NEW TEST
NOTIFICATION DATE: 01/09/2023
EFFECTIVE DATE: 01/09/2023

MYELOID NEOPLASM (AML, MDS) NGS
Epic Test Code: LAB1231014

EXPLANATION: The Molecular Diagnostics Laboratory at WVU Hospital will transition testing for **Acute Myeloid Leukemia (AML) and Myelodysplastic Syndromes (MDS) by next generation sequencing (NGS)** on January 9, 2023.

- New Orderable: **Myeloid Neoplasm (AML, MDS) NGS (LAB1231014)**
- Replaced Test: LeukoVantage, Acute Myeloid Leukemia (LAB15096)

REPORTING: The variant interpretations are categorized into Tiers based on evidence in the current medical and scientific literature and by adapting guidelines recommended by AMP, ASCO, and CAP [J Mol Diagn. 2017 Jan; 19(1): 4–23.]. Clinical significance may not be completely established for all the targeted genes/variants identified (including Tier 3 variants) in this panel and such genes/variants will NOT be reported. Variants reported in this test should be interpreted in conjunction with other clinical and laboratory findings before using the results to diagnose a myeloid neoplasm or malignancy.

INTERPRETATION: Certain alterations in genes identified by this panel can assist in the diagnosis, prognosis, and treatment of myeloid neoplasms. Findings may aid enrollment in clinical trials.

LIMITATIONS:

1. Variants are only detected within genes (exons) interrogated in this panel. Variants outside the targeted region (exons/ preferred transcript) will not be detected.
2. Specimens containing at least 20% abnormal cells are preferred to ensure reliable results. At least 10% abnormal cells are required to detect variants (SNVs/indels) near the stated level of sensitivity of the assay (5% Variant Allelic Fraction, VAF). A low sample tumor cell percentage may affect the true mutation VAF and/or sensitivity.
3. Insertions up to 102 bp and deletions up to 52 bp have been detected/validated in clinical specimens. Sensitivities for detecting larger size variants are unknown; these may or may not be detected.
4. Polymorphisms, benign and likely benign variants are not reported. Some variants classified as Variants of Unknown Clinical Significance (VUS/Tier 3) may represent rare or low frequency polymorphisms and may not be reported.
5. This targeted panel does not specifically distinguish between somatic and germline alterations for genes with a VAF close to 50% or above.
6. For some targeted regions, the depth of sequencing coverage may vary. Variants may not be identified in certain targeted regions due to the presence of pseudogenes, repetitive regions, polymorphisms in primer binding sites or other technical limitations.
7. Large genomic alterations (insertions/deletions/duplications/rearrangements) are not detected by this assay.
8. This test is NOT intended to detect minimal residual disease.
9. Prior or current treatment for a hematologic malignancy could affect the results.

METHOD: Genomic DNA extracted from peripheral blood or bone marrow samples is used to generate a library targeting the exonic regions of the genes listed below utilizing Anchored-Multiplex PCR (AMP™) enrichment chemistry. Massively parallel sequencing is performed using Illumina® next-generation sequencing (NGS) instruments to determine the variant status of the targeted genes. Archer Analysis Unlimited (AAU) platform is used to analyze data. This test was developed, and performance characteristics determined by the WVUH-Molecular Diagnostic Laboratory consistent with CLIA requirements. It has not been cleared or approved by the US Food and Drug Administration (FDA).

Genes Tested in the WVUH-Molecular Diagnostic Myeloid Targeted Panel (MD-MP) include:

ABL1: NM_005157 exon: 4-10; **ANKRD26**: NM_014915 exon:1(c.-113-c.-134); **ASXL1**: NM_015338.5 exon:1-13; **ASXL1**:NM_001164603.1 exon:5; **BCOR**: NM_017745 exon:2-15; **BCOR**:NM_001123385 exon:8; **BCORL1**:NM_021946 exon:1-12; **BRAF**:NM_004333 exon:3-15; **CALR**:NM_004343 exon:8-9; **CBL**: NM_005188 exon:2-16; **CBLB**: NM_170662 exon:3,9-10; **CEBPA**:NM_004364 exon:1; **CSF3R**:NM_156039 exon:17; **CSF3R**:NM_172313 exon:10,18; **CSF3R**:NM_000760 exon:14-16; **CUX1**: NM_001202543 exon:15-24; **CUX1**:NM_001913 exon:1-23; **CUX1**: NM_181552 exon:1; **DDX41**: NM_016222 exon: 1-17; **DNMT3A**: NM_022552 exon: 2-3,5-23; **DNMT3A**: NM_153759 exon: 1-2; **DNMT3A**: NM_175630 exon:4; **ETNK1**: NM_018638 exon:3; **ETV6**:NM_001987 exon:1-8; **EZH2**: NM_004456 exon: 2-20; **FLT3**: NM_004119 exon:8-21; **GATA1**: NM_002049 exon:2; **GATA2**: NM_032638 exon:2-6; **GNAS**: NM_000516 exon: 8-11; **IDH1**: NM_005896 exon:3-4; **IDH2**: NM_002168 exon: 4-6; **JAK2**: NM_004972 exon:12-25; **KDM6A**: NM_021140 exon: 1-29; **KDM6A**: NM_001291415 exon:14; **KIT**: NM_000222 exon: 1-2,5,8-15,17-18; **KMT2A**: NM_005933 exon: 1-36; **KMT2A**: NM_001197104 exon: 14; **KRAS**: NM_004985 exon: 2-4; **MPL**: NM_005373 exon:10,12; **NF1**: NM_000267 exon:1-57; **NF1**: NM_001128147 exon:15; **NF1**: NM_001042492 exon: 31; **NPM1**: NM_002520 exon: 11; **NRAS**: NM_002524 exon: 2-5; **PHF6**: NM_032335 exon:2-8; **PHF6**: NM_001015877 exon:10; **PHF6**: NM_032458 exon:9; **PPM1D**: NM_003620 exon:6; **PTPN11**: NM_002834 exon:3,4,7,8,12,13; **PTPN11**: NM_080601 exon:11; **RAD21**: NM_006265 exon:2-14; **RUNX1**: NM_001754 exon:2-3,5-9; **RUNX1**: NM_001122607 exon:1,5; **SETBP1**: NM_015559 exon:4 (p.799-p.950); **SF3B1**: NM_012433 exon:13-21; **SH2B3**: NM_005475 exon:2-8; **SMC1A**: NM_001281463 exon:2; **SMC3**: NM_005445 exon:10,13,19,23,25,28; **SRSF2**: NM_003016 exon:1-2; **STAG2**: NM_006603 exon:2-33; **STAG2**: NM_001042749 exon:32; **TET2**: NM_001127208 exon:4-11; TET2: NM_017628 exon:3; **TP53**: NM_000546 exon:1-11; **TP53**: NM_001276696 exon:10; TP53: NM_001276695 exon:10; **U2AF1**: NM_006758 exon:2,6,7; **U2AF1**: NM_001025204 exon:6; **U2AF2**: NM_007279 exon:1-12; **WT1**: NM_000378 exon:1-9; **WT1**: NM_001198552 exon:8; **ZRSR2**: NM_005089 exon:1-11

REFERENCE VALUES: No clinically reportable variant detected

SAMPLE REQUIREMENTS:

- Preferred Specimen(s): 5 mL bone marrow collected in an EDTA (lavender-top) tube having lesions cells ≥20% or a minimum ≥10% determined either by morphology (blasts), or flow cytometry
- Alternative Specimen(s): 5 mL Whole blood collected in an EDTA (lavender-top) tube or extracted DNA minimum 200 ng (5-10 ng/ μL)
- Minimum Volume: 3 mL whole blood or bone marrow
- NOTE: Heparinized samples are not accepted. Heparin may act as an inhibitor of PCR.

SPECIMEN STABILITY INFORMATION:

- Room temperature: 2 days
- Refrigerated: 7 days
- Frozen: Unacceptable
- Genomic DNA (once sample has been extracted) can be stored at +2-8°C for one week or -20°C for 1 month

CPT CODE: 81450
DAYS(S) TEST SET UP: Weekly (Sample should be received by Friday: 12:30 pm to be included in the next run)
TURNAROUND TIME: 14-21 days

QUESTIONS ABOUT THIS TESTING

Subit Barua PhD, FACMG, Director Molecular Pathology, subit.barua@hsc.wvu.edu
Joyce Nicola, Molecular Laboratory Supervisor, jvincent@wvumedicine.org

ADDITIONAL INORMATION IS AVAILABLE ONLINE

WVU Medicine Online Test Catalog