

7-Aminoactinomycin D (7-AAD) Viability Staining Solution

Cat# C2069

Store at -20°C for 1 year and protected from light

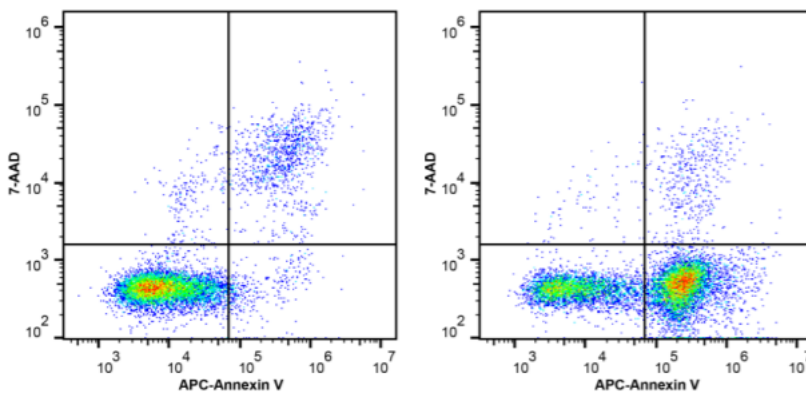
INTRODUCTION

Concentration: 100 µg/mL

7-Aminoactinomycin D 7-AAD Viability Staining Solution is developed to identify apoptotic and necrotic cells.

7-Aminoactinomycin D 7-AAD (7-amino-actinomycin D) has a high DNA binding constant and is efficiently excluded by intact cells. It is useful for DNA analysis and dead cell discrimination during flow cytometric analysis. Due to the loss of integrity of membrane, 7-AAD can enter late apoptotic or necrotic cells to stain DNA. Cells at different apoptotic stages can be distinguished by using 7-AAD and Annexin V.

Jurkat cells were treated with 1 µM Camptothecin and detected with this reagent and Annexin V-APC



Jurkat cells were cultured with (Right) or without (Left) 1 µM Camptothecin for 4 h. Annexin V-APC single-positive cells were early apoptotic cells, Annexin V-APC and 7-AAD double-positive cells were necrotic or late apoptotic cells, and PI single-positive cells were naked nuclei.

STAINING PROCEDURE

1. Induce apoptosis of suspension cells with reagents of interest. Collect cell cultures, centrifuge at 300 g for 5 min and discard the supernatant. Add PBS to wash the cells and resuspend the cells gently followed by the cell counting.
Tip: This product is only validated in suspension cells. Good cell viability is the key to the experiment. When the adherent cells are used for apoptotic detection, treatments like digestion may increase the ratio of necrotic or apoptotic cells and cause uncontrollable effects on the experimental results. Please be aware !
2. Split the cell suspension into tubes, $1 \sim 5 \times 10^5$ cells for each, centrifuge at 300 g for 5 min and discard the supernatant. Add PBS to wash the cells and discard the supernatant. Add 500 µL of 1 × Annexin V Binding Buffer to resuspend the cells.
3. Add 5 µl of Annexin V-APC and 5 µl of 7-AAD Viability Staining Solution to each tube.
4. Gently vortex the cells and incubate at room temperature for 15~20 min in the dark.
5. Analyze the cells immediately with proper machine settings. Otherwise, place the cells on ice in the dark and analyze within 1 h

CAUTIONS

1. For maximal assay performance, this reagent should be used within 12 months. Avoid freeze / thaw cycles.
2. Detect apoptosis as soon as possible after staining to avoid the increase number of apoptosis or necrosis.
3. Avoid extended exposure of the samples to direct light to protect the fluorophores from quenching.
4. For your safety and health, please wear the lab coat and disposable gloves before the experiments.

PRODUCT USE LIMITATION

These products are intended for research use only.