Cytek® 20-Color AML Panel

Improve Operational Efficiency in Acute Myeloid Leukemia Therapy Development

Acute myeloid leukemia (AML) is a myeloid-lineage malignant neoplasm disorder. According to the American Cancer Society, AML is one of the most common types of leukemia in adults and it makes up 31% of all adult leukemia cases. The typical treatment for patients with AML is intensive chemotherapy. However, personalized and target-specific therapies are being developed as alternatives to broad cytotoxic chemotherapy for the treatment of AML. The development of new therapy methods has extensively improved disease prognosis and patient survival.

Multiparametric flow cytometry assays are widely used in detecting and monitoring AML in drug discovery, translational research, and clinical trials. On conventional flow cytometers, markers used for assessing AML are typically split into multiple tubes due to the limitation of detection channels, forcing the use of redundant markers and greater sample volume.

Cytek has developed a single-tube, 20-color panel for AML analysis using the Cytek Northern Lights™ and Aurora systems equipped with violet, blue, and red lasers. This panel consists of the Cytek 20-Color AML Panel, cFluor® Reagent Kit (19C) (P/N R7-40009), and CD19 Monoclonal Antibody (clone SJ25C1), Super Bright™ 780, eBioscience™ (P/N 78-0198-42).

Cytek's 20-Color AML Panel is an effective and sensitive flow cytometric approach for identifying and characterizing normal and aberrant cells, immunophenotyping, and evaluating measurable residual disease in AML samples. The marker selection for this panel was based on two publications with 2-tube[3] and 3-tube[1] AML assays and input from experts in the field. This single-tube assay enhances laboratory operational efficiency by saving time and labor for sample preparation and acquisition, as well as preserving precious samples and eliminating the use of redundant reagents.

Violet			Blue			Red		
Specifcity	**Clone**	**Fluorochrome**	**Specifcity**	**Clone**	**Fluorochrome**	**Specifcity**	**Clone**	**Fluorochrome**
CD16	3G8	cFluor® V420	CD7	CD7-6B7	cFluor® B515	CD13	WM15	cFluor® R659
CD14	MEM18	cFluor® V450	CD15	HI98	cFluor® B548	CD5	L17F12	cFluor® R685
HLA-DR	L243	cFluor® V505	CD34	4H11	cFluor® BYG575	CD123	6H6	cFluor® R720
CD4	SK3	cFluor® V547	CD33	WM53	cFluor® BYG610	CD64	10.1	cFluor® R780
CD11b	ICRF44	cFluor® V610	CD71	OKT9	cFluor® BYG667	CD45	HI30	cFluor® R840
CD19	SJ25C1	Super Bright™ 780	CD38	HB-7	cFluor® B690			
			CD117	104D2	cFluor® BYG710			
			CD56	LT56	cFluor® BYG750			
			CD10	HI10a	cFluor® BYG781			

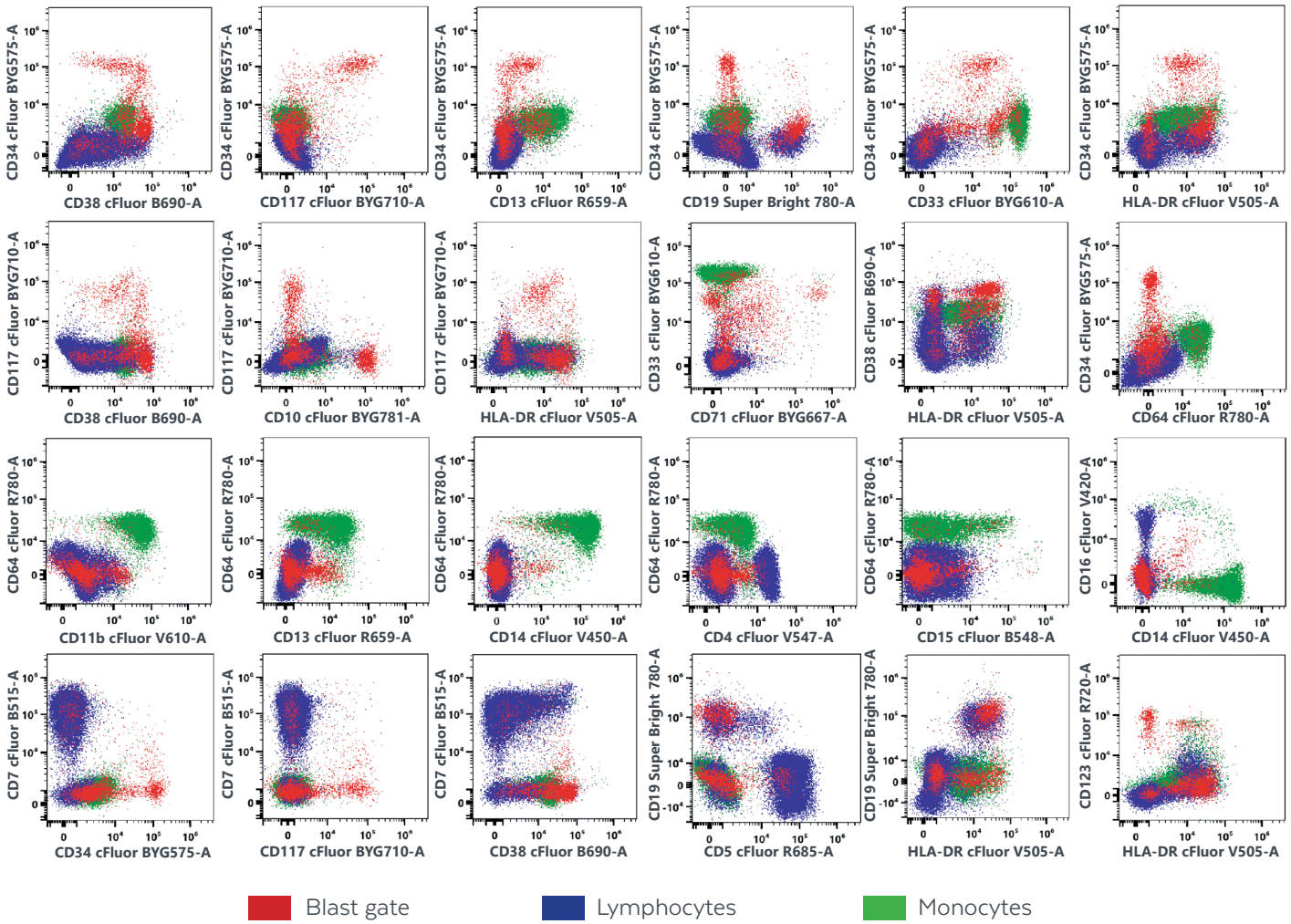
***Table 1**: Cytek 20-Color AML Panel, cFluor® Reagent Kit (19C) (P/N R7-40009), CD19 Monoclonal Antibody (clone SJ25C1), Super Bright™ 780, eBioscience™ (P/N 78-0198-42), and ViaDye™ Red (P/N R7-60008) are available to purchase separately.*

The Cytek 20-Color AML Panel is pre-optimized, eliminating the need for costly and labor-intensive panel design and optimization. Cytek scientists have evaluated the panel's performance in fresh and frozen bone marrow as well as peripheral blood samples with both stain/lyse and lyse/stain methods. Cytek also provides the following tools to streamline your laboratory workflow:

- Complete protocols for sample preparation, staining, and acquisition
- Experiment and analysis templates to streamline workflow and improve laboratory efficiency

Data

A. Mononuclear cells



B. Myeloid cells

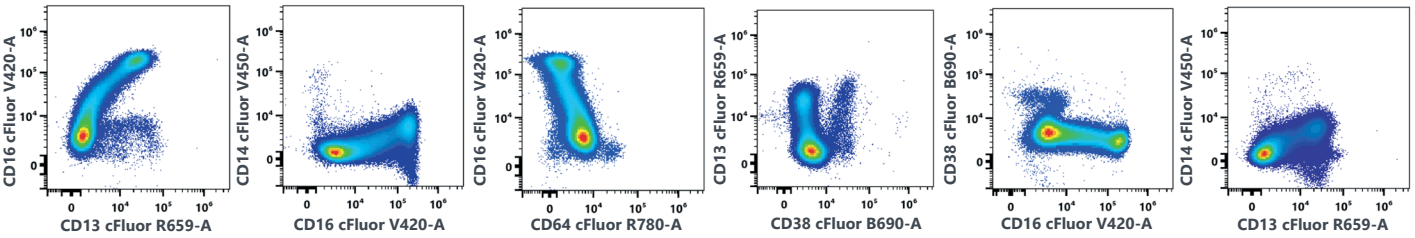


Figure 1. Representative data of healthy donor bone marrow samples. A fresh bone marrow sample collected in a heparin tube was stained with the Cytek 20-Color AML panel and analyzed on a 5 laser Aurora system. Data was analyzed using the gating schema and plot layout published by Wood et al[1] and Soh et al[2]. Data shown was sequentially gated with time gate, singlet gate and then followed with lymphocyte, monocyte, myeloid and blast gates on CD45 vs. SSC plot. Cells from lymphocyte, monocyte and blast gates were combined as mononuclear cells.

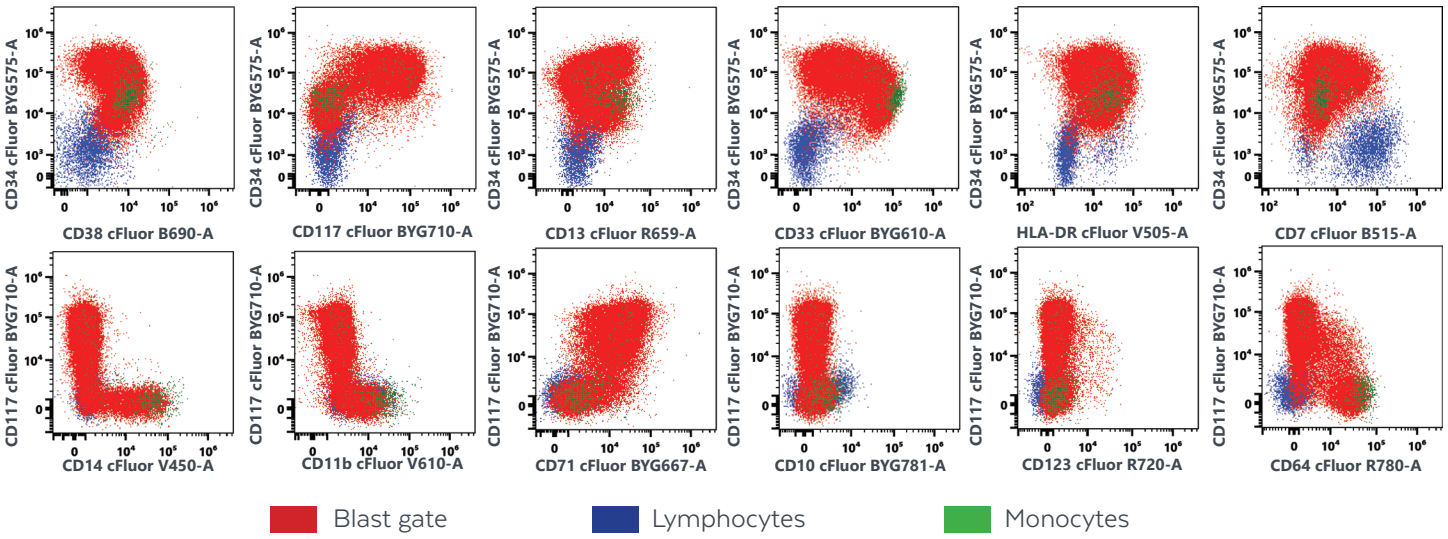


Figure 2. Representative data of AML frozen bone marrow samples stained with the Cytek 20-Color AML Panel. Data was generated using a cryo-preserved AML bone marrow sample. The layout of the data plots is based on those published by Wood et al[1] and Soh et al[2].

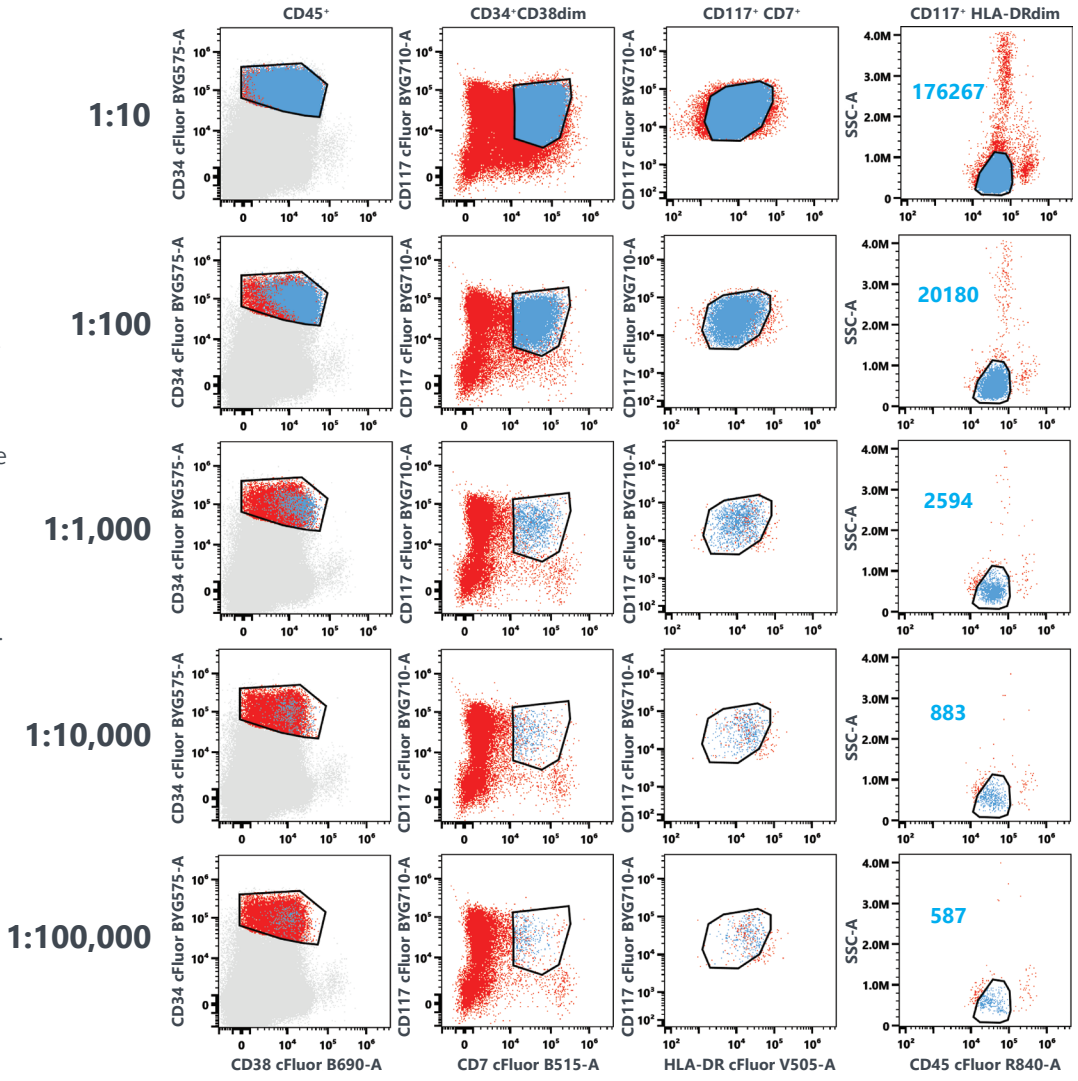


Figure 3. Empirical lower limit of quantification (LLOQ) determination of the 20-color AML panel for measurable residual disease (MRD) evaluation. A frozen AML bone marrow sample was spiked into a fresh normal bone marrow sample at different dilutions as shown in the graph and tested in triplicates. Representative data from one replicate is shown. The cancer cells are still detectable at 10^{-5} dilution.

References

1. Wood, B. L. (2020). Acute myeloid leukemia minimal residual disease detection: The difference from normal approach. Current Protocols in Cytometry, 93, e73. doi: 10.1002/cpcy.73

2. Soh, K. T., et al. (2022). Development of a 27-color panel for the detection of measurable residual disease in patients diagnosed with acute myeloid leukemia. Cytometry A, 101, 970-983.

3. Hedley, B. D., et al. (2021). A multicenter study evaluation of the ClearLLab 10C panels. Cytometry B Clin Cytom. 100, 225-234.

For Research Use Only. Not for use in diagnostic procedures.

[1]cFluor® V547, cFluor® B515, cFluor® B548, cFluor® BYG610, cFluor® R685, cFluor® R720 and cFluor® R840 are equivalent to CF®405L, CF®488A, CF®514, PE-CF®596R, CF®660C, CF®700 and APC-CF®790T respectively, manufactured and provided by Biotium, Inc. under an Agreement between Biotium and Cytek (LICENSEE). The manufacture, use, sale, offer for sale, or import of the product is covered by one or more of the patents or pending applications owned or licensed by Biotium. The purchase of this product includes a limited, non-transferable immunity from suit under the foregoing patent claims for using only this amount of product for the purchaser's own internal research. No right under any other patent claim, no right to perform any patented method, and no right to perform commercial services of any kind, including without limitation reporting the results of purchaser's activities for a fee or other commercial consideration, is conveyed expressly, by implication, or by estoppel.
[2]cFluor® BYG610, cFluor® BYG667, cFluor® BYG710, cFluor® BYG750, and cFluor® BYG781 are tandem dyes made with R-PE. cFluor® B690 is a tandem dye made with PerCP. cFluor® R780 and cFluor® R840 are tandem dyes made with APC. Caution – Tandem dyes may show changes in their emission spectra with prolonged exposure to light or fixatives.

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N9-20102, Rev. A
January 2023