Cell Meter™ IX830 fixable viability dye

Catalog number: 22529
Unit size: 200 Tests

Component	Storage	Amount (Cat No. 22529)
Cell Meter™ IX830 fixable viability dye	Freeze (< -15 °C), Minimize light exposure	200 tests

OVERVIEW

Discrimination and exclusion of dead cells from live cells allows cleaner separation and identification of cell populations. Cell Meter™ fixable viability dyes are a large family of cell-impermeable fluorescent viability dyes that are optimized to match the major excitation lasers of common flow cytometers, such as 350, 405, 488, 633 and 647 nm. These dyes are impermeant to live cells but permeant to cells with compromised membranes. They irreversibly react with amine- and thiol-containing proteins and other cellular components. Since dead or fixed cells with a compromised membrane more readily react with Cell Meter™ fixable cell stains, thus stain brighter than live cells with an intact membrane, these dyes can be used to assess live vs. dead status of mammalian cells. There are a few factors to be considered when using these dyes, e.g., the titration of each dye to ensure that live cells have minimal to no staining. Cell Meter™ IX830 fixable cell stain is optimized to be excited with the IR laser at 808 nm with emission at 830 nm. Compared to other commercially similar viability dyes, this fixable viability dye is much more robust and stable.

AT A GLANCE

Protocol summary

1. Prepare samples in HHBS buffer (0.5 mL/assay).
2. Add Cell Meter™ IX830 to the cell suspension.
3. Stain the cells at room temperature for 20 - 60 minutes.
4. Wash the cells.
5. Fix the cells (optional).
6. Examine the sample with flow cytometer using 780/50 nm filter (APC-Cy7 channel) and/or IR filters.

Important Note

Thaw at room temperature before starting the experiment.

KEY PARAMETERS

Flow cytometer

Emission	780/50 nm or IR filters
Excitation	634 laser or IR lasers
Instrument specification(s)	APC/Cy7 or IR filters

PREPARATION OF STOCK SOLUTIONS

Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles

Cell Meter™ IX830 stock solution (500X)

1. Add 200 µL of DMSO to the Cell Meter™ IX830 vial to make a 500X stock solution.

   Note: Any unused Cell Meter™ IX830 stock solution should be divided into single-use aliquots and stored at -20°C. Avoid repeated freeze/thaw cycles.

SAMPLE EXPERIMENTAL PROTOCOL

1. Treat cells as desired.
2. Wash cells once with HHBS or the azide- and serum/protein-free buffer of your choice.
3. Resuspend cells at 5 - 10 × 10^6/mL in HHBS or in the azide- and serum/protein-free buffer of your choice.
4. Add 1 µL of 500X Cell Meter™ IX830 stock solution to 0.5 mL of cells/assay and mix it well.
5. Incubate at room temperature or 37°, 5% CO_2 incubator for 20 - 60 minutes, protected from light.

   Note: The optimal stain concentrations and incubation time should be experimentally determined for different cell lines.
6. Wash cells twice and resuspend cells with HHBS or the buffer of your choice.
7. Fix cells as desired (optional).
8. Analyze cells with a flow cytometer using 780/50 nm filter (APC-Cy7 channel) or IR filters.

EXAMPLE DATA ANALYSIS AND FIGURES

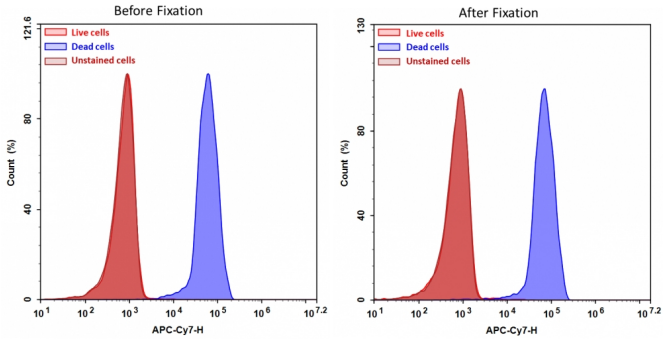

Figure 1. Detection of Jurkat cell viability by Cell Meter™ fixable viability dye. Jurkat cells were treated and stained with Cell Meter™ IX830 (Cat#22529), and then fixed in 3.7% formaldehyde and analyzed by flow cytometry. The dead cell population (Blue peak) is easily distinguished from the live cell population (Red peak) with APC-Cy7 channel, and nearly identical results were obtained before and after fixation.

DISCLAIMER

AAT Bioquest provides high-quality reagents and materials for research use only. For proper handling of potentially hazardous chemicals, please consult the Safety Data Sheet (SDS) provided for the product. Chemical analysis and/or reverse engineering of any kit or its components is strictly prohibited without written permission from AAT Bioquest. Please call 408-733-1055 or email info@aatbio.com if you have any questions.