UNC HOSPITALS

MEMORANDUM#9

TO: UNC Hospitals Attending Physicians, Housestaff, Nursing Coordinators, Department Heads and Supervisors

FROM: Margaret L. Gulley, MD, Director of Molecular Pathology
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DATE: May 27, 2008

SUBJECT: New Molecular tests assess prognosis in Acute Myelogenous Leukemia

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The UNCH Molecular Genetics Laboratory now offers mutation analysis of the *FLT3* and *NPM1* genes to assess prognosis in acute myelogenous leukemia (AML) of normal karyotype. Internal tandem duplication (ITD) in the *FLT3* gene confers a worse prognosis while insertional mutation in the *NPM1* gene confers a better prognosis, while AML harboring both changes has an intermediate prognosis.

Clinical indications for testing: Testing is recommended in AML patients whose tumor is cytogenetically normal, particularly when prognostic information would impact therapeutic decision-making.

Sample requirements: 3ml EDTA blood (purple-top tube) or 1 mL marrow aspirate (EDTA), refrigerated up to 2 days before receipt in laboratory. The specimen must contain **at least 10% leukemic cells**.

Turn around time: Assays are performed weekly.

Method and Reporting: DNA is extracted, amplified using PCR primers targeting exons 14 and 15 of *FLT3*, and sized by capillary electrophoresis to detect an abnormally large amplicon implying ITD. An *NPM1* gene segment is amplified using PCR primers targeting exon 12 followed by capillary electrophoresis to detect a four-nucleotide enlargement of one allele. Results for both *FLT3* and *NPM1* assays are reported as positive or negative and interpreted for their prognostic significance.

References:

1. Gale RE et al. The impact of FLT3 internal tandem duplication mutant level, number, size, and interaction with NPM1 mutations in a large cohort of young adult patients with acute myeloid leukemia. *Blood.* 2008;111:2776.
2. Scholl S et al. Clinical impact of nucleophosmin mutations and Flt3 internal tandem duplications in patients older than 60 yr with acute myeloid leukaemia. *Eur J Haematol.* 2008;80:208.
3. Schlenk RF et al. Mutations and treatment outcome in cytogenetically normal acute myeloid leukemia. *N Engl J Med.* 358:1909, 2008.
4. Murphy KM et al. Detection of FLT3 internal tandem duplication and D835 mutations by a multiplex polymerase chain reaction and capillary electrophoresis assay. *J Mol Diagn.* 2003;5:96.
5. Chen W et al. Nucleophosmin gene mutations in acute myeloid leukemia. *Arch Pathol Lab Med.* 2006;130:1687.

For further information, consult the McLendon Clinical Laboratories Manual of Pathology and Laboratory Medicine Clinical Services (Website= http://labs.unchealthcare.org/directory/molecular_pathology/index_html), or contact the Molecular Genetics Laboratory at 966-4408 or Dr. Gulley at margaret_gulley@med.unc.edu.