NPM1 mutation to Monitor Acute Myeloid Leukemia

The UNC Hospitals Molecular Genetics Laboratory measures *NPM1* mutant transcripts as a marker of tumor burden in serial blood specimens of leukemia patients.

NPM1 Mutation as a Marker of Acute Myeloid Leukemia

Acute myeloid leukemia (AML) management increasingly relies on molecular test results that add value for monitoring tumor burden in serial samples collected over time. *NPM1* mutation is the most prevalent of the commonly tested molecular events in AML, occurring in approximately 50% of affected adults, and has been shown to be a molecularly stable event which allows for use as a clinical marker of relapse and to monitor minimal residual disease.

A somatic, frameshift mutation in exon 12 of *NPM1* confers a more favorable prognosis in AML that is otherwise intermediate prognosis. Many variants of mutant *NPM1* have been described, typically 4-base insertions, yet the majority (~80%) are characterized as mutation type A (956 ins tctg). This common variant is measurable using quantitative rtPCR, and levels reflect tumor burden and may serve as harbingers of relapse.

Laboratory Measurement of *NPM1* type A mutation by Q-rtPCR:

Specimens must be **delivered promptly** to the laboratory to minimize RNA degradation. The preferred sample is EDTA blood (3mL purple-top), although EDTA marrow is also acceptable (0.5mL). RNA is extracted and converted to cDNA that is PCR-amplified using primers flanking the insertion hotspot, and a fluorescent probe permits detection of PCR products in real time. A separate control assay targeting *ABL1* cDNA normalizes for the amount of amplifiable cDNA in the sample. Results relative to internal and external calibrators are reported as the number of "mutant *NPM1* transcripts per 10,000 cell equivalents". Analytic sensitivity is 1 in 10^{-4} (one tumor cell in 10,000 normal cells). In serial specimens, changes of 10-fold (one log) are considered to be significant.

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To consult a pathologist about indications for testing or the significance of a result, call the Molecular Genetics Lab at (984) 974-1825 or email Karen Weck at Karen.Weck@unchealth.unc.edu

Website: Molecular Genetics | McLendon Clinical Laboratories | UNC Medical Center