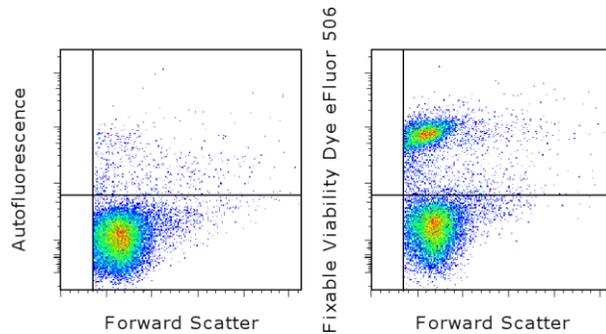


## Fixable Viability Dye eFluor<sup>®</sup> 506

Catalog Number: 65-0866

GPR: General Purpose Reagents. For Laboratory Use.



Staining of C57Bl/6 thymocytes cultured overnight with staining buffer (autofluorescence) (left) or Fixable Viability Dye eFluor<sup>®</sup> 506 (right). Total cells were used for analysis.

### Product Information

**Contents:** Fixable Viability Dye eFluor<sup>®</sup> 506  
**Catalog Number:** 65-0866



**Formulation:** DMSO, pre-diluted to test size

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



**Batch Code:** Refer to vial



**Use By:** Refer to vial

### Description

Fixable Viability Dye eFluor<sup>®</sup> 506 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<sup>®</sup> 506 can be excited by the violet (405 nm) laser line and has a peak emission of 506 nm that can be detected using a 510/50 band pass filter (equivalent to AmCyan). 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.

Fixable Viability Dye eFluor<sup>®</sup> 506 is supplied as a pre-diluted solution prepared in high-quality, anhydrous DMSO. It should be protected from light and moisture. Store at less than or equal to -70°C with desiccant. It may be freeze-thawed up to 20 times. Allow vial to equilibrate to room temperature before opening.

### Applications Reported

Fixable Viability Dye eFluor<sup>®</sup> 506 has been reported for use in flow cytometric analysis.

### Applications Tested

Fixable Viability Dye eFluor<sup>®</sup> 506 has been pre-titrated and 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 1-10x10<sup>6</sup> 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<sup>®</sup> 506 may be done before or after surface staining. Cells may be

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## Fixable Viability Dye eFluor® 506

**Catalog Number:** 65-0866

**GPR: General Purpose Reagents. For Laboratory Use.**

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cryopreserved after staining with Fixable Viability Dye eFluor® 506 with no adverse effect on staining intensity of dead cells after thawing.

### Related Products

00-5521 Foxp3 Fixation/Permeabilization Concentrate and Diluent

65-0863 Fixable Viability Dye eFluor® 450

65-0864 Fixable Viability Dye eFluor® 660

65-0865 Fixable Viability Dye eFluor® 780

88-8823 Fixation & Permeabilization Buffers

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## Fixable Viability Dye Cell Staining Protocol

Research Use Only

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### Protocol: Fixable Viability Dye Cell Staining

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#### 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  $\mu$ 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.*