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MESGEN[™]
INNOVATION BIOTECHNOLOGY

Introduction

Distinguishing between live and dead cells is very important for investigation of growth control and cell death. The Live-Dead Cytotoxicity Kit provides the ready-to-use reagents for convenient discrimination between live and dead cells. The kit utilizes Calcein,AM, a cell-permeable green fluorescent dye (Ex/Em = 488/518 nm), to stain live cells. Dead cells can be easily stained by propidium iodide (PI), a cell non-permeable red fluorescent dye (Ex/Em = 535/615). Stained live and dead cells can be visualized by fluorescence microscopy using a band-pass filter.

Kit Components

	100T	500T	1000T
Solution A (1 mM Calcein,AM)	50µl	250µl	500µl
Solution B (1 mg/ml PI)	50µl	250µl	500µl
Staining Buffer	50ml	250ml	500ml

Cell Staining Protocol

1. Prepare enough Staining Solution for your assay (0.5 ml per well in 24 well dish) : mix 1µl of Solution A and 1µl of Solution B in 1 ml of Staining Buffer. Scale up accordingly for larger numbers of assays.
2. Collect cells (1×10^6 cells) by centrifugation at 500 X g for 5 min.
3. Resuspend to 0.5 ml Staining Solution
4. Incubate for 15 min at 37°C.
5. Place the cell suspension on a glass slide. Cover the cells with a glass coverslip. For analyzing adherent cells, grow cells directly on a coverslip. Following incubation with the Staining Solution, invert coverslip on a glass slide and visualize cells.
6. Observe cells immediately under a fluorescence microscope using a band-pass filter. Healthy cells stain only the cell-permeable Calcein,AM, fluorescing green. Dead cells can stain both the cell-permeable Calcein,AM and the cell non-permeable PI (red), the overlay of green and red appears to be yellow-red.

Caution

As the optimal staining conditions may vary among different cell types, we recommend that a suitable concentration of Solution A and B be determined individually. Please note that PI is suspected to be highly carcinogenic, so careful handling of the reagent is required.

Storage

Store at -20°C. Protect from light.

For Research Use Only. Not For Use In Diagnostic Procedures.