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HLID#: **PATIENT NAME:**
PATIENT ID#: **DOB :** **SEX:**
NPI: **ORDERING PHYSICIAN:**
SPECIMEN TYPE: Bone Marrow Aspirate
COLLECTION DATE: **RECEIPT DATE:**
REPORT DATE: **ICD Code:** C92.00 **UNITS:**
CLINIC ID#:
CPT: 81450
ACCOUNT:

Last Name, First Name HLID#

MOLECULAR ANALYSIS REPORT

Specimen Type: Bone Marrow Aspirate (Client Pathology Number)

Clinical History/Indications: An adult patient with a clinical history of AML.

AML (NGS) mutation panel results:

LOW POSITIVE – *continued presence*

– *clinically significant/ potentially clinically significant:* **TET2, ZRSR2**

variant	collection date	HLID	Specimen	variant allelic frequency
ZRSR2 NM_005089.3:c.446dupA; NP_005080.1:p.Asn149LysfsTer23	3/2024	X-XXXX	PB	7.59%
	1/2025	X-XXXX	BMA	13.27%
	8/2025	X-XXXX	BMA	56.44%
	9/2025	X-XXXX	BMA	3.11%
	2/2026	X-XXXX	BMA	**0.86%**
TET2 NM_001127208.2:c.4280A>T; NP_001120680.1:p.Asp1427Val	1/2025	X-XXXX	BMA	7.42%
	8/2025	X-XXXX	BMA	35.10%
	9/2025	X-XXXX	BMA	9.50%
	2/2026	X-XXXX	BMA	**2.52%**

Analysis/Conclusions:

- The specimen tested ***low positive*** for the continued presence of the previously detected missense mutation in codon 1427 (exon 10) of the **TET2** gene with a variant allelic frequency (VAF) of 2.52%.

Missense mutations in the ten-eleven translocation 2 tumor suppressor gene (TET2) have been reported in myeloid malignancies, including AML and may confer a poor prognosis in intermediate-risk AML [Chou et al. Blood. 2011; 118:3803-10; Metzeler et al. J Clin Oncol. 2011; 29:1373-81; Weissmann et al. Leukemia. 2012; 26:934-42].

- The specimen tested ***low positive*** for the continued presence of the previously detected duplication (frameshift) mutation in exon 7 of the **ZRSR2** gene with a VAF of 0.87%. *Please note that this finding is at or below the sensitivity level of the analysis (established at ~2-5% for diagnostics cases and ~1-0.1% for monitoring variant) and therefore needs to be interpreted with caution.*

Loss-of-function nonsense and frameshift mutations in ZRSR2, a member of the U2AF1-related protein family involved in the spliceosome pathway have been reported in MDS and other myeloid malignancies. In MDS mutations are associated with transformation to AML and adverse prognosis; in AML mutations are frequently associated with secondary AML [Madan et al. Blood. 2014; 124:4609; Larson et al. Mol Cancer Res. 2013;11:815-827; Lindsley et al. Blood. 2015;125:1367-76].

- *No additional variants were detected in the genes targeted by the AML panel.*

The AML panel targets the following genes:

ANKRD26, ASXL1, ***BCOR, BCORL1, CEBPA,*** *CSF3R, DDX41,* ***DNMT3A, ETV6, EZH2,*** *FLT3 (ITD+TKD), GATA2, IDH1, IDH2, JAK2, KIT, KRAS, NPM1, NRAS, NF1,* ***PHF6,*** *PPM1D, PTPN11,* ***RAD21, RUNX1,*** *SETBP1, SF3B1, SRSF2,* ***STAG2, TET2, TP53,*** *U2AF1, WT1,* ***ZRSR2***

Genes marked in **bold** have full coding sequence (CDS) coverage; for further information on covered regions, including transcripts visit our website: https://www.hematologics.com

Please note:

- The presence of additional variants below the sensitivity limit (*established at ~2-5% for diagnostics cases and ~1-0.1% for monitoring variants)* cannot be ruled out.
- Currently classified benign variants detected by this assay are not reported.
- Variant classification is based on current versions (date of the report) of the following public databases: NCBI Short Genetic Variations Database (dbSNP) and Clinically Relevant Sequence Variations (ClinVar), as well as literature and published guidelines. Clinical knowledge can change over time and variant classification can change in significance.
- Variants in the above genes may also occur in healthy older individuals (clonal hematopoiesis of indeterminate potential (CHIP)). [Jaiswal et al. N Engl J Med. 2014;371:2488-2498; Genovese et al. N Engl J Med. 2014;371:2477-2487]
- A variant allele frequency of ~50% or 100% can be in keeping with heterozygous or homozygous germline mutations. **This analysis cannot distinguish between germline and somatic mutations.** Genetic counseling may be considered if a germline variant is suspected.
- Clinical and histological correlation required.

Method: Next-generation deep amplicon sequencing (NGS) was utilized to simultaneously detect mutations in selected genes and gene regions (coding sequence and splice region) recurrently reported mutated in AML.[1-3] Fastq files were aligned to hg19 version of the human genome and single-nucleotide and small indel calling was performed using the PiVAT software from Pillar Bioscience version: 2023.1.0. Secondary analysis was performed using NextGENe software v2.4.1 -also utilized to visualize reads. Variants were annotated utilizing Variant Studio v3.0.12 with cDNA and amino acid changes, and the number of reads supporting the variant allele and population allele frequency. The assay is validated to detect deletions up to 52 bp, and insertions to a size of approximately 100 bp. Orthogonal polymerase chain reaction combined with fragment length analysis is performed to detect FLT3 internal tandem duplications >100 bp in size. Gene fusions and intronic gene regions are not targeted by this assay.

References:

1. Pollyea et al. J Natl Compr Canc Netw 2023;21(5):503–513
2. Döhner et al. Blood 2022; 140 (12): 1345–1377
3. Huber, S., Baer, C., Hutter, S. et al. Leukemia. 2023; 37, 1413–1420

Although molecular testing is highly accurate, rarely false-positive and false-negative diagnostic errors may occur. DW/MRL/DX

Electronically signed by: Arun K. Nalla, Ph.D., HCLD (ABB), Associate Director of Molecular Analysis - 02/2026 15:20 PT; Michael H. Kalnoski, MD, FCAP - 02/2026 16:25 PT