

Sample Case 3: AML MRD confirmed by Molecular Genetics

“Difference from Normal” Multidimensional Cytometry Results

Specimen Type: Bone Marrow Aspirate

Clinical History/Indications: A xy-year-old patient with a clinical history of FLT3-positive AML. A previous bone marrow specimen showed no evidence of abnormal myeloid progenitor cells.

Analysis/Conclusions: The flow cytometric findings reveal a **suspicious myeloblast population at 0.03% of non-erythroid cells, see comment**. Histopathologic, cytogenetic, and clinical data are required for complete interpretation.

Comment: The suspicious cell population lacks expression of CD13, a characteristic not identified in the previous specimen. Without a diagnostic phenotype to compare, it is difficult to interpret the significance of this small population. **Cell sorting and molecular studies suggested.**

Flow Cytometric SSC/CD45 Differential: 29% lymphocytes, 3.7% monocytes, 61% myeloid forms (of all stages of development), 0% lymphoblasts, and 0.1% myeloblasts (CD117) including 0.03% suspicious myeloid progenitor cells.

Immunophenotypic Findings: Independent immunophenotypic analysis of the myeloblast population reveals abnormal surface antigen expression consisting of HLA-DR, CD34, CD33, CD123, and CD117, without expression of CD11b, CD13, CD14, CD16, CD64 and all lymphoid antigens tested. Analysis of the maturing myeloid populations shows normal antigenic relationships. Total non-erythroid cells expressing CD34 are present at 0.2%.

Molecular Genetic Confirmation on Flow Cytometric Sorted Cells

Specimen Type: **Flow cytometry sorted CD34+CD13+ sorted cell fraction** from Bone Marrow Aspirate (36345312). Inadequate DNA yield for NGS analysis.

Clinical History/Indications: A xy-year-old patient with a clinical history of FLT3-positive AML. Current flow cytometric findings are suspicious for the presence of an aberrant myeloblast population at 0.03%.

FMS-like tyrosine kinase 3 mutation detection: **POSITIVE (ITD)**

Analysis/Conclusions:

- These findings are *positive* for the presence of an internal tandem duplication (ITD) (~42 bp) of the juxtamembrane domain of the Fms-Like Tyrosine kinase 3 receptor in the **CD34-positive, CD13-positive cell fraction of this specimen.**
- Clinical and histological correlation required for definitive diagnosis.

Activating mutations of the fms-like tyrosine kinase 3 receptor have been described to be an important prognostic factor in AML [Thiede et al. Blood 2002, 99:4326-4335] [Kottaridis et al. Blood 2001, 98:1752-1759].