

# Dyes and Stains

## Propidium Iodide

Cell viability dye (DNA-labeling dye)



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Catalog # 75002

10 mg

## Product Description

Propidium Iodide (PI) is a red-fluorescent cell viability dye which is excluded from live cells with intact membranes, but penetrates dead or damaged cells to bind to DNA and RNA by intercalating between the bases. It is widely used as a counterstain to differentiate and exclude non-viable cells in flow cytometric analyses, and can be excited using blue (488 nm), green (532 nm), or yellow-green (561 nm) laser lines, with detection in the FL2 or FL3 channels. PI is used in DNA fluorescence imaging applications to discriminate early and late stages of apoptosis, to study cell-mediated cytotoxicity, and for chromosome analysis. It is also commonly used in quantitative DNA assays.

Chemical Name:	3,8-diamino-5-[3-(diethylmethylammonio)propyl]-6-phenylphenanthridinium diiodide
Alternative Names:	3,8-Diamino-5-{3-[diethyl(methyl)ammonio]propyl}-6-phenylphenanthridinium diiodide; PI; Propidium diiodide
CAS Number:	25535-16-4
Chemical Formula:	$C_{27}H_{34}N_4 \cdot 2I$
Molecular Weight:	668.4 g/mol
Excitation Wavelength:	488 - 535 nm (DNA or RNA complex)
Emission Wavelength:	617 nm (DNA or RNA complex)

## Properties

Storage:	Store at -20°C.
Shelf Life:	Product stable until expiry date (EXP) on label. Protect product from prolonged exposure to light.
Format/Formulation:	A crystalline solid

## Applications

Verified:	FC
Reported:	FC, FISH, Fluorescence microscopy, Fluorometry, ICC
Special Applications:	This product has been verified for viability assessments of cells isolated with EasySep™ and RosetteSep™ kits.

Abbreviations: CellSep: Cell separation; ChIP: Chromatin immunoprecipitation; FA: Functional assay; FC: Flow cytometry; ICC: Immunocytochemistry; IF: Immunofluorescence microscopy; IHC: Immunohistochemistry; IP: Immunoprecipitation; RIA: Radioimmunoassay; WB: Western blotting

## Handling/Directions for Use

### PREPARATION

A stock solution may be made by dissolving PI in the diluent of choice. PI is soluble in aqueous buffers and in organic solvents.

Guidelines for the solubility of PI are as follows:

- Phosphate-buffered saline (PBS), pH 7.2  $\leq$  2 mg/mL
- Ethanol  $\leq$  0.2 mg/mL
- DMSO  $\leq$  2.5 mg/mL
- Dimethyl formamide  $\leq$  3.3 mg/mL

NOTE: If making a stock solution using an organic solvent, further dilution into aqueous buffers or isotonic saline should be made prior to performing biological experiments. Ensure that the residual amount of organic solvent is insignificant, as organic solvents may have physiological effects at low concentrations.

Wherever possible, prepare and use the stock solutions on the same day. Protect stock solutions from prolonged exposure to light. If stock solutions must be made in advance, aliquot and store in tightly sealed vials at  $-20^{\circ}\text{C}$ , protected from prolonged exposure to light. Generally these will be stable for up to 1 month.

### FLOW CYTOMETRY

1. Prepare a 1 mg/mL (1.5 mM) stock solution by dissolving solid PI in PBS.
2. Add to cells at a final concentration of  $\leq 1 \mu\text{g/mL}$ .
3. Incubate for 5 - 10 min in the dark, then analyze immediately.

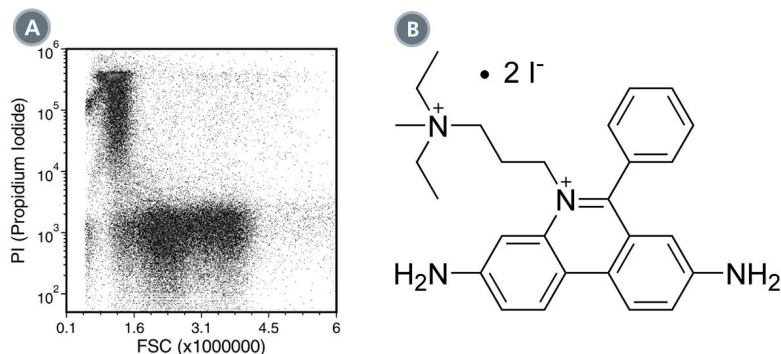
The dye should be titrated for optimal performance for each application.

## Notes and Tips

For flow cytometric analysis, PI can be detected in the FL2 (DNA content) or FL3 (viability) channels. Use FL2 to analyze PI staining if it is being used as a counterstain with fluorescein-conjugated Annexin V.

For microscopy analysis, PI can be viewed using a rhodamine (red) filter. Cells will be stained with PI if the membrane has been permeated, e.g. as a result of natural cell death or detergent treatment.

## Data/Structure



(A) Flow cytometry analysis of human peripheral blood mononuclear cells (PBMCs) labeled with Propidium Iodide.

(B) Chemical structure of Propidium Iodide.

## References

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