

Client: Example Client ABC123
123 Test Drive
Salt Lake City, UT 84108
UNITED STATES

Physician: Doctor, Example

Patient: Patient, Example

DOB: 6/15/1958
Gender: Female
Patient Identifiers: 01234567890ABCD, 012345
Visit Number (FIN): 01234567890ABCD
Collection Date: 01/01/2017 12:34

Hereditary Cancer Panel, Sequencing and Deletion/Duplication

ARUP test code 2012032

Cancer Panel, Hereditary, Spcm whole Blood

Cancer Panel, Hereditary, Interp

Positive

INDICATION FOR TESTING
Patient with a family history suggestive of a hereditary cancer syndrome.

RESULT

One likely pathogenic variant was detected in the FH gene. Two variants of uncertain significance were detected, one each in the BARD1 and DICER1 genes.

LIKELY PATHOGENIC VARIANT

Gene: FH (NM_000143.3)
Nucleic Acid Change: c.1431_1433dupAAA Heterozygous
Amino Acid Alteration: p.Lys477dup
Inheritance: Autosomal Dominant

VARIANT OF UNCERTAIN SIGNIFICANCE

Gene: BARD1 (NM_000465.3)
Nucleic Acid Change: c.1226C>G; Heterozygous
Amino Acid Alteration: p.Ser409Cys
Inheritance: Autosomal Dominant

VARIANT OF UNCERTAIN SIGNIFICANCE

Gene: DICER1 (NM_177438.2)
Nucleic Acid Change: c.2776G>C; Heterozygous
Amino Acid Alteration: p.Asp926His
Inheritance: Autosomal Dominant

INTERPRETATION

One copy of the likely pathogenic variant, c.1431_1433dupAAA; p.Lys477dup, was detected in the FH gene by massively parallel sequencing and confirmed by Sanger sequencing. Pathogenic variants in FH are associated with autosomal dominant leiomyomatosis and renal cell cancer (MIM: 150800); therefore this individual is predicted to have the predisposition to hereditary cancers associated with this variant. Offspring of this individual have a 50 percent chance of inheriting this variant. Additionally, pathogenic FH variants are associated with autosomal recessive fumarate hydratase deficiency (MIM: 606812); therefore, this individual is at least a carrier of this disorder.

One copy of a variant of uncertain clinical significance, c.1226C>G; p.Ser409Cys, was detected in the BARD1 gene by massively parallel sequencing. Pathogenic BARD1 variants are

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inherited in an autosomal dominant manner, and are associated with susceptibility to breast cancer (MIM: 114480). However, it is uncertain if this variant is pathogenic or benign (see evidence for variant classifications section).

One copy of a variant of uncertain clinical significance, c.2776G>C; p.Asp926His, was detected in the DICER1 gene by massively parallel sequencing. Pathogenic DICER1 variants are inherited in an autosomal dominant manner, and are associated with an increased risk for several types of hereditary cancers/tumors including pleuropulmonary blastoma (MIM: 601200), cystic nephroma, multinodular goiter with/without Sertoli-Leydig cell tumors (MIM: 138800), and embryonal rhabdomyosarcoma (MIM: 180295); lifetime risks for different cancers/tumors vary (Doros 1993-2019). However, it is uncertain if this variant is pathogenic or benign (see evidence for variant classifications section).

No additional pathogenic variants were identified in the targeted genes by massively parallel sequencing or deletion/duplication analysis. Please refer to the background information included in this report for a list of the genes analyzed and limitations of this test.

Evidence for variant classifications:

The FH c.1431_1433dupAAA; p.Lys477dup variant (rs367543046), also known as Lys434ins, 1302insAAA, 1431insAAA, 1433_1434dupAAA, is commonly found in the compound heterozygous state in individuals with fumarase deficiency (Coughlin 1998, Ezgu 2013, Rustin 1997), and is considered the most frequent pathogenic variant associated with fumarase deficiency (Ewbank 2013). It is also reported in the heterozygous state in individuals with cancer predisposition syndrome including hereditary leiomyomatosis and renal cell cancer syndrome as well as unaffected individuals (Chen 2014, Ezgu 2013, Martinek 2015, Whitworth 2018). This variant is reported in ClinVar (Variation ID: 42095). It is found in the general population with an overall allele frequency of 0.1% (285/280892 alleles, including 2 homozygotes) in the Genome Aggregation Database. Based on available information, this variant is considered to be likely pathogenic.

The BARD1 c.1226C>G; p.Ser409Cys variant (rs786202226), to our knowledge, is not reported in the medical literature but is reported as having uncertain significance in ClinVar (Variation ID: 185502). This variant is only observed on one allele in the Genome Aggregation Database, indicating it is not a common polymorphism. The serine at codon 409 is moderately conserved, but computational analyses (SIFT, PolyPhen-2) predict that this variant is tolerated. However, given the lack of clinical and functional data, the significance of this variant is uncertain at this time.

The DICER1 c.2776G>C; p.Asp926His variant (rs1060503645), to our knowledge, is not reported in the medical literature but is reported as having uncertain significance in ClinVar (Variation ID: 412155). This variant is absent from general population databases (Exome Variant Server, Genome Aggregation Database), indicating it is not a common polymorphism. The aspartic acid at codon 926 is highly conserved, but computational analyses (SIFT: Tolerated, PolyPhen-2: Probably Damaging) predict conflicting effects of this variant on protein structure/function. Due to limited information, the clinical significance of this variant is uncertain at this time.

RECOMMENDATIONS

Medical screening and management should rely on clinical findings and family history. Due to the complex nature of this result, genetic consultation is recommended. At risk family members should be offered testing for the identified likely pathogenic FH variant (Familial Mutation, Targeted Sequencing;

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ARUP test code 2001961). Surveillance of the literature for new information concerning the uncertain variants is recommended.

LIKELY BENIGN VARIANT

Gene: ALK (NM_004304.4) Variant: c.3173-11C>T - Heterozygous
The ALK c.3173-11C>T variant (rs79339096) is reported in ClinVar (Variation ID: 335696). This variant is found in the general population with an overall allele frequency of 0.009% (25/278250 alleles) in the Genome Aggregation Database. This is an intronic variant in a weakly conserved nucleotide, and computational analyses (Alamut v.2.11) predict that this variant does not alter splicing. Based on available information, this variant is considered to be likely benign.

COMMENTS

Variants in the following region could not be detected with sufficient confidence in this sample due to technical limitations:

PHOX2B (NM_003924.3) exon 3 - chr4: 41747804 - 41748360

Unless otherwise specified, confirmation by Sanger sequencing was not performed for variants with acceptable quality metrics. Benign variants are not included in this report.

REFERENCES

Chen YB et al. Hereditary leiomyomatosis and renal cell carcinoma syndrome-associated renal cancer: recognition of the syndrome by pathologic features and the utility of detecting aberrant succination by immunohistochemistry. *Am J Surg Pathol.* 2014 May;38(5):627-37.
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Ezgu F et al. Mild clinical presentation and prolonged survival of a patient with fumarase deficiency due to the combination of a known and a novel mutation in FH gene. *Gene.* 2013 Jul 25;524(2):403-6.
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Rustin P et al. Inborn errors of the Krebs cycle: a group of unusual mitochondrial diseases in human. *Biochim Biophys Acta.* 1997 Aug 22;1361(2):185-97.
Whitworth J et al. Comprehensive Cancer-Predisposition Gene Testing in an Adult Multiple Primary Tumor Series Shows a Broad Range of Deleterious Variants and Atypical Tumor Phenotypes. *Am J Hum Genet.* 2018 Jul 5;103(1):3-18.

This result has been reviewed and approved by [REDACTED]

BACKGROUND INFORMATION: Hereditary Cancer Panel, Sequencing and Deletion/Duplication

CHARACTERISTICS: Pathogenic variants in multiple genes have been implicated in hereditary cancer often characterized by early age of onset (typically before 50 years), and the development of two or more cancers, multifocal cancers, or similar cancers in an individual or closely related family member(s). Pathogenic

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variants in the genes analyzed by this panel cause variable phenotypes and cancer risks.

EPIDEMIOLOGY: Approximately 1,700,000 new cases of cancer are diagnosed in the U.S. each year.

CAUSE: Approximately 5-10 percent of cancer is associated with a hereditary cause.

INHERITANCE: Varies depending on the gene and specific variant.

GENES TESTED: ALK, APC, ATM, ATR, AXIN2**, BAP1, BARD1, BMPR1A, BRCA1, BRCA2, BRIP1, CDH1, CDK4, CDKN1B, CDKN2A, CHEK2*, DICER1, EPCAM****, FH, FLCN, MAX, MEN1, MET, MLH1, MRE11, MSH2, MSH3**, MSH6, MUTYH, NBN, NF1**, NF2, NTHL1, PALB2, PHOX2B, PMS2, POLD1, POLE, PTEN, RAD51C, RAD51D, RB1, RECQL***, RET, SDHAF2, SDHB, SDHC*, SDHD*, SMAD4, SMARCA4**, SMARCB1, STK11, SUFU, TMEM127, TP53, TSC1, TSC2, VHL, WT1**

* - One or more exons are not covered by sequencing for the indicated gene; see limitations section below.

** - Deletion/duplication detection is not available for this gene.

*** - One or more exons are not covered by sequencing, and deletion/duplication detection is not available for this gene; see limitations section below.

**** - Deletion/duplication only; sequencing is not available for this gene.

METHODOLOGY: Targeted capture of all coding exons and exon-intron junctions of the targeted genes, including the PTEN promoter region, followed by massively parallel sequencing. Sanger sequencing was performed as necessary to fill in regions of low coverage and confirm reported variants. A custom tiled comparative genomic hybridization array (aCGH) was used to detect large deletions or duplications in the indicated subset of genes. Human genome build 19 (Hg 19) was used for data analysis. Analysis of the PMS2 gene was performed by bidirectional Sanger sequencing of coding regions and their respective exon intron boundaries as well as multiplex ligation-dependent probe amplification (MLPA). Targeted sequencing was performed for the CHEK2 c.1100delC variant.

ANALYTICAL SENSITIVITY: The analytical sensitivity of this test is approximately 99 percent for single nucleotide variants (SNVs) and greater than 93 percent for insertions/duplications/deletions from 1-10 base pairs in size. Variants greater than 10 base pairs may be detected, but the analytical sensitivity may be reduced.

LIMITATIONS: A negative result does not exclude a heritable form of cancer. This test only detects variants within the coding regions and intron-exon boundaries of the targeted genes. Regulatory region variants and deep intronic variants will not be identified and breakpoints of large deletions/duplications will not be determined. Single exon deletions/duplications or deletions/duplications less than 1kb may not be detected. Deletions/duplications/insertions of any size may not be detected by massive parallel sequencing. Diagnostic errors can occur due to rare sequence variations. In some cases, variants may not be identified due to technical limitations in the presence of pseudogenes, repetitive, or homologous regions. This assay may not detect low-level somatic variants associated with disease. Interpretation of this test result may be impacted if this patient has had an allogeneic stem cell transplantation. Non-coding transcripts were not analyzed.

The following regions are not sequenced due to technical limitations of the assay:
CHEK2(NM_001349956) exon(s) 4
CHEK2(NM_001005735) exon(s) 3

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CHEK2(NM_007194) exon(s) 10,12,13,14,15
RECQL(NM_002907) exon(s) 14,15
SDHC(NM_001035511) exon(s) 5
SDHD(NM_001276506) exon(s) 4

Single exon deletions/duplications will not be called for the following exons:

APC(NM_001127511) 1;BAP1(NM_004656) 1;BARD1(NM_000465) 1;BMPRIA(NM_004329) 9;BRCA1(NM_007300) 13;CDH1(NM_004360) 1;CDKN2A(NM_000077) 2;CDKN2A(NM_058195) 2;CHEK2(NM_001005735) 3;CHEK2(NM_007194) 11,12,14,15;FH(NM_000143) 1;FLCN(NM_144997) 8;MAX(NM_001320415) 5;MAX(NM_145113) 5;MRE11(NM_005591) 2;MSH2(NM_000251) 1;MSH2(NM_001258281) 2;MSH6(NM_000179) 10;MUTYH(NM_001128425) 1;NF2(NM_000268) 7,13,16;NTHL1(NM_002528) 3,4,5,6;PALB2(NM_024675) 1;POLDI(NM_002691) 6,18,25;PTEN(NM_000314) 8,9;PTEN(NM_001304717) 1;RAD51D(NM_002878) 1;RB1(NM_000321) 1;RET(NM_020975) 1;SDHD(NM_001276506) 4;SMARCB1(NM_003073) 5;SUFU(NM_001178133) 11;SUFU(NM_016169) 1;TP53(NM_001126113) 10;TP53(NM_001126114) 10;TSC2(NM_000548) 17,29,41;VHL(NM_000551) 1

Test developed and characteristics determined by ARUP Laboratories. See Compliance Statement C: aruplab.com/CS

VERIFIED/REPORTED DATES

| Procedure | Accession | Collected | Received | Verified/Reported |
|----------------------------------|---------------|----------------------|----------------------|---------------------|
| Cancer Panel, Hereditary, Spcm | 19-024-402530 | 1/23/2019 3:00:00 PM | 1/26/2019 9:33:00 AM | 3/7/2019 8:45:00 AM |
| Cancer Panel, Hereditary, Interp | 19-024-402530 | 1/23/2019 3:00:00 PM | 1/26/2019 9:33:00 AM | 3/7/2019 8:45:00 AM |

END OF CHART

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