Chronic Lymphocytic Leukemia Mutation Panel by Next Generation Sequencing

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Patient: [redacted]
DOB: [redacted] Age: 60 Gender: M
Patient Identifiers: [redacted]
Visit Number (FIN): [redacted]

Client: [redacted]
Physician: [redacted]

ARUP Test Code: 3001858
Collection Date: 04/01/2021
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Comment:

Submitted diagnosis or diagnosis under consideration for variant interpretation: Chronic lymphocytic leukemia (CLL)

TIER 1: Variants of Known Clinical Significance in Hematologic Malignancies

Gene	Transcript ID	DNA Variant	Protein Variant	Variant Frequency
NOTCH1	NM_017617.3	c.7541_7542del	p.Pro2514fs	8.4%
KRAS	NM_004985.4	c.38G>A	p.Gly13Asp	15.7%

TIER 2: Variants of Unknown Clinical Significance in Hematologic Malignancies

Gene	Transcript ID	DNA Variant	Protein Variant	Variant Frequency

None Detected

Interpretation

NOTCH1 c.7541_7542del - NOTCH1 encodes a transmembrane receptor that functions as a transcription factor that regulates stem cell maintenance, cell differentiation, proliferation, and apoptosis (1) (10). NOTCH1 activating mutations occur in various hematologic malignancies including approximately 5-22% of chronic lymphocytic leukemia (CLL) patients (4) (11) (12) (13) (15).These mutations, most commonly Pro2514fs, are often frameshift and nonsense mutations in the C-terminal heterodimerization (HD) and PEST domains as well as the 3' UTR of NOTCH1 (6). This particular frameshift mutation (Pro2514fs) is a recurrent activating mutation within the PEST domain (15). NOTCH1 activating mutations are associated with poor prognosis, including increased risk of progression and resistance to therapy in patients with CLL (15).

KRAS c.38G>A - The RAS genes (KRAS and NRAS) encode a family of membrane-associated signal-transduction proteins involved in regulating cell growth (2) (3). RAS mutations are found in a variety of hematologic malignancies including approximately 2-7% of patients with CLL (9) (14). These mutations predominantly occur at codons 12, 13, 61, 117 and 146, leading to activation of the RAS-ERK pathway (8) (9). This particular missense mutation has been reported in lymphoid malignancies (5). In CLL, one study concluded that RAS mutations were not associated with overall survival (7). Another study showed that RAS mutations were associated with shorter therapy-free survival and patients with KRAS mutations showed a higher incidence of somatic trisomy 12 (14). Correlation with cytogenetic findings is recommended.

Low Coverage Regions

This list contains regions where the average sequencing depth (number of times a particular position is sequenced) for 20% or more of the region is below our stringent cutoff of 300. Sensitivity for detection of low allelic frequency mutations may be reduced in areas with low depth of coverage. The sequencing reads from these regions were manually reviewed. If high quality variants are detected in these regions they will be listed above in Tier 1 or Tier 2.

None

This result has been reviewed and approved by [redacted]

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BACKGROUND INFORMATION: Chronic Lymphocytic Leukemia (CLL) Mutation Panel by Next Generation Sequencing

CHARACTERISTICS: Chronic lymphocytic leukemia (CLL) is a hematopoietic disorder characterized by monoclonal B cell proliferation. Recent studies have identified recurrently mutated genes with diagnostic and/or prognostic impact in CLL and other lymphoid malignancies. The presence of certain mutations may inform clinical management. This multi-gene panel by massively parallel sequencing (next generation sequencing) is a more cost-effective approach when compared to the cost of multiple single-gene tests. This test can be used to complement the morphologic and cytogenetic workup of CLL and other lymphoid malignancies.

GENES TESTED: ATM, BCL2, BIRC3*, BRAF, BTG1, BTK, CARD11, CD79B, CXCR4, DDX3X, FBXW7, IKZF3, KRAS, MAP2K1, MED12, MGA, MYD88, NOTCH1, NRAS, PLCG2, POT1, RPS15*, SAMHD1, SF3B1, TP53, XPO1, ZMYM3

* - One or more exons of the preferred transcript were not covered by sequencing for the indicated gene; see limitations

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section below.
METHODOLOGY: Genomic DNA was isolated from peripheral blood or bone marrow then enriched for the targeted exonic regions of the tested genes. The variant status of the targeted genes was determined by massively parallel sequencing. The hg19 (GRCh37) human genome assembly was used as a reference for identifying genetic variants.
LIMITATIONS: Variants outside the targeted regions or below the limit of detection are not identified. Variants in regions that are not included in the preferred transcript for the targeted genes are not detected. In some cases, variants may not be identified due to technical limitations in the presence of pseudogenes or in repetitive or homologous regions. It is also possible some insertion/deletion variants may not be identified. The following regions were not sequenced due to technical limitations of the assay:
BIRC3 (NM_001165) exon 5
RPS15 (NM_001018) exon 3
LIMIT OF DETECTION (LOD): 5 percent variant allele fraction (VAF) for single nucleotide variants (SNV) and small variants less than 24 base pairs (bp). Variants greater than 24bp may be detected at LOD, but the analytical sensitivity may be reduced.
ANALYTICAL SENSITIVITY: The positive percent agreement (PPA) estimate for the respective variant classes (with 95 percent credibility region) are listed below. Genes included on this test are a subset of a larger methods-based validation from which the PPA values are derived.
Single nucleotide variants (SNVs): 96.9 percent (95.1 - 98.1 percent)
Insertions/Duplications (1-24bp): 98.1 percent (95.5 - 99.3 percent)
Insertions/Duplications (greater than 24bp): Greater than 99 percent (92.9 - 100.0 percent)
Deletions (1-24bp): 96.7 percent (92.8 - 98.7 percent)
Deletions (greater than 24bp): 90 percent (79.5 - 96.1 percent)
Multi-nucleotide variants (MNVs): 97 percent (93.0 - 99.0 percent)

CLINICAL DISCLAIMER: Results of this test must always be interpreted within the context of clinical findings and other relevant data and should not be used alone for a diagnosis of malignancy. This test is not intended to detect minimal residual disease.

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

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