, Exon 4, c.520G>A (p.Ala174Thr), heterozygous, Uncertain Significance

- This sequence change replaces alanine, which is neutral and non-polar, with threonine, which is neutral and polar, at codon 174 of the MOCS1 protein (p.Ala174Thr).
- This variant is present in population databases (rs143573353, gnomAD 0.03%).
- This variant has not been reported in the literature in individuals affected with MOCS1-related conditions.
- ClinVar contains an entry for this variant (Variation ID: 356663).
- Algorithms developed to predict the effect of missense changes on protein structure and function output the following: SIFT: "Not Available"; PolyPhen-2: "Possibly Damaging"; Align-GVGD: "Not Available". The threonine amino acid residue is found in multiple mammalian species, which suggests that this missense change does not adversely affect protein function.
- In summary, the available evidence is currently insufficient to determine the role of this variant in disease. Therefore, it has been classified as a Variant of Uncertain Significance.

MYO18B, Exon 31, c.4984G>T (p.Val1662Leu), heterozygous, Uncertain Significance

- This sequence change replaces valine, which is neutral and non-polar, with leucine, which is neutral and non-polar, at codon 1662 of the MYO18B protein (p.Val1662Leu).
- This variant is not present in population databases (gnomAD no frequency).
- This variant has not been reported in the literature in individuals affected with MYO18B-related conditions.
- Algorithms developed to predict the effect of missense changes on protein structure and function output the following: SIFT: "Not Available"; PolyPhen-2: "Benign"; Align-GVGD: "Not Available". The leucine amino acid residue is found in multiple mammalian species, which suggests that this missense change does not adversely affect protein function.
- In summary, the available evidence is currently insufficient to determine the role of this variant in disease. Therefore, it has been classified as a Variant of Uncertain Significance.

MYO18B, Exon 35, c.5593C>T (p.His1865Tyr), heterozygous, Uncertain Significance

- This sequence change replaces histidine, which is basic and polar, with tyrosine, which is neutral and polar, at codon 1865 of the MYO18B protein (p.His1865Tyr).
- The frequency data for this variant in the population databases is considered unreliable, as metrics indicate poor data quality at this position in the gnomAD database.
- This variant has not been reported in the literature in individuals affected with MYO18B-related conditions.
- Algorithms developed to predict the effect of missense changes on protein structure and function are either unavailable or do not agree on the potential impact of this missense change (SIFT: "Not Available"; PolyPhen-2: "Possibly Damaging"; Align-GVGD: "Not Available").
- In summary, the available evidence is currently insufficient to determine the role of this variant in disease. Therefore, it has been classified as a Variant of Uncertain Significance.

PNPLA2, Exon 3, c.236G>A (p.Arg79Gln), heterozygous, Uncertain Significance

- This sequence change replaces arginine, which is basic and polar, with glutamine, which is neutral and polar, at codon 79 of the PNPLA2 protein (p.Arg79Gln).
- This variant is present in population databases (rs139576982, gnomAD 0.1%), and has an allele count higher than expected for a pathogenic variant.
- This missense change has been observed in individual(s) with clinical features of PNPLA2-related conditions (PMID: 32041611).

- ClinVar contains an entry for this variant (Variation ID: 465789).
- Algorithms developed to predict the effect of missense changes on protein structure and function are either unavailable or do not agree on the potential impact of this missense change (SIFT: "Deleterious"; PolyPhen-2: "Probably Damaging"; Align-GVGD: "Class C0").
- Experimental studies have shown that this missense change affects PNPLA2 function (PMID: 21170305).
- In summary, the available evidence is currently insufficient to determine the role of this variant in disease. Therefore, it has been classified as a Variant of Uncertain Significance.

SPEG, Exon 30, c.5635C>T (p.Arg1879Cys), heterozygous, Uncertain Significance

- This sequence change replaces arginine, which is basic and polar, with cysteine, which is neutral and slightly polar, at codon 1879 of the SPEG protein (p.Arg1879Cys).
- This variant is present in population databases (rs113853448, gnomAD 0.04%).
- This variant has not been reported in the literature in individuals affected with SPEG-related conditions.
- Algorithms developed to predict the effect of missense changes on protein structure and function (SIFT, PolyPhen-2, Align-GVGD) all suggest that this variant is likely to be disruptive.
- In summary, the available evidence is currently insufficient to determine the role of this variant in disease. Therefore, it has been classified as a Variant of Uncertain Significance.

GALC, Exon 15, c.1685T>C (p.Ile562Thr), heterozygous, Benign (Pseudodeficiency allele)

- This sequence change replaces isoleucine, which is neutral and non-polar, with threonine, which is neutral and polar, at codon 562 of the GALC protein (p.Ile562Thr).
- This variant is present in population databases (rs398607, gnomAD 61%).
- This variant is a known pseudodeficiency allele and individuals with this variant can exhibit low galactocerebrosidase activity during enzyme analysis. On its own, this variant mildly reduces enzyme activity. However, it has been shown to further reduce GALC enzyme activity when it is located on the same chromosome (in cis) with pathogenic GALC variants (PMID: 26795590, 26865610, 27126738, 27638593). Individuals with pseudodeficiency alleles may exhibit false positive results on related biochemical tests, but pseudodeficiency alleles are not known to cause disease. Although pseudodeficiency alleles do not cause disease, other carrier relatives may have abnormal enzyme testing.
- This variant is also known as p.Ile546Thr or p.I546T.
- ClinVar contains an entry for this variant (Variation ID: 92497).
- Advanced modeling of protein sequence and biophysical properties (such as structural, functional, and spatial information, amino acid conservation, physicochemical variation, residue mobility, and thermodynamic stability) performed at Invitae indicates that this missense variant is not expected to disrupt GALC protein function.
- For these reasons, this variant has been classified as a Benign pseudodeficiency allele.

Genes analyzed

This table represents a complete list of genes analyzed for this individual, including the relevant gene transcript(s). If more than one transcript is listed for a single gene, variants were reported using the first transcript listed unless otherwise indicated in the report. Results are negative unless otherwise indicated in the report. Benign and Likely Benign variants are not included in this report but are available upon request. An asterisk (*) indicates that this gene has a limitation. Please see the Limitations section for details.

GENE	TRANSCRIPT
ABAT	NM_020686.5
ABCD1	NM_000033.3
ABCD4	NM_005050.3
ABHD5	NM_016006.4
ACAD9	NM_014049.4
ACADM	NM_000016.5
ACADVL	NM_000018.3
ACAT1	NM_000019.3
ACO2	NM_001098.2
ACTA1	NM_001100.3
ADSL	NM_000026.2
ADSSL1	NM_199165.2
AGA	NM_000027.3
AGK	NM_018238.3
AGL	NM_000642.2
AGRN	NM_198576.3
AHCY	NM_000687.3
ALDH18A1	NM_002860.3
ALDH3A2	NM_000382.2
ALDH4A1	NM_003748.3
ALDH5A1	NM_001080.3
ALDH7A1	NM_001182.4
ALDOA	NM_000034.3
ALG14	NM_144988.3
ALG2	NM_033087.3
AMACR	NM_014324.5
AMN*	NM_030943.3
AMPD1	NM_000036.2
AMT	NM_000481.3
ANO5	NM_213599.2
AP1S1	NM_001283.3
AP4M1	NM_004722.3
APTX	NM_175073.2
ARG1	NM_000045.3
ARHGEF9	NM_015185.2;NM_001173479.1

GENE	TRANSCRIPT
ARSA	NM_000487.5
ASAH1	NM_177924.3
ASL	NM_000048.3
ASNS	NM_133436.3
ASPA	NM_000049.2
ASS1	NM_000050.4
ATAD1	NM_001321967.1
ATP13A2	NM_022089.3
ATP1A2	NM_000702.3
ATP2A1	NM_173201.3
ATP5D	NM_001001975.1
ATP5E	NM_006886.3
ATP6AP1	NM_001183.5
ATP6AP2	NM_005765.2
ATP7A	NM_000052.6
ATP7B	NM_000053.3
AUH	NM_001698.2
B3GALNT2	NM_152490.4
B4GAT1	NM_006876.2
BAG3	NM_004281.3
BCKDHA	NM_000709.3
BCKDHB	NM_183050.2
BCKDK	NM_005881.3
BIN1	NM_139343.2
BSCL2	NM_032667.6
BSND	NM_057176.2
BTD	NM_000060.3
C19orf12	NM_001031726.3
C1QBP	NM_001212.3
CA5A	NM_001739.1
CACNA1S	NM_000069.2
CAD	NM_004341.4
CAPN3	NM_000070.2
CASQ1	NM_001231.4
CASR	NM_000388.3

GENE	TRANSCRIPT
CAV3	NM_033337.2
CBS	NM_000071.2
CCDC78	NM_001031737.2
CD320	NM_016579.3
CFL2	NM_021914.7
CHAT	NM_020549.4
CHCHD10	NM_213720.2
CHKB	NM_005198.4
CHRNA1	NM_000079.3
CHRNB1	NM_000747.2
CHRND	NM_000751.2
CHRNE	NM_000080.3
CLCN1	NM_000083.2
CLCNKB*	NM_000085.4
CLDN16	NM_006580.3
CLDN19	NM_148960.2
CLN3	NM_001042432.1
CLN5	NM_006493.2
CLN6	NM_017882.2
CLN8	NM_018941.3
CLPB	NM_030813.5
CNNM2	NM_017649.4
CNTN1	NM_001843.3
COASY	NM_025233.6
COL12A1	NM_004370.5
COL13A1	NM_001130103.1
COL6A1	NM_001848.2
COL6A2	NM_001849.3
COL6A3	NM_004369.3
COLQ	NM_005677.3
COQ2	NM_015697.7
COQ4	NM_016035.4
COQ6	NM_182476.2
COQ7	NM_016138.4
COQ8A	NM_020247.4

GENE	TRANSCRIPT
COQ8B	NM_024876.3
COQ9	NM_020312.3
COX15	NM_004376.6
COX20	NM_198076.5
COX6B1	NM_001863.4
CP	NM_000096.3
CPOX	NM_000097.5
CPS1	NM_001875.4
CPT1A	NM_001876.3
CPT2	NM_000098.2
CRAT	NM_000755.3
CRYAB	NM_001885.2
CTDP1*	NM_004715.4
CTSD	NM_001909.4
CUBN	NM_001081.3
CYP27A1	NM_000784.3
D2HGDH	NM_152783.4
DAG1	NM_004393.5
DBH	NM_000787.3
DBT	NM_001918.3
DCAF17	NM_025000.3
DDC*	NM_000790.3
DES	NM_001927.3
DGUOK	NM_080916.2
DHCR7	NM_001360.2
DHFR	NM_000791.3
DLAT	NM_001931.4
DLD	NM_000108.4
DMD	NM_004006.2
DNA2	NM_001080449.2
DNAJB6	NM_058246.3
DNAJC12	NM_021800.2
DNM2	NM_001005360.2
DOK7	NM_173660.4
DPAGT1	NM_001382.3
DPM1	NM_003859.1
DPM2	NM_003863.3
DPM3	NM_153741.1
DYSF	NM_003494.3
EGF	NM_001963.5
EMD	NM_000117.2
ENO3	NM_053013.3
ETFA	NM_000126.3
ETFB	NM_001985.2
ETFDH	NM_004453.3
ETHE1	NM_014297.3
FA2H	NM_024306.4
FAM111A	NM_022074.3
FBXL4	NM_012160.4
FDX2	NM_001031734.3
FH*	NM_000143.3
FHL1	NM_001449.4
FKBP14	NM_017946.3
FKRP	NM_024301.4
FKTN	NM_001079802.1
FLAD1	NM_025207.4
FLNC*	NM_001458.4
FOLR1	NM_016725.2
FTL	NM_000146.3
FUCA1	NM_000147.4
FXYD2	NM_001680.4
GAA	NM_000152.3
GAD1	NM_000817.2
GALC*	NM_000153.3
GAMT	NM_000156.5
GATM	NM_001482.2
GBE1	NM_000158.3
GCDH	NM_000159.3
GCH1	NM_000161.2
GCLC	NM_001498.3
GFER	NM_005262.2
GFPT1*	NM_001244710.1
GIF	NM_005142.2
GJA1	NM_000165.4
GLA	NM_000169.2
GLB1	NM_000404.2
GLDC	NM_000170.2
GLRA1	NM_000171.3
GLRB	NM_000824.4
GLUD1	NM_005271.4
GMPPB	NM_021971.2
GNE	NM_001128227.2
GNS	NM_002076.3
GOSR2	NM_004287.3
GOT2	NM_002080.3
GPHN	NM_020806.4
GSS	NM_000178.2
GTPBP2	NM_019096.4
GUSB	NM_000181.3
GYG1	NM_004130.3
GYS1	NM_002103.4
HACD1	NM_014241.3
HADH	NM_005327.4
HADHA	NM_000182.4
HADHB	NM_000183.2
HCFC1	NM_005334.2
HEXA	NM_000520.4
HEXB	NM_000521.3
HGSNAT	NM_152419.2
HLCS	NM_000411.6
HMBS	NM_000190.3
HMGCL	NM_000191.2
HMGCS2	NM_005518.3
HNF1B	NM_000458.3
HNRNPA2B1	NM_031243.2
HNRNPDL	NM_031372.3
HPRT1	NM_000194.2
HSD17B10	NM_004493.2
IDH2	NM_002168.3
IDS*	NM_000202.6
IDUA	NM_000203.4
ISCU	NM_213595.3
ISPD	NM_001101426.3
ITGA7	NM_002206.2
IVD	NM_002225.3
KBTBD13	NM_001101362.2
KCNA1	NM_000217.2

GENE	TRANSCRIPT
KCNJ10	NM_002241.4
KCNJ2	NM_000891.2
KCTD7	NM_153033.4
KIF1A	NM_004321.6
KLHL40	NM_152393.3
KLHL41	NM_006063.2
KLHL9	NM_018847.3
L2HGDH	NM_024884.2
LAMA2	NM_000426.3
LAMB2	NM_002292.3
LAMP2	NM_002294.2
LARGE1	NM_004737.4
LDB3	NM_001080116.1;NM_001171610.1;NM_007078.3
LDHA	NM_005566.3
LIMS2	NM_001136037.2
LIPA	NM_000235.3
LMBRD1	NM_018368.3
LMNA	NM_170707.3
LMOD3	NM_198271.4
LPIN1	NM_145693.2
LRP4	NM_002334.3
MAN2B1	NM_000528.3
MAOA	NM_000240.3
MAP3K20	NM_016653.2
MAT1A	NM_000429.2
MATR3	NM_199189.2
MCCC1	NM_020166.4
MCCC2	NM_022132.4
MCEE	NM_032601.3
MCM3AP	NM_003906.4
MEGF10	NM_032446.2
MFSD8	NM_152778.2
MGME1	NM_052865.3
MICU1	NM_006077.3
MLYCD	NM_012213.2
MMAA	NM_172250.2
MMAB	NM_052845.3
MMACHC	NM_015506.2

GENE	TRANSCRIPT
MMADHC	NM_015702.2
MOCOS	NM_017947.2
MOCS1	NM_001358530.2
MOCS2A	NM_176806.3
MOCS2B	NM_004531.4
MPV17	NM_002437.4
MSMO1	NM_006745.4
MTHFR*	NM_005957.4
MTM1	NM_000252.2
MTMR14	NM_022485.4
MTR	NM_000254.2
MTRR	NM_002454.2
MUSK	NM_005592.3
MUT	NM_000255.3
MYH2	NM_017534.5
MYH3	NM_002470.3
MYH7	NM_000257.3
MYL2	NM_000432.3
MYO18B	NM_032608.6
MYOT	NM_006790.2
MYPN	NM_032578.3
NAGLU	NM_000263.3
NAGS	NM_153006.2
NAXE	NM_144772.2
NEB*	NM_001271208.1
NGLY1	NM_018297.3
NPC1	NM_000271.4
NPC2	NM_006432.3
NT5C3A	NM_016489.12
OAT*	NM_000274.3
OPA1	NM_015560.2;NM_130837.2
OPA3	NM_025136.3
ORAI1	NM_032790.3
OTC	NM_000531.5
OXCT1	NM_000436.3
PAH	NM_000277.1
PANK2	NM_153638.2
PC	NM_000920.3
PCBD1	NM_000281.3

GENE	TRANSCRIPT
PCCA	NM_000282.3
PCCB	NM_000532.4
PDHA1	NM_000284.3
PDHB	NM_000925.3
PDHX	NM_003477.2
PDP1	NM_018444.3
PDSS1*	NM_014317.4
PDSS2	NM_020381.3
PEX1*	NM_000466.2
PEX10	NM_153818.1
PEX11B	NM_003846.2
PEX12	NM_000286.2
PEX13	NM_002618.3
PEX14	NM_004565.2
PEX16	NM_004813.2
PEX19	NM_002857.3
PEX2	NM_000318.2
PEX26	NM_017929.5
PEX3	NM_003630.2
PEX5	NM_001131025.1
PEX6	NM_000287.3
PFKM	NM_000289.5
PGAM2	NM_000290.3
PGK1	NM_000291.3
PGM1*	NM_002633.2
PGM3	NM_001199917.1
PHGDH	NM_006623.3
PHKA1	NM_002637.3
PHKB	NM_000293.2;NM_001031835.2
PHYH	NM_006214.3
PLA2G6	NM_003560.2
PLEC	NM_000445.4;NM_201378.3
PNP	NM_000270.3
PNPLA2	NM_020376.3
PNPLA8	NM_015723.4
PNPO	NM_018129.3
POGLUT1	NM_152305.2
POLG	NM_002693.2
POLG2	NM_007215.3

GENE	TRANSCRIPT
POMGNT1	NM_017739.3
POMGNT2	NM_032806.5
POMK	NM_032237.4
POMT1	NM_007171.3
POMT2	NM_013382.5
PPM1K	NM_152542.4
PPT1	NM_000310.3
PRDX1	NM_002574.3
PREPL	NM_006036.4
PROSC	NM_007198.3
PRPS1	NM_002764.3
PSAT1	NM_058179.3
PSPH*	NM_004577.3
PTS	NM_000317.2
PUS1	NM_025215.5
PYGM	NM_005609.3
PYROXD1	NM_024854.3
QDPR	NM_000320.2
RAPSN	NM_005055.4
RBCK1	NM_031229.3
REPS1	NM_001286611.1
RNASEH1	NM_002936.4
RRM2B	NM_015713.4
RXYLT1	NM_014254.2
RYR1	NM_000540.2
SCN4A	NM_000334.4
SCP2	NM_002979.4
SDHA*	NM_004168.3
SELENON*	NM_020451.2
SGCA	NM_000023.2
SGCB	NM_000232.4
SGCD	NM_000337.5
SGCG	NM_000231.2
SGSH	NM_000199.3
SIL1	NM_022464.4
SLC12A1	NM_000338.2
SLC12A3	NM_000339.2
SLC13A5	NM_177550.4
SLC16A1	NM_003051.3

GENE	TRANSCRIPT
SLC18A2	NM_003054.4
SLC18A3	NM_003055.2
SLC19A1	NM_194255.2
SLC19A2	NM_006996.2
SLC19A3	NM_025243.3
SLC1A3	NM_004172.4
SLC22A5	NM_003060.3
SLC25A1	NM_005984.4
SLC25A12	NM_003705.4
SLC25A13	NM_014251.2
SLC25A15	NM_014252.3
SLC25A19	NM_021734.4
SLC25A20	NM_000387.5
SLC25A3	NM_005888.3
SLC25A32	NM_030780.4
SLC25A4	NM_001151.3
SLC25A42	NM_178526.4
SLC2A1	NM_006516.2
SLC30A10	NM_018713.2
SLC33A1	NM_004733.3
SLC39A14	NM_001128431.2;NM_015359.5
SLC39A8	NM_022154.5
SLC46A1	NM_080669.5
SLC5A7	NM_021815.2
SLC6A19	NM_001003841.2
SLC6A3	NM_001044.4
SLC6A5	NM_004211.3
SLC6A8	NM_005629.3
SLC6A9	NM_201649.3
SMCHD1	NM_015295.2
SMN1	NM_000344.3
SMN2	NM_017411.3
SMPX	NM_014332.2
SNAP25	NM_130811.2
SPEG	NM_005876.4
SPR	NM_003124.4
SQSTM1	NM_003900.4
STAC3	NM_145064.2
STIM1	NM_003156.3

GENE	TRANSCRIPT
SUCLA2	NM_003850.2
SUCLG1	NM_003849.3
SUN1	NM_001130965.2
SUN2	NM_015374.2
SUOX	NM_000456.2
SYNE1	NM_033071.3
SYNE2	NM_182914.2
SYT2	NM_177402.4
TANGO2	NM_152906.6
TAT	NM_000353.2
TAZ	NM_000116.4
TCAP	NM_003673.3
TCN1	NM_001062.3
TCN2	NM_000355.3
TH	NM_199292.2
TIA1	NM_022173.2
TK2	NM_004614.4
TMEM43	NM_024334.2
TNNT1	NM_003283.5
TNNT3	NM_006757.3
TNPO3	NM_012470.3
TOP3A	NM_004618.4
TOR1AIP1	NM_001267578.1
TPI1	NM_000365.5
TPK1	NM_022445.3
TPM2	NM_003289.3
TPM3*	NM_152263.3
TPP1	NM_000391.3
TRAPPC11	NM_021942.5
TRIM32	NM_012210.3
TRMT5	NM_020810.3
TRPM6	NM_017662.4
TSFM*	NM_001172696.1
TTN*	NM_001267550.2
TTPA	NM_000370.3
TWNK	NM_021830.4
TYMP	NM_001953.4
VAMP1	NM_014231.3
VCP	NM_007126.3

GENE	TRANSCRIPT
VMA21	NM_001017980.3
WDR45	NM_007075.3
XDH	NM_000379.3
YARS2	NM_001040436.2

Methods

- Complete list of tests performed: Invitae Comprehensive Neuromuscular Disorders Panel, Add-on Preliminary-evidence Genes for Neuromuscular Disorders, Invitae Comprehensive Muscular Dystrophy Panel, Invitae Limb-Girdle Muscular Dystrophy Panel, Invitae Comprehensive Myopathy Panel, Invitae Comprehensive Neurometabolic Disorders Panel
- Genomic DNA obtained from the submitted sample is enriched for targeted regions using a hybridization-based protocol, and sequenced using Illumina technology. Unless otherwise indicated, all targeted regions are sequenced with ≥50x depth or are supplemented with additional analysis. Reads are aligned to a reference sequence (GRCh37), and sequence changes are identified and interpreted in the context of a single clinically relevant transcript, indicated below. Enrichment and analysis focus on the coding sequence of the indicated transcripts, 20bp of flanking intronic sequence, and other specific genomic regions demonstrated to be causative of disease at the time of assay design. Promoters, untranslated regions, and other non-coding regions are not otherwise interrogated. For some genes only targeted loci are analyzed (indicated in the table above). Exonic deletions and duplications are called using an in-house algorithm that determines copy number at each target by comparing the read depth for each target in the proband sequence with both mean read-depth and read-depth distribution, obtained from a set of clinical samples. Markers across the X and Y chromosomes are analyzed for quality control purposes and may detect deviations from the expected sex chromosome complement. Such deviations may be included in the report in accordance with internal guidelines. Confirmation of the presence and location of reportable variants is performed based on stringent criteria established by Invitae (1400 16th Street, San Francisco, CA 94103, #05D2040778), as needed, using one of several validated orthogonal approaches (PubMed ID 30610921). The following analyses are performed if relevant to the requisition. For PMS2 exons 12-15, the reference genome has been modified to force all sequence reads derived from PMS2 and the PMS2CL pseudogene to align to PMS2, and variant calling algorithms are modified to support an expectation of 4 alleles. If a rare SNP or indel variant is identified by this method, both PMS2 and the PMS2CL pseudogene are amplified by long-range PCR and the location of the variant is determined by Pacific Biosciences (PacBio) SMRT sequencing of the relevant exon in both long-range amplicons. If a CNV is identified, MLPA or MLPA-seq is run to confirm the variant. If confirmed, both PMS2 and PMS2CL are amplified by long-range PCR, and the identity of the fixed differences between PMS2 and PMS2CL are sequenced by PacBio from the long-range amplicon to disambiguate the location of the CNV. Technical component of confirmatory sequencing is performed by Invitae Corporation (1400 16th Street, San Francisco, CA 94103, #05D2040778). For C9orf72 repeat expansion testing, hexanucleotide repeat units are detected by repeat-primed PCR (RP-PCR) with fluorescently labeled primers followed by capillary electrophoresis. Interpretation Reference Ranges: Benign (Normal Range): <25 repeat units, Uncertain: 25-30 repeat units, Pathogenic (Full Mutation): >=31 repeat units. A second round of RP-PCR utilizing a non-overlapping set of primers is used to confirm the initial call in the case of suspected allele sizes of 22 or more repeats. For RNA analysis of the genes indicated in the Genes Analyzed table, complementary DNA is synthesized by reverse transcription from RNA derived from a blood specimen and enriched for specific gene sequences using capture hybridization. After high-throughput sequencing using Illumina technology, the output reads are aligned to a reference sequence (genome build GRCh37; custom derivative of the RefSeq transcriptome) to identify the locations of exon junctions through the detection of split reads. The relative usage of exon junctions in a test specimen is assessed quantitatively and compared to the usage seen in control specimens. Abnormal exon junction usage is evaluated as evidence in the Sherloc variant interpretation framework. If an abnormal splicing pattern is predicted based on a DNA variant outside the typical reportable range, as described above, the presence of the variant is confirmed by targeted DNA sequencing. RNA sequencing is performed by Invitae Corporation (1400 16th Street, San Francisco, CA 94103, #05D2094793). Technical component of Fibroblast cell-culturing and gDNA extraction from skin punch biopsy is performed by Invitae Corporation (5 Technology Drive, Irvine CA 92618, #05D1052995).
- A PMID is a unique identifier referring to a published, scientific paper. Search by PMID at **http://www.ncbi.nlm.nih.gov/pubmed**.
- An rsID is a unique identifier referring to a single genomic position, and is used to associate population frequency information with sequence changes at that position. Reported population frequencies are derived from a number of public sites that aggregate data from large-scale population sequencing projects, including ExAC (**http://exac.broadinstitute.org**), gnomAD (**http://gnomad.broadinstitute.org**), and dbSNP (**http://ncbi.nlm.nih.gov/SNP**).
- A MedGen ID is a unique identifier referring to an article in MedGen, NCBI's centralized database of information about genetic disorders and phenotypes. Search by MedGen ID at **http://www.ncbi.nlm.nih.gov/medgen**. An OMIM number is a unique identifier referring to a comprehensive entry in Online Mendelian Inheritance of Man (OMIM). Search by OMIM number at **http://omim.org/**.
- Invitae uses information from individuals undergoing testing to inform variant interpretation. If "Invitae" is cited as a reference in the variant details this may refer to the individual in this requisition and/or historical internal observations.

Limitations

Based on validation study results, this assay achieves >99% analytical sensitivity and specificity for single nucleotide variants, insertions and deletions <15bp in length, and exon-level deletions and duplications. Invitae's methods also detect insertions and deletions larger than 15bp but smaller than a full exon but sensitivity for these may be marginally reduced. Invitae's deletion/duplication analysis determines copy number at a single exon resolution at virtually all targeted exons. However, in rare situations, single-exon copy number events may not be analyzed due to inherent sequence properties or isolated reduction in data quality. Certain types of variants, such as structural rearrangements (e.g. inversions, gene conversion events, translocations, etc.) or variants embedded in sequence with complex architecture (e.g. short tandem repeats or segmental duplications), may not be detected. Additionally, it may not be possible to fully resolve certain details about variants, such as mosaicism, phasing, or mapping ambiguity. Unless explicitly guaranteed, sequence changes in the promoter, non-coding exons, and other non-coding regions are not covered by this assay. Please consult the test definition on our website for details regarding regions or types of variants that are covered or excluded for this test. This report reflects the analysis of an extracted genomic DNA sample. While this test is intended to reflect the analysis of extracted genomic DNA from a referred patient, in very rare cases the analyzed DNA may not represent that individual's constitutional genome, such as in the case of a circulating hematolymphoid neoplasm, bone marrow transplant, blood transfusion, chimerism, culture artifact or maternal cell contamination. Invitae's RNA analysis is not designed for use as a stand-alone diagnostic method and cannot determine absolute RNA levels. Results from the RNA analysis may not be informative for interpreting copy number gains. CTDP1: Sequencing analysis is not offered for exon 8. FH: Sequencing analysis for exons 9 includes only cds +/- 10 bp. GALC: Deletion/duplication analysis is not offered for exon 6. TPM3: Deletion/duplication analysis is not offered for exon 10. OAT: Deletion/duplication analysis is not offered for exon 2. SMN1 or SMN2: The SMN1 gene is identical to the SMN2 gene with the exception of exon 8 (typically referred to as exon 7). This assay unambiguously detects SMN1 exon 8 copy number and sequence variants. Sequence variants outside of exon 8 will also be detected, but this assay cannot determine whether the variant is located in SMN1 or SMN2. SMN2 exon 8 copy number and the SMN2 exon 8 c.859G>C (p.Gly287Arg) modifier variant will be reported for individuals with a positive result in SMN1. CNVs of exons 1-7 of SMN1 or SMN2 (typically referred to as exons 1-6 in the literature) will not be reported. Variants in all exons with no evidence towards pathogenicity are not reported, but are available upon request. This assay cannot detect silent carriers (individuals that have 2 functional copies of SMN1 on one chromosome and zero copies on the other). Therefore a negative result for carrier testing greatly reduces but does not eliminate the chance that a person is a carrier. For individuals with 2 copies of SMN1, the residual risk of being a carrier has been reported to be 1 in 121 in African Americans, 1 in 345 in Ashkenazi Jewish individuals, 1 in 628 in Asians, 1 in 632 in Caucasians, and 1 in 1061 in Hispanic individuals (PMID: 23788250). The SMA-STAT test does not detect sequence variants in SMN1 or SMN2, and therefore cannot be used to identify compound heterozygotes. SDHA: Deletion/duplication analysis is not offered for this gene and sequencing analysis is not offered for exon 14. Sequencing analysis for exons 6-8 includes only cds +/- 10 bp. MTHFR: The NM_005957.4:c.665C>T (p.Ala222Val) (aka 677C>T) and c.1286A>C (p.Glu429Ala) (aka 1298A>C) variants are not reported in our primary report. CLCNKB: Deletion/duplication analysis is not offered for this gene. PGM1: Deletion/duplication analysis is not offered for exon 11. GFPT1: Sequencing analysis for exons 20 includes only cds +/- 10 bp. IDS: Detection of complex rearrangements not offered (PMID: 7633410, 20301451). PEX1: Sequencing analysis for exons 16 includes only cds +/- 0 bp. PDSS1: Deletion/duplication analysis is not offered for exon 2. AMN: Deletion/duplication analysis is not offered for exon 1. TTN: Exons 45-46, 147, 149, 164, 172-201 (NM_001267550.2) are excluded from analysis. TTN variants are included in the primary report based on functional effect and/or location. A complete list of variants of uncertain significance, likely benign and benign variants in TTN is available upon request. Variants are named relative to the NM_001267550.2 (meta) transcript. Variants in the coding sequence and intronic boundaries of the clinically relevant NM_133378.4 (N2A) and fetal isoforms are reported (PMID: 25589632, 29598826, 29691892, 31660661), with the exception of the PEVK tandem repeat region (172-198) (PMID: 28040389). SELENON: Deletion/duplication analysis is not offered for exon 1. FLNC: Deletion/duplication analysis is not offered for exon 47. Sensitivity and specificity for single nucleotide variants, insertions and deletions in exons 47-48 may be reduced due to the presence of segmental duplications overlapping the region. DDC: Deletion/duplication analysis is not offered for exons 10-11. TSFM: Sequencing analysis is not offered for exon 5. PSPH: Deletion/duplication and sequencing analysis is not offered for exons 4-5. NEB: Deletion/duplication analysis is not offered for exons 82-105. NEB variants in this region with no evidence towards pathogenicity are not included in this report, but are available upon request.

Disclaimer

DNA studies do not constitute a definitive test for the selected condition(s) in all individuals. It should be realized that there are possible sources of error. Errors can result from trace contamination, rare technical errors, rare genetic variants that interfere with analysis, recent scientific developments, and alternative classification systems. This test should be one of many aspects used by the healthcare provider to help with a diagnosis and treatment plan, but it is not a diagnosis itself. This test was developed and its performance characteristics determined by Invitae. It has not been cleared or approved by the FDA. The laboratory is regulated under the Clinical Laboratory Improvement Act (CLIA) as qualified to perform high-complexity clinical tests (CLIA ID: 05D2040778). This test is used for clinical purposes. It should not be regarded as investigational or for research.

This report has been reviewed and approved by:

Matteo Vatta, Ph.D., FACMG
Clinical Molecular Geneticist

INVITAE

POSITIVE RESULTS GUIDE

What positive results mean for you

Your genetic test results were positive. This means that you have a significant genetic change(s) in one or more of the genes tested. On your test report, this is called likely pathogenic variant or pathogenic variant ("mutation").

Create a plan with your healthcare provider

Whether or not you develop a disease is not determined by your genetics alone. However, your results are important. There may be tests and treatments available to help you prevent or manage a condition caused by a genetic variant. It is important to share these results with your healthcare provider so you can make informed medical decisions together.

What positive results mean for your family

Relatives can share genetic features. Your first-degree relatives (parents, children, and siblings), and even more distant relatives, may also have the same variant(s). We encourage you to share your test results with your relatives so they may discuss their potential health risks with their own healthcare providers. The medical community recommends genetic counseling and testing for family members who may be affected.

We (and others) are here to help

Genetic counseling is recommended to help you clearly and accurately understand your results so it's important to talk to your genetic counselor or other healthcare provider about your test results.

Log in to your patient portal (invitae.com) to view your results, search for a local or Invitae genetic counselor, or join Invitae's Patient Insight Network (PIN), a community where you can connect with other patients and share your experience.

This information in this results guide is meant to be used along with your genetic test results and other health information. It is not meant to replace a discussion with your healthcare provider and should not be considered or interpreted as medical advice.

1400 16th Street, San Francisco, CA 94103 • www.invitae.com • 800-436-3037 • 415-276-4164