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

Physician: Doctor, Example

## Patient: Patient, Example

| | |
|---|---|
| **DOB** | 10/22/1951 |
| **Gender:** | Female |
| **Patient Identifiers:** | 01234567890ABCD, 012345 |
| **Visit Number (FIN):** | 01234567890ABCD |
| **Collection Date:** | 00/00/0000 00:00 |

## BRCA1 and BRCA2-Associated HBOC Syndrome Panel, Sequencing and Deletion/Duplication

ARUP test code 3001855

BRCA Specimen: Whole Blood

BRCA Interp: Positive

RESULT
One likely pathogenic variant was detected in the BRCA1 gene.

LIKELY PATHOGENIC VARIANT
Gene: BRCA1 (NM_007294.4)
Nucleic Acid Change: c.5096G>A; Heterozygous
Amino Acid Alteration: p.Arg1699Gln
Inheritance: Autosomal dominant

INTERPRETATION
One likely pathogenic variant, c.5096G>A; p.Arg1699Gln, was detected in the BRCA1 gene by massively parallel sequencing. Pathogenic BRCA1 variants are inherited in an autosomal dominant manner, and are associated with hereditary breast and ovarian cancer (HBOC) syndrome. HBOC syndrome increases the risk for several types of hereditary cancers including female/male breast, ovarian, prostate, and pancreatic; lifetime risks for different cancers vary. National Comprehensive Cancer Network (NCCN) guidelines are available for cancer risk management in heterozygous individuals. Other genetic/environmental factors may influence an individual's risk of developing cancer. This individual's offspring have a 50 percent chance of inheriting the likely pathogenic variant.

In addition, autosomal recessive inheritance of two BRCA1 pathogenic variants may be associated with Fanconi anemia, a condition characterized by congenital anomalies  bone marrow failure, and a predisposition to malignancies (Sawyer 2015; MIM: 617883); thus, this individual is predicted to be at least a carrier of this disorder.

Please refer to the background information included in this report for a list of the genes analyzed, methodology, and limitations of this test.

Evidence for variant classification:
The BRCA1 c.5096G>A; p.Arg1699Gln variant (rs41293459), also known as 5215G>A, is reported in the literature in multiple individuals affected with breast or ovarian cancer (Couch 2015, Cunningham 2014, Rostagno 2003, Song 2014), but with reduced penetrance and an intermediate cancer risk compared to other pathogenic BRCA1 variants (Moghadasi 2018, Spurdle 2012). Functional analyses of the variant protein show conflicting results, with significant reductions in transcriptional activation (Lovelock 2007) and protein interactions (Coquelle

H=High, L=Low, *=Abnormal, C=Critical

2011), but retention of an intermediate level of homologous recombination activity (Bouwman 2013), altogether suggestive of a hypomorphic and damaging effect. This variant is classified as pathogenic by an expert panel in ClinVar (Variation ID: 37636). It is only found on six alleles in the Genome Aggregation Database, indicating it is not a common polymorphism. Computational analyses predict that this variant is deleterious (REVEL: 0.785). Additionally, another variant at this codon (c.5095C>T; p.Arg1699Trp) has been reported in individuals with breast or ovarian cancer and is considered pathogenic (Song 2014, Spurdle 2012). Based on available information, this variant is considered to be likely pathogenic with reduced penetrance and an intermediate risk for breast and ovarian cancer.

RECOMMENDATIONS
Genetic consultation is indicated, including a discussion of medical screening and management. At-risk family members should be offered testing for the identified likely pathogenic BRCA1 variant (Familial Targeted Sequencing, ARUP test code 3005867).

COMMENTS
Likely benign and benign variants are not reported.
Variants in the following region(s) may not be detected by NGS with sufficient confidence in this sample due to technical limitations:
NONE

REFERENCES
Bouwman P et al. A high-throughput functional complementation assay for classification of BRCA1 missense variants. Cancer Discov 2013 3(10):1142-55. PMID: 23867111.

Coquelle N et al. Impact of BRCA1 BRCT domain missense substitutions on phosphopeptide recognition. Biochemistry 2011 50(21):4579-89. PMID: 21473589.

Couch FJ et al. Inherited mutations in 17 breast cancer susceptibility genes among a large triple-negative breast cancer cohort unselected for family history of breast cancer. J Clin Oncol. 2015 Feb 1;33(4):304-11. PMID: 25452441.

Cunningham JM et al. Clinical characteristics of ovarian cancer classified by BRCA1, BRCA2, and RAD51C status. Sci Rep. 2014 Feb 7;4:4026. PMID: 24504028.

Lovelock P et al. Identification of BRCA1 missense substitutions that confer partial functional activity: potential moderate risk variants? Breast Cancer Res 2007 9(6):R82. PMID: 18036263.

Moghadasi S et al. The BRCA1 c. 5096G>A p.Arg1699Gln (R1699Q) intermediate risk variant: breast and ovarian cancer risk estimation and recommendations for clinical management from the ENIGMA consortium. J Med Genet. 2018 Jan;55(1):15-20. PMID: 28490613.

National Comprehensive Cancer Network. Genetic/Familial High-Risk Assessment: Breast, Ovarian, and Pancreatic (3.2023): https://www.nccn.org/professionals/physician_gls/pdf/genetics_bop.pdf

Rostagno P et al. A mutation analysis of the BRCA1 gene in 140 families from southeast France with a history of breast and/or ovarian cancer. J Hum Genet. 2003;48(7):362-6. PMID: 12827452.

Sawyer S et al. Biallelic Mutations in BRCA1 Cause a New Fanconi Anemia Subtype Cancer Discov. 2015 February; 5(2): 135-142. PMID: 25472942.

Song H et al. The contribution of deleterious germline mutations in BRCA1, BRCA2 and the mismatch repair genes to ovarian cancer in the population. Hum Mol Genet. 2014 Sep 1;23(17):4703-9.

**H=High, L=Low, *=Abnormal, C=Critical**

PMID: 24728189.

Spurdle A et al. BRCA1 R1699Q variant displaying ambiguous functional abrogation confers intermediate breast and ovarian cancer risk. J Med Genet 2012 49(8):525-32. PMID: 22889855.

This result has been reviewed and approved by [redacted]

BACKGROUND INFORMATION: BRCA1 and BRCA2-Associated HBOC Syndrome Panel, Sequencing and Deletion/Duplication

CHARACTERISTICS: Individuals with a single germline BRCA1 or BRCA2 pathogenic variant have an increased risk for breast (female and male), ovarian, fallopian tube, peritoneal, pancreatic, and prostate cancers. Additionally, BRCA2 carriers may be at increased risk for melanoma.

EPIDEMIOLOGY: 1 in 40 individuals of Ashkenazi Jewish decent or 1 in 400 individuals in the general population have a germline BRCA1 or BRCA2 pathogenic variant; 5-10 percent of breast cancers and 10-15 percent of ovarian cancers are associated with a hereditary cause.

CAUSE: Pathogenic germline variants in the tumor suppressor genes BRCA1 and BRCA2 cause hereditary breast and ovarian cancer (HBOC) syndrome. Approximately 20-60 percent of inherited breast and/or ovarian cancers are due to pathogenic germline variants in BRCA1 and BRCA2.

INHERITANCE: Autosomal dominant

CLINICAL SENSITIVITY: Greater than 98 percent of BRCA1 and BRCA2 pathogenic variants.

GENES TESTED: BRCA1* (NM_007294), BRCA2 (NM_000059)
* - One or more exons are not covered by deletion/duplication analysis for the indicated gene; see limitations section below.

METHODOLOGY: Probe hybridization-based capture of all coding exons and exon-intron junctions of the BRCA1 and BRCA2 genes, followed by massively parallel sequencing. Sanger sequencing was performed as necessary to fill in regions of low coverage and to confirm reported variants that do not meet acceptable quality metrics. A proprietary bioinformatic algorithm was used to detect large (single exon-level or larger) deletions or duplications in the indicated genes. Large deletions/duplications confirmed using an orthogonal exon-level microarray. Human genome build 19 (Hg 19) was used for data analysis.

ANALYTICAL SENSITIVITY/SPECIFICITY: The analytical sensitivity is approximately 99 percent for single nucleotide variants (SNVs) and greater than 93 percent for insertions/duplications/deletions (indels) from 1-10 base pairs in size. Indels greater than 10 base pairs may be detected, but the analytical sensitivity may be reduced. Deletions of 2 exons or larger are detected with sensitivity greater than 97 percent; single exon deletions are detected with 62 percent sensitivity. Duplications of 3 exons or larger are detected at greater than 83 percent sensitivity. Specificity is greater than 99.9 percent for all variant classes.

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 BRCA1 and BRCA2 genes. Deletions/duplications/insertions of any size may not be detected by massively parallel sequencing. Regulatory region variants and deep intronic variants will not be identified. Precise breakpoints for large deletions or duplications are not

determined in this assay and single exon deletions/duplications may not be detected based on the breakpoints of the rearrangement. The actual breakpoints for the deletion or duplication may extend beyond or be within the exon(s) reported. This test is not intended to detect duplications of 2 or fewer exons in size, though these may be identified. Single exon deletions are reported but called at a lower sensitivity. Diagnostic errors can occur due to rare sequence variations. In some cases, variants may not be identified due to technical limitations caused by the presence of pseudogenes, repetitive, or homologous regions. This test is not intended to detect low-level mosaic or somatic variants, gene conversion events, complex inversions, translocations, mitochondrial DNA (mtDNA) variants, or repeat expansions. Interpretation of this test result may be impacted if this patient has had an allogeneic stem cell transplantation. Noncoding transcripts were not analyzed.

Single exon deletions/duplications will not be called for the following exons: BRCA1 (NM_007294) 2.

This test was developed and its performance characteristics determined by ARUP Laboratories. It has not been cleared or approved by the U.S. Food and Drug Administration. This test was performed in a CLIA-certified laboratory and is intended for clinical purposes.

Counseling and informed consent are recommended for genetic testing. Consent forms are available online.

VERIFIED/REPORTED DATES

| Procedure | Accession | Collected | Received | Verified/Reported |
|---|---|---|---|---|
| BRCA Specimen | 23-118-402887 | 00/00/0000 00:00 | 00/00/0000 00:00 | 00/00/0000 00:00 |
| BRCA Interp | 23-118-402887 | 00/00/0000 00:00 | 00/00/0000 00:00 | 00/00/0000 00:00 |

END OF CHART

**H=High, L=Low, *=Abnormal, C=Critical**