PI Brite™

Catalog number: 17583, 17584
Unit size: 1 mg, 5 mg

Component	Storage	Amount (Cat No. 17583)	Amount (Cat No. 17584)
PI Brite™	Freeze (< -15 °C), Minimize light exposure	1 mg	5 mg

OVERVIEW

PI Brite™ is a propidium iodide (PI) analog developed by AAT Bioquest to improve the brightness and fixability of propidium iodide in cells. Under the same conditions, PI Brite™ is significantly brighter and better survives cell fixation process than PI. Propidium iodide is a fluorescent intercalating agent commonly used for staining cells with compromised membranes, such as dead cells, apoptotic or fixed cells. One of the primary applications of propidium iodide is for cell viability assays. Since it only stains cells with compromised cell membranes, it is often used to distinguish between live and dead cells in a population. In addition, PI is frequently used in flow cytometry to assess the cell cycle distribution of a population of cells. It allows researchers to analyze the DNA content of individual cells and identify different phases of the cell cycle. It binds to DNA by intercalating between the bases. PI is often used for detecting and quantifying DNA in various applications, such as flow cytometry, fluorescence microscopy, and gel electrophoresis.

AT A GLANCE

Spectral Properties

Ex/Em = 536/620 nm (bound to DNA)

KEY PARAMETERS

Fluorescence microscope

Emission	Texas Red filter set
Excitation	Texas Red filter set
Recommended plate	Black wall/clear bottom

SAMPLE EXPERIMENTAL PROTOCOL

Important Note

PI Brite™ staining is typically conducted as a final step after all other types of staining. This staining method is used to identify only dead cells. The procedure can be adjusted to work with most cell types, but factors such as growth medium, cell density, the presence of other cell types, and other variables may affect the staining process. Additionally, any residual detergent on glassware may impact the actual or perceived staining of many organisms. This can cause brightly stained material to appear in both solutions with and without cells present.

1. Make a 1 to 10 mM PI Brite™ stock solution in high-quality DMSO.

   Note: Any unused PI Brite™ stock solution should be divided into single-use aliquots and stored at -20 °C, protected from light. Avoid repeated freeze-thaw cycles.

2. Use the fixation protocol appropriate for your sample.

3. Pellet cells by centrifugation and resuspend the cells in buffered salt solutions or media, with optimal dye staining at pH 7.4. Adherent cells in culture may be stained *in situ* on coverslips or in the cell culture wells.

4. Add PI Brite™ using the concentrations between 0.5 and 5 µM and incubate it for 15 to 60 minutes as a guide.

Note: In initial experiments, it may be best to try several dye concentrations over the entire suggested range to determine the concentration that yields optimal staining.

EXAMPLE DATA ANALYSIS AND FIGURES

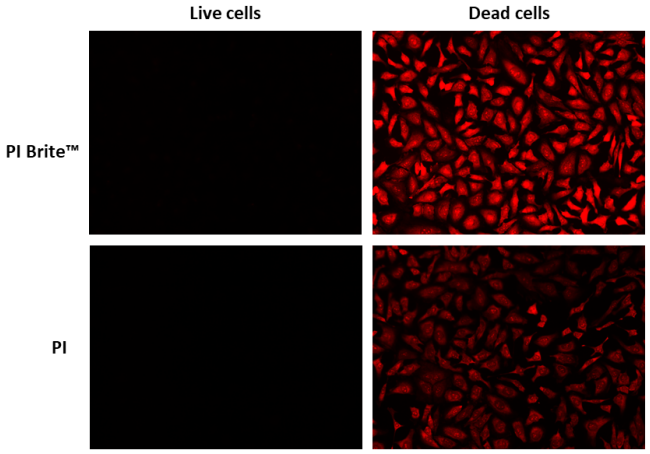

Figure 1. Image of live and dead HeLa cells stained with propidium iodide (PI) and PI Brite™ at similar concentrations and analyzed by fluorescence microscopy using a Cy3/TRITC filter set.

DISCLAIMER

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