

FLT3 Gene Mutation Analysis

Test: FLT3 D835 and FLT3 ITD

CPT code: 81245 . 81246

Methodology:

The FLT3 ITD gene region (exon 14) and FLT3 TKD region (exon 20) are amplified. The size of the ITD PCR product is determined by capillary electrophoresis to assess the percent mutant allele burden. The D835 PCR product is digested with EcoRV and assessed by capillary electrophoresis to determine the mutant burden.

Clinical Use:

FLT3 is a tyrosine kinase receptor with important role in hematopoietic stem cell proliferation and survival. Activating mutations in FLT3-ITD are seen in approximately 20-30% of acute myeloid leukemia (AML) patients. There are two major types of clinically significant FLT3 mutations: can be seen as either internal tandem duplication (ITD) within the juxtamembrane domain of FLT3 gene or as a missense point mutation within tyrosine kinase domain (TKD) at codon 835. FLT3-ITD mutations range from 3-400 bp and are always in-frame. Since the presence of ITD mutations in normal cytogenetics AML patients are associated with poorer outcomes and more aggressive therapy, knowledge of these mutations may benefit the patients from the currently available clinical trials using FLT3 inhibitors.

Specimen Requirements:

- 5-10 mL of blood or bone marrow-purple (EDTA) tube or yellow (ACD) tube within 24 hours of collection.
- DNA 200ng at a minimum of 25ng/μL.

Note: If sample cannot arrive at laboratory within 24 hours of draw, refrigerate until sample can be transported and then transfer it at room temperature.

Test Performed:

Weekly

Turn Around Time:

7-10 days

References:

1. Marcucci et al. 2011. Molecular Genetics of adult acute myeloid leukemia: prognostic and therapeutic implications. *Journal of Clinical Oncology* 29: 475-486.
2. Gale et al. 2008. 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* 111: 2776-2784.
3. Kiyoi et al. 1997. Internal tandem duplication of FLT3 associated with leukocytosis in acute promyelocytic leukemia. *Leukemia*. 11: 1447-1452.
4. Murphy et al. 2003. Detection of FLT3 Internal Tandem Duplication and D835 Mutations by a Multiplex Polymerase Chain Reaction and Capillary Electrophoresis Assay. *Journal of Molecular diagnostics* 5(2): 96-102.
5. Acute Myeloid Leukemia, Clinical Practice Guidelines in Oncology, (v.2.2007) National Comprehensive Cancer Network.