Miltenyi Biotec

Hoechst 33342 Staining Solution

Order no. 130-111-569

Contents

1. Description

This product is for research use only.

Components	1 mL Hoechst 33342 Staining Solution:
Capacity	For 10^8 total cells, up to 100 tests.
Product format	The ready-to-use Hoechst 33342 Staining Solution is supplied in distilled water at a concentration of 1.5 mg/mL.
Storage	Store protected from light at 2–8 °C. Do not freeze. The expiration date is indicated on the vial label.

1.1 Background information

Hoechst 33342 is a cell permeable nucleic acid stain that binds to minor groove A-T rich regions of double-stranded DNA. Therefore, the staining intensity differs between different cell types. All nucleated cells are Hoechst positive.
The fluorescence emission maximum for DNA-bound Hoechst 33342 is about 461 nm. With an excitation maximum of 355 nm it can be excited by a ultraviolet (UV) laser as well as by a violet laser (405 nm) and detected within the blue fluorescence channel, e.g., channel V1 on the MACSQuant® Analyzer 10.

1.2 Applications

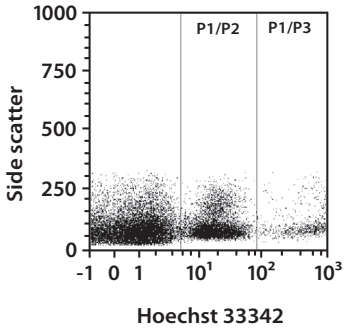
- Enumeration of nucleated cells.
- Nuclear counterstain in imaging and flow cytometry.
- Cell cycle analysis.

1.3 Recommended dilution

It is recommended to use Hoechst 33342 Staining Solution at a final concentration of 15 µg/mL. Since application vary, each investigator should titrate the reagent to obtain optimal results.
Incubation time can vary between 5–30 minutes at room temperature (max. 37 °C).
For enumeration of nucleated cells add 10 µL of Hoechst 33342 Staining Solution to 10^6 cells in 1 mL buffer and incubate for 15 minutes in the dark at room temperature before analysis.

2. Example of cell staining with the Hoechst 33342 Staining Solution

10^6 human peripheral blood mononuclear cells (PBMCs), 8 days old, were stained with the Hoechst 33342 Staining Solution, incubated for 15 minutes in the dark at room temperature, and analyzed by flow cytometry using the MACSQuant Analyzer 10. Cell debris was excluded from the analysis based on scatter signals. Viable nucleated cells are positive for Hoechst 33342 and belong to the P2 population. The bright P3 population consists of nucleated cells with damaged cell membranes, which are no longer viable.



Refer to **www.miltenyibiotec.com** for all data sheets and protocols. Miltenyi Biotec provides technical support worldwide. Visit www.miltenyibiotec.com/local to find your nearest Miltenyi Biotec contact.

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