TBMNK Cell

Multi-Color Flow Cytometry Panel

Catalog Number: FMC-P-002

PRODUCT DESCRIPTION

T cells, B cells, and NK cells are the major lymphocyte subsets in peripheral blood. Monocytes are critical mediators of host immune responses. This panel contains 7 conjugated antibodies that can be used for the single step staining of T cells, B cells, NK cells, classical monocytes, and non-classical monocytes.

INTENDED USE

This multicolor flow cytometry panel was validated on human peripheral blood mononuclear cells (PBMCs).

MATERIALS PROVIDED & STORAGE

Store the unopened kit at 2-8 °C. Do not use past expiration date.

MARKER	CLONE	FLUOROCHROME	SIZE	RECOMMENDED CONCENTRATION
CD3	UCHT1	Alexa Fluor® 405	100 tests	5 µL/10^6 cells
CD4	11830	FITC	100 tests	10 µL/10^6 cells
CD8	37006	APC	100 tests	10 µL/10^6 cells
CD14	134620	Alexa Fluor® 750	100 tests	5 µL/10^6 cells
CD16	245536	PerCP	100 tests	10 µL/10^6 cells
CD19	4G7-2E3	PE	100 tests	10 µL/10^6 cells
CD56	2524C	Alexa Fluor® 700	100 µg	0.25 - 1 µg/10^6 cells

Note: Recommended concentrations are given as reference point for antibody titration. Optimal concentrations should be determined by each laboratory for their experimental conditions.

PRECAUTIONS

Wear protective gloves, clothing, eye, and face protection. Wash hands thoroughly after handling. Refer to the SDS on our website prior to use.

OTHER SUPPLIES REQUIRED

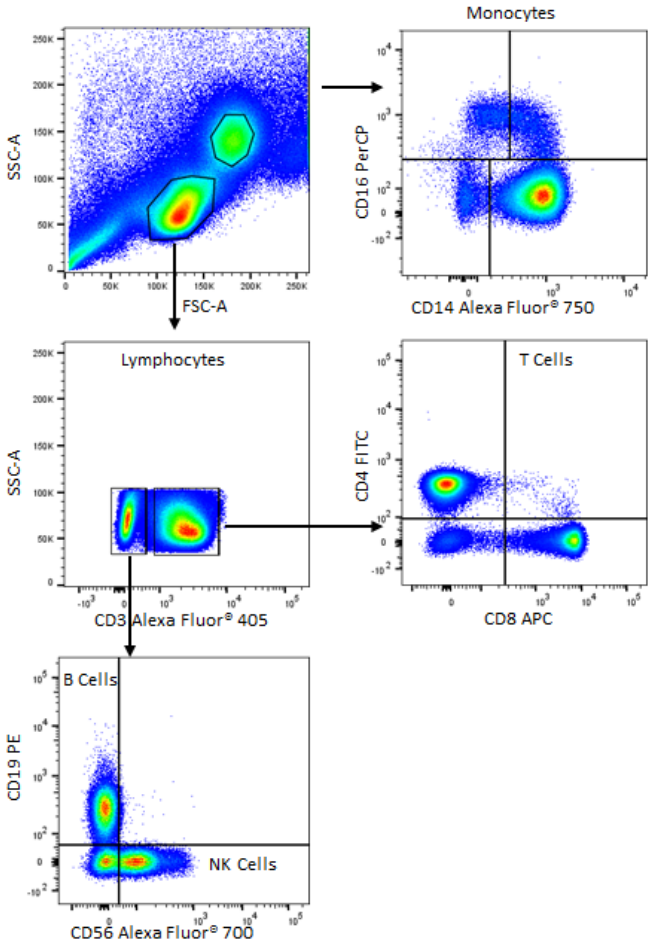
- PBS
- Flow Cytometry Staining Buffer (R&D Systems®, Catalog # FC001)
- Fc-block (blocking IgG)
- (Optional; Isotype Control Antibodies)
- 5 mL Flow cytometry tubes

PROTOCOL

1. Wash human PBMCs (1 x 10^6 cells per sample) with 2 mL of Staining Buffer (1X) (R&D Systems® Catalog # FC001) or other BSA-containing buffer, by spinning at 300 x g for 5 minutes, using 5 mL flow cytometry tubes. Decant/aspirate supernatant.
2. Fc-block cells with blocking IgG (1 µg IgG/10^6 cells) for 10 minutes at room temperature.
3. Add or previously titrated amount of each primary conjugated antibody. Vortex tubes.
4. *(optional)* To a separate tube, add 5 µL of each of the isotype control antibodies. Vortex tubes.
5. Incubate the mixtures for 30-45 minutes at room temperature **in the dark**.
6. Wash with 2 mL of Staining Buffer (1X), by spinning at 300 x g for 5 minutes at end of incubation. Decant/aspirate supernatant.
7. Resuspend the cells in 0.2 - 0.5 mL Staining Buffer (1X) and acquire on a Flow Cytometer.

Global bio-techne.com info@bio-techne.com TEL +1 612 379 2956 **USA** TEL 800 343 7475 **Canada** TEL 855 668 8722
Europe | Middle East | Africa TEL +44 (0)1235 529449 **China** TEL +86 (21) 52380373

DATA EXAMPLES



Multicolor Flow Cytometry Panel to identify human T Cells, B Cells, NK cells, and Monocyte subsets. PBMCs were stained with anti-human CD3 Alexa Fluor® 405, CD4 FITC, CD8 APC, CD56 Alexa Fluor 700, CD19 PE, CD14 Alexa Fluor 750, and CD16 PerCP. All antibodies are validated for flow cytometry. Classical monocytes are defined as $CD14^{+}$, non-classical monocytes are $CD16^{+}$; CD4 T cells are defined as $CD3^{+}$, $CD4^{+}$; CD8 T cells are $CD3^{+}$, $CD8^{+}$; B cells are $CD3^{-}$, $CD19^{+}$; and NK cells are $CD3^{-}$, $CD56^{+}$.