COS-0103

Live vs. Dead Cell Differentiation Staining Kit

Introduction:
Differentiating between live and dead cells is a common but very important task when investigating growth regulation controls and cell death mechanisms and pathways. AkrivisBio makes it simple with our ready-to-use reagents. We combine a proprietary cell-permeable green, fluorescent dye to stain live cells and propidium iodide, a red non-permeable dye, to stain all cells. Stained cells are visualized by fluorescence microscopy. The kit provides sufficient materials for 4 X 24-well plates.

Kit Components:

Live-Cell Dye	50 µl	Green	COS-0103-A
All-Cell Dye	50 µl	Red	COS-0103-B
Staining Buffer	50 ml	NM	COS-0103-C

Storage and Stability:
Store unopened kit at –20°C. Staining Buffer and All-Cell Dye (Propidium Iodide) can be stored at 4°C after opening. Live-Cell Dye should be stored at -20°C.

Staining Protocol:
1. Prepare sufficient Staining Solution for the number of wells to be stained. Each well will require 0.5 µl of Live-Cell Dye, 0.5 µl of All-cell Dye and 0.5 ml of Staining Buffer. Scale up accordingly for larger numbers of assays.
2. Collect cells (10^6) by gentle centrifugation at 500 X g for 5 min. Remove/discard supernatant.
3. Add 500 µl staining solution and gently resuspend the cell pellet.
4. Incubate for 15 min at 37°C.
5. Transfer the cell suspension to a glass slide and cover the cell suspension with a glass coverslip. Place on microscope stage to visualize.

NOTE: Grow adherent cells directly on a coverslip. To stain, tilt and remove growth medium, then lay flat, apply 0.5 ml of staining solution and incubate for 15 minutes. Invert coverslip onto a glass slide and place on stage.

6. Observe cells immediately with fluorescence microscopy using a band-pass filter which detects fluorescein and rhodamine, excluding shorter wavelengths.

Live cells stain with the cell-permeable Live Cell Dye, with a green fluorescence(Excitation 488 nm; emission 518 nm). Dead cell stain will stain both the live and dead cells red (excitation 488 nm; emission 615 nm), giving a combined yellow-red appearance.

Note: Staining conditions vary among different cell types, so it will be necessary to manually work through conditions to find suitable concentrations of the two dyes. PI is a suspected carcinogen, so use caution when handling the reagent.

FOR RESEARCH USE ONLY! Not to be used on humans.