

## Hoechst 33342 Ready Stain for Flow Cytometry

<http://www.lumiprobe.com/p/hoechst-33342-ready-stain>

Hoechst 33342 (bisbenzimidazole, HOE 33342) is a cell-permeant blue-emitting fluorescent dye that binds strongly to adenine-thymine-rich regions in the minor groove of double-stranded DNA. Although Hoechst 33342 can bind to all nucleic acids, AT-rich dsDNA strands enhance its fluorescence considerably.

Hoechst 33342 bound with DNA has excitation/emission maxima at 351/461 nm, respectively. The fluorescence intensity of Hoechst 33342 increases with the pH of the solvent. The unbound dye fluoresces in the 510–540 nm range. The green fluorescence of unbound dye may be observed when an excessive dye concentration is used or the sample is insufficiently washed out.

Hoechst 33342 is used extensively in flow cytometry for staining chromosomes and nuclei in live and fixed cells. The dye is often used to distinguish condensed pycnotic nuclei in apoptotic cells and cell sorting. Hoechst 33342 is less toxic than DAPI, which ensures a higher viability of stained cells.

Hoechst 33342 is quenched by [bromodeoxyuridine \(BrdU\)](#), commonly used to detect dividing cells. When BrdU is integrated into DNA, the bromine is supposed to deform the minor groove so that Hoechst dyes cannot reach their optimal binding site. This property of Hoechst 33342 is used to study cell-cycle progression.

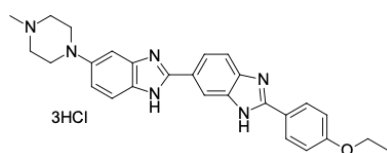
This product is a ready-to-use Hoechst solution for flow cytometry. We also offer Hoechst 33342 as a powder ([1H010](#)) and a concentrated 10 mg/mL solution ([2G010](#)).

## Protocol

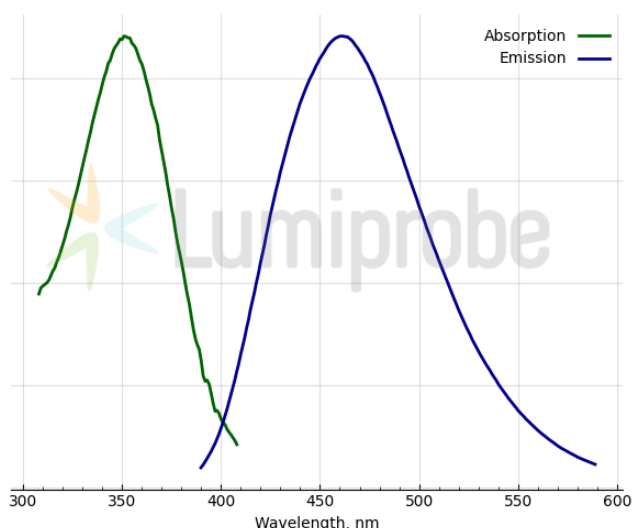
**Step 1:** Add 2 drops per  $10^6$  cells in 1 mL.

**Step 2:** Incubate for 60 minutes at 37 °C.

**Step 3:** Proceed with flow cytometry.



**Structure of Hoechst 33342**



**Absorption and emission spectra of Hoechst 33342 (DNA-dye complex)**

### General properties

Appearance: slightly yellow solution

Quality control: NMR 1H and HPLC-MS (95+%), functional testing

Storage conditions: 12 months after receipt at 2-8°C in the dark. Transportation: at room temperature for up to 3 weeks. Desiccate.

Legal statement: This Product is offered and sold for research purposes only. It has not been tested for safety and efficacy in food, drug, medical device, cosmetic, commercial or any other use. Supply does not express or imply authorization to use for any other purpose, including, without limitation, in vitro diagnostic purposes, in the manufacture of food or pharmaceutical products, in medical devices or in cosmetic products.

### **Spectral properties**

Excitation/absorption maximum, nm: 351 (complex)

Emission maximum, nm: 461 (complex)