Lab Dept: Anatomic Pathology

Test Name: FLT3 ASSAY (UWASH)

General Information

Lab Order Codes:	FLT3
Synonyms:	FLT3 ITD Gene Mutation Analysis; FLT3 ITD DNA Screen
CPT Codes:	81245 – Chromosome analysis for breakage syndromes; baseline Sister Chromatid Exchange (SCE), 20-25 cells
Test Includes:	PCR amplification of the juxtamembrane portion of the FLT3 gene. Specific for internal tandem duplication mutations.

Logistics

Test Indications:	Genomic DNA is amplified using flanking primers and then size fractionated by capillary electrophoresis. For positive patients, a FLT3 ITD to wild type allelic ratio is calculated and reported. This test should be performed on AML patients at diagnosis. FLT3 mutations tend to be unstable and can change with relapse, so this assay is not a reliable method for detecting minimal residual disease in AML. A mutation in the FLT3 gene on chromosome 13 results from internal tandem duplications (ITD) in exons 14 and 15 of the juxtamembrane portion of the gene and causes activation of the FLT3 protein. Approximately 20-30% of patients with acute myeloid leukemia have this mutation which has been associated with adverse prognosis.
Lab Testing Sections:	Chemistry - Sendouts
Referred to:	Molecular Hematopathology @ Seattle Cancer Care Alliance, 825 Eastlake Ave, E Room G7801, Seattle, Washington 98109 (UWASH Test Code: **FLT3**)
Phone Numbers:	MIN Lab: 612-813-6280 STP Lab: 651-220-6550
Test Availability:	Daily, 24 hours (Reference lab performs Monday through Friday)
Turnaround Time:	2 days, reflex testing may take an additional 5 days.
Special Instructions:	N/A

Specimen

Specimen Type:	Blood or Bone Marrow
Container:	Blood: Lavender top (EDTA) tube Bone Marrow: Dry syringe **immediately** transferred to lavender EDTA tube to prevent clotting. Alternate: Blood in green top no gel (heparin) or bone marrow first collected in heparinized syringe will be accepted for testing, however there is documentation that heparin can interfere with some PCR assays and is not preferred.
Draw Volume:	6 mL (Minimum: 1 mL) blood 1-2 mL (Minimum: 0.5 mL) bone marrow aspirate
Processed Volume:	Same as Draw Volume
Collection:	Routine venipuncture or bone marrow collection. Clearly label specimen as blood or bone marrow. Invert specimen gently to mix.
Special Processing:	Lab Staff: **Do not** centrifuge specimen. Submit blood specimen in original collection container. Bone marrow aspirate should be transferred and shipped in an EDTA vacutainer tube. Store and ship specimen at ambient temperature. Specimen must be received within 48 hours of collection. If sample is expected to arrive at the testing lab beyond 48 hours, store and transport refrigerated at 2 – 6˚C. The stability limit is seven days. Mark for Overnight delivery. If sample must be drawn for weekend delivery, please contact the lab ahead of time, and include tracking number, to ensure receipt and processing of sample. Forward promptly. Lab to send with UWash Fred Hutch/Seattle Cancer Care Alliance Hematopathology Requisition Requisition form
Patient Preparation:	None
Sample Rejection:	Mislabeled or unlabeled specimens; specimens collected with incorrect anticoagulant; specimens with unacceptable volume or inadequate white blood cell count; specimens received in testing lab past the stability limit.

Interpretive

Reference Range:	Not detected
Critical Values:	N/A

Limitations:	The FLT3 Assay does not detect an internal duplication (ITD) or a point mutation at D835. A negative result does not rule out the presence of a small population of cells containing either FLT3 mutation [FLT3(+)] that fall below the sensitivity of the assay (5% allele fraction). The clinical significance of small populations of FLT3(+) cells is unknown. FDA required disclaimer: This test was developed and it performance characteristics determined by the SCCA Molecular Oncology Laboratory. It has not been cleared or approved by the U.S. Food and Drug Administration. The FDA has determined, however, that in most cases, such approval is not necessary. This test is used for clinical purposes. It should not be regarded as investigational or for research.
Methodology:	PCR (Polymerase Chain Reaction)
References:	Molecular Hematopathology @ University of Washington https://testguide.labmed.uw.edu/ (February 2023)
Updates:	1/24/2011: Testing moved from Fred Hutchinson to Seattle Cancer Care Alliance. 2/5/2013: CPT update 2/20/2023: Billing, reference range and specimen collection updates. Lab address update to reflect Seattle Cancer Care and Fred Hutch alliance.