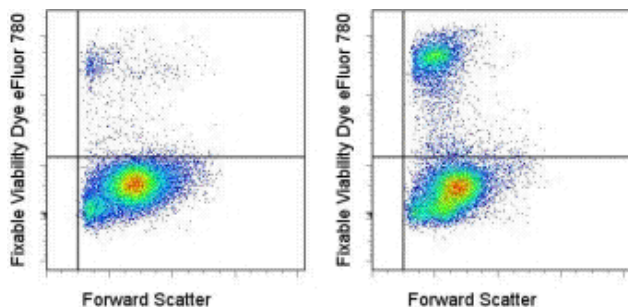


Fixable Viability Dye eFluor® 780

Catalog Number: 65-0865

GPR: General Purpose Reagents. For Laboratory Use.



BALB/c thymocytes were uncultured (left) or cultured overnight at 37°C (right) and then stained with Fixable Viability Dye eFluor® 780. Total cells were used for analysis.

Product Information

Contents: Fixable Viability Dye eFluor® 780

REF Catalog Number: 65-0865

Formulation: Dye in DMSO, pre-diluted to test size



Temperature Limitation: Store at less than or equal to -70°C. Protect from light and moisture.



LOT Batch Code: Refer to Vial



Use By: Refer to Vial

Description

Fixable Viability Dye eFluor® 780 is a viability dye that can be used to irreversibly label dead cells prior to cryopreservation, fixation and/or permeabilization procedures. Unlike 7-AAD and propidium iodide, cells labeled with Fixable Viability Dyes can be washed, fixed, permeabilized, and stained for intracellular antigens without any loss of staining intensity of the dead cells. Thus, using Fixable Viability Dyes allows dead cells to be excluded from analysis when intracellular targets are being studied. Fixable Viability Dyes may be used to label cells from all species.

Fixable Viability Dye eFluor® 780 can be excited by the red (633 nm) laser line and has a peak emission of 780 nm that can be detected using a 780/60 band pass filter (equivalent to APC-eFluor® 780 or APC-Alexa Fluor® 750). Please make sure that your instrument is capable of detecting this dye. For compensation, it is recommended to use a sample of the cells of interest stained with the Fixable Viability Dye. If the percentage of dead cells is expected to be less than 5%, then it is recommended to take a small aliquot of cells and heat them at 65°C for 1 minute then immediately place on ice for 1 minute. After this treatment, the heat-killed cells can be combined 1:1 with live cells and then stained with the Fixable Viability Dye. Testing at eBioscience suggests that compensation out of most detectors is negligible, with compensation out of PE-Cy7 being the highest at <5%. Actual compensation values will depend on each investigator's specific instrument, filter sets, and PMT voltage settings.

Fixable Viability Dye eFluor® 780 is supplied as a pre-diluted solution prepared in high-quality, anhydrous DMSO. It should be protected from light and moisture. Store at -80°C with dessicant. It may be freeze-thawed up to 20 times. Allow vial to equilibrate to room temperature before opening.

Applications Reported

Fixable Viability Dye eFluor® 780 has been reported for use in flow cytometric analysis.

Applications Tested

Fixable Viability Dye eFluor® 780 has been tested by flow cytometric analysis of mouse thymocytes. Fixable Viability Dyes are fully compatible with both IC Fixation and Permeabilization Buffers and the Foxp3 Buffer Set. This can be used at 1 µL/mL of cells resuspended at 1-10x10⁶ cells per mL in azide-free and serum/protein-free PBS. It is recommended that the concentration used be determined by each investigator for optimal performance in the assay of interest.

Special Notes

Staining with Fixable Viability Dye eFluor® 780 may be done before or after surface staining. Cells may be cryopreserved after staining with Fixable Viability Dye eFluor® 780 with no adverse effect on staining intensity of dead cells after thawing.

References

Gray EE, Suzuki K, Cyster JG. Cutting edge: Identification of a motile IL-17-producing gammadelta T cell population in the dermis. J Immunol. 2011 Jun 1;186(11):6091-5.

Related Products

00-5523 Foxp3 / Transcription Factor Staining Buffer Set

65-0863 Fixable Viability Dye eFluor® 450
65-0864 Fixable Viability Dye eFluor® 660
65-0866 Fixable Viability Dye eFluor® 506
88-8823 Fixation & Permeabilization Buffers

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Fixable Viability Dye Cell Staining Protocol

Research Use Only

Protocol: Fixable Viability Dye Cell Staining

Experimental Procedure

Allow vial of Fixable Viability Dye to equilibrate to room temperature before opening. Staining with Fixable Viability Dye must be done in azide-free and serum/protein-free PBS. For consistent staining of cells, do not stain cells in less than 0.5 mL.

1. Prepare cells as desired.
2. Wash cells 2 times in azide-free and serum/protein-free PBS.
3. Resuspend cells at $1-10 \times 10^6$ /mL in azide-free and serum/protein-free PBS.
4. Add 1 μ L of Fixable Viability Dye per 1 mL of cells and vortex immediately.
5. Incubate for 30 min at 2-8°C, protect from light.
6. Wash cells 1-2 times with flow staining buffer or equivalent.
7. Fix and/or permeabilize cells as desired.

Note: Cells may be stained with Fixable Viability Dyes before or after surface staining. After staining with Fixable Viability Dyes, cells may also be cryopreserved for analysis at a later time. It is recommended that each investigator determine the optimal concentration for the assay of interest.