Hoechst 33342 Solution in Water

Cat. No. CTG-AP0011 Store the kit at -15 to -25°C

Experimental Protocol

1. Preparing Stock Solutions of Hoechst Dyes
The solid dyes may be dissolved in either water, dimethylformamide (DMF), or DMSO to make concentrated stock solutions up to 10 mg/mL.

2. Basic Protocol for Staining Cells
The following procedure can be adapted for most cell types. Note that different concentration ranges for the Hoechst dyes are suggested depending on the cell type (see below). Growth medium, cell density, the presence of other cell types and other factors may influence staining. Residual detergent on glassware may also affect real or apparent staining of many organisms, causing brightly stained material to appear in solutions with or without cells present. Glassware should be washed in a mild detergent and rinsed with hot tap water followed by several rinses with deionized, distilled water.

Pellet cells by centrifugation and resuspend in buffered salt solutions or media, with optimal dye binding at pH 7.4. Adherent cells in culture may be stained in situ on coverslips. Add Hoechst stain using the concentrations listed below as a guide. 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. Unbound dye has its maximum fluorescence emission in the 510–540 nm range, this green fluorescence may be observed on samples using too high a concentration of dye.

Recommended conditions for staining cells with Hoechst stains.

Cell Type	Hoechst Dye Concentration	Incubation Conditions
Bacteria	0.1 to 12 µg/mL	10 to 30 minutes
Live animal cells	0.2 to 5 µg/mL	20 to 30 minutes
Fixed animal cells	0.2 to 2 µg/mL	1 to 15 minutes

References

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