CellBlox™ Monocyte and Macrophage Blocking Buffer

Catalog Numbers B001T02F01, B001T03F01, B001T06F01

Pub. No. MAN0025651 Rev. A.0

WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from thermofisher.com/support.

Product details

Size	25, 100, or 500 tests[1]
Type	Buffer
Form	Liquid
Purification	HPLC
Storage buffer	PBS, pH 7.2
Storage conditions	4°C. Do not freeze!
Applications reported	Flow cytometry
Tested dilution	5 µL/test

[1] A test is defined as the amount (µg) of antibody that will stain a cell sample in a final volume of 100 µL. Cell number can range from 10^5 to 10^8 cells/test.

Product description

CellBlox™ Monocyte and Macrophage Blocking Buffer has been specifically formulated to block the non-specific binding of NovaFluor™ dyes and most tandem dyes with macrophages and monocytes. It also assists in reducing background staining when using NovaFluor™ dyes.

Application

CellBlox™ Monocyte and Macrophage Blocking Buffer should be included whenever NovaFluor™ dyes or other tandem dyes are used in order to block non-specific binding of dyes to monocytes or macrophages and to reduce background staining. CellBlox™ blocking buffer works best when included in a master mix as it increases the dilution and reduces contact time with cells. However, it can also be added directly to a cell suspension if required.

Add 5 µL of CellBlox™ blocking buffer per test to the antibody master mix, prior to staining cells, then incubate on ice for 5 minutes. A test is defined as the amount (µg) of antibody that will stain a cell sample in a final volume of 100 µL. Cell number can range from 10^5 to 10^8 cells/test.

If adding directly to a suspension, add 5 µL of CellBlox™ blocking buffer to a suspension of cells prior to the addition of antibodies.

Compatibility

CellBlox™ Monocyte and Macrophage Blocking Buffer is compatible with other blocking reagents such as Fc Block, irrelevant proteins, rat/mouse serum, BD Horizon™ Brilliant Stain Buffer, and Invitrogen™ Super Bright Complete Staining Buffer. It does not impact compensation beads.

NovaFluor™ dyes are not compatible with DNA-binding dyes such as propidium iodide, 7-actinomycin D (7-AAD), and DAPI. Invitrogen™ LIVE/DEAD™ Fixable Dead Cell stains are recommended for use with NovaFluor™ dyes.

Applications tested

CellBlox™ Monocyte and Macrophage Blocking Buffer has been pre-titrated and tested by flow cytometric analysis of normal human peripheral blood cells. See "Experimental results."

Experimental results

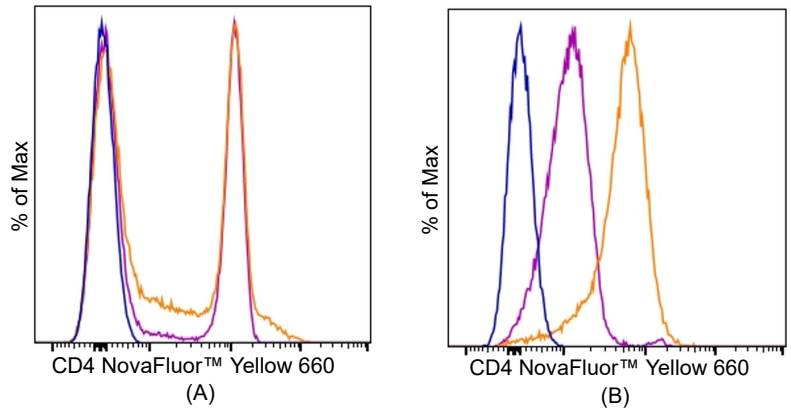

Figure 1 CellBlox™ Monocyte and Macrophage Blocking Buffer mitigates non-specific interactions of NovaFluor™ dyes

Peripheral Blood Mononuclear Cells (PBMC) were either unlabeled (blue) or labeled CD4 Monoclonal Antibody (SK3), NovaFluor™ Yellow 660, eBioscience™ direct conjugate with and without the addition of CellBlox™ blocking buffer. Histogram overlay plots of CD4 expression are shown using a lymphocyte gate (A) and a monocyte gate (B). CD4 labeling combined with CellBlox™ blocking buffer (purple) is shown to reduce non-specific labeling of monocytes and macrophages as compared with CD4 labeling without CellBlox™ blocking buffer (orange), leading to an improvement in signal accuracy. Data was acquired on a 4-laser Invitrogen™ Attune™ NxT Flow Cytometer using the 561 nm laser with a 695/40 nm bandpass filter.

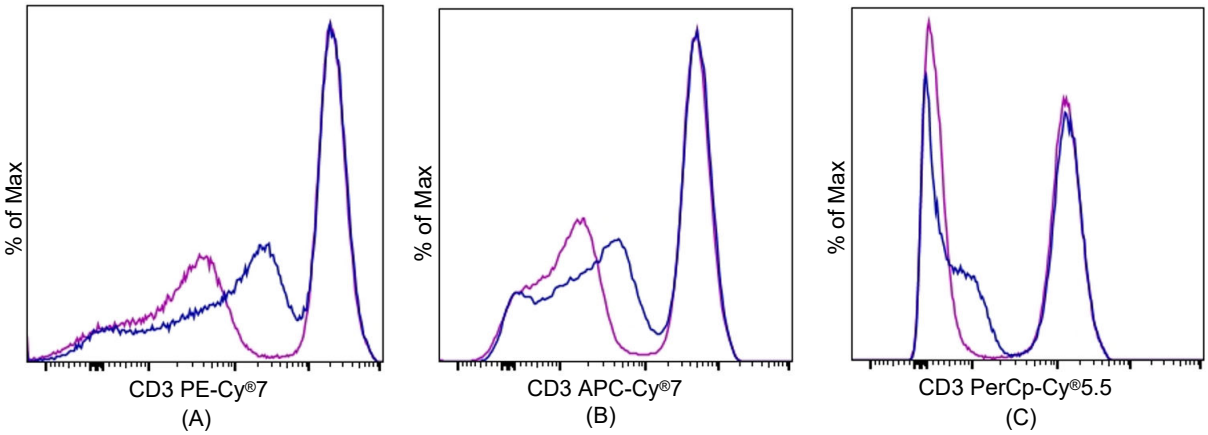

Figure 2 CellBlox™ Monocyte and Macrophage Blocking Buffer mitigates non-specific interactions of PE, APC, and PerCP Cyanine tandem dyes

Peripheral Blood Mononuclear Cells (PBMC) were labeled with CD3 direct conjugates of PE-Cy®7, APC-Cy®7, and PerCP-Cy®7 with and without CellBlox™ blocking buffer. Histogram overlay plots of CD3 expression are shown using a lymphocyte gate. Data is displayed with CellBlox™ blocking buffer (purple) or without blocking buffer (blue). CD3 labeling of tandem dyes combined with CellBlox™ blocking buffer (purple) is shown to reduce non-specific labelling of monocytes and macrophages as compared with CD3 labeling without CellBlox™ blocking buffer (blue), leading to a reduction in background noise and an improvement in signal strength and accuracy. Data was acquired on a 4-laser Invitrogen™ Attune™ NxT Flow Cytometer using the 488 nm laser with a 695/40 nm bandpass for PE-Cy®7 (A), the 561 nm laser with a 780/60 nm bandpass filter for APC-Cy®7 (B), and the 638 nm laser with a 780/60 nm bandpass filter for PerCP-Cy®5.5 (C).

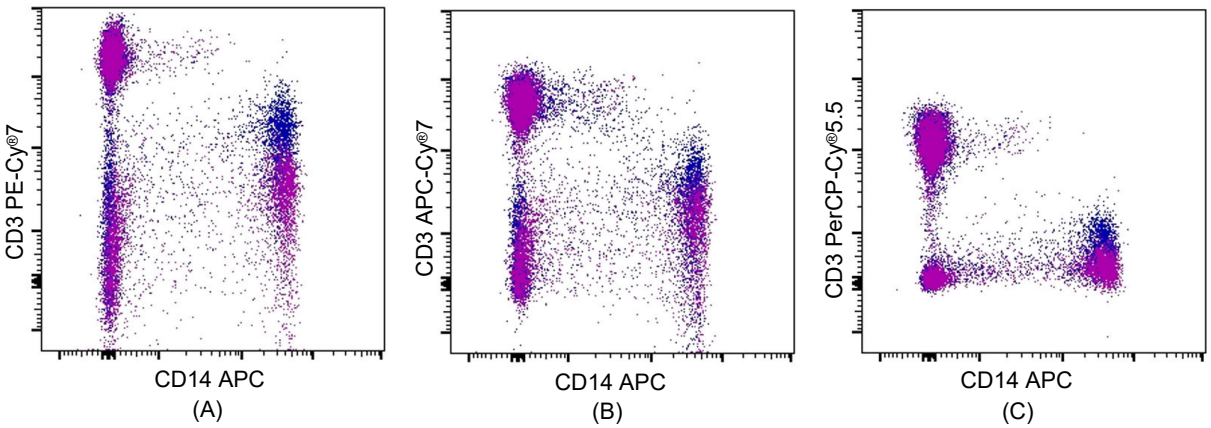

Figure 3 CellBlox™ Monocyte and Macrophage Blocking Buffer mitigates non-specific interactions of PE, APC, and PerCP Cyanine tandem dyes

Peripheral Blood Mononuclear Cells (PBMC) were labeled with CD3 direct conjugates of PE-Cy®7, APC-Cy®7, PerCP-Cy®7, and CD14 APC with and without CellBlox™ blocking buffer. Overlay plots of CD3 and CD14 expression are shown . Data is displayed with CellBlox™ blocking buffer (purple) or without blocking buffer (blue). CD3 labeling of tandem dyes combined with CellBlox™ blocking buffer (purple) is shown to reduce non-specific labelling of monocytes and macrophages as compared with CD3 labeling without CellBlox™ blocking buffer (blue), leading to a reduction in background noise and an improvement in signal strength and accuracy. Data was acquired on a 4-laser Invitrogen™ Attune™ NxT Flow Cytometer using the 488 nm laser with a 695/40 nm bandpass for PE-Cy®7 (A), the 561 nm laser with a 780/60 nm bandpass filter for APC-PE-Cy®7 (B), and the 638 nm laser with a 780/60 nm bandpass filter for PerCP-Cy®5.5 (C).

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Revision	Date	Description
A.0	20 September 2021	New product manual (part of Phitonex™ integration)

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