

Hoechst 33342/PI Double Staining Kit

Cat#: orb219888 (User manual)

I. INTRODUCTION

Hoechst 33342 is a cell permeable fluorescent minor groove-binding probe for DNA, and through the living cell membrane. The Hoechst 33342 and DNA complex show light blue fluorescent color. Propidium iodide (PI) has been widely used as a fluorescent stain for DNA in cells; it specificity binds double-stranded nucleic acids in apoptosis, necrosis and fixed cells, but cannot enter the normal living cell. PI and DNA complex show light red fluorescent color. Hoechst 33342 and PI have been used for double fluorescence staining. This product is 1mg/ml chromogen. Solute it with suitable density for applies. The recommend density is 5 µg/ml.

II. KIT COMPONENTS

Component	Volume	Storage
Hoechst 33342 Chromogen (1mg/ml)	100 µl x 1	-20 °C
PI Chromogen (1mg/ml)	100 µl x 1	-20 °C
Dilution Buffer	20 ml x 2	4 °C
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III. STORAGE AND STABILITY

Store at 2-8 °C for short time; -20 °C for long time. Each component is stable for up to 12 months.

IV. PROCEDURE

1. For double or triple fluorescence staining in immunofluorescence tests, the Hoechst 33342/PI staining is the last step after all fluorescence antibodies incubation;

2. For culture cell, add 10 μ l Hoechst 33342/PI Chromogen equally to 2 ml Dilution Buffer in the same tube and mix them (the end density is 5 μ g/ml); incubate about 5 minutes in a dark incubator, at 30 °C; and then wash it with PBS/TBS 3 times for 3 minutes each time;

3. Observation with the fluorescence microscope.

Note:

- 1. The Hoechst 33342/PI are faded with light, all experiment process need keep away from light;
- 2. The Hoechst 33342/PI are suspected carcinogens, operation with gloves.