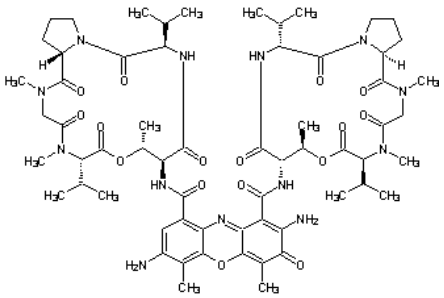
7-AAD [7-Aminoactinomycin D]

Ordering Information:	*Storage Conditions:*
Product Number: 17501 (1mg)	Keep at -20 °C and desiccated

Chemical and Physical Properties

Molecular Weight: 1270.43
Solvents: water and DMSO
Excitation = 546 nm
Emission = 647 nm



Biological Applications

7-Amino actinomycin D (7-AAD) is a non-permeant dye that can be used to identify non-viable cells. Cells with damaged plasma membranes or with impaired/no cell metabolism are unable to prevent the dye from entering the cell. Once inside the cell, the dyes bind to intracellular DNA producing highly fluorescent adducts which identify the cells as non-viable. It is widely used in flow cytometry. 7-AAD can be excited by the 488 nm laser line of an argon laser with fluorescence detected above 650 nm. Although the emission intensity of 7-AAD is lower than that of Propidium iodide (PI), the longer wavelength emission make it more useful for multiplexing assays in combination with other 488 nm-excited fluorochromes such as FITC and PE due to the minimal spectral overlap between these emissions.

Sample Protocol for Staining Cells

7-AAD staining is normally performed after all other stainings. It stains dead cells only. The following procedure can be adapted for most cell types. Growth medium, cell density, the presence of other cell types and other factors may influence staining. Residual detergent on glassware may also affect real or apparent staining of many organisms, causing brightly stained material to appear in solutions with or without cells present.

1). Make 1-10 mM DMSO stock solution. The DMSO stock solution is good for 6 months if stored at -20 °C.
2). Use the fixation protocol appropriate for your sample.
3). Pellet cells by centrifugation and resuspend the cells in buffered salt solutions or media, with optimal dye staining at pH 7.4. Adherent cells in culture may be stained *in situ* on cover slips or in the cell culture wells.
4). Add 7-AAD stain using the concentrations from 0.5 to 5 µM and incubate it for 15 to 60 minutes as a guide.
Note: In initial experiments, it may be best to try several dye concentrations over the entire suggested range to determine the concentration that yields optimal staining.
5). Measure the fluorescence intensity at Ex/Em = 545/650nm (use FL3 for flow cytometric analysis)
CAUTION: 7-AAD is a potential carcinogen. It is recommended that the user wear gloves, protective clothing, and eye/face protection in order to avoid contact with skin and eyes.

References

1. Kovalev AE, Iakovenko AA, Vekshin NL. (2004) [A study of interaction of 7-aminoactinomycin D with DNA by fluorescence correlation spectroscopy]. Biofizika, 49,1030.
2. Gaforio JJ, Serrano MJ, Algarra I, Ortega E, Alvarez de Cienfuegos G. (2002) Phagocytosis of apoptotic cells assessed by flow cytometry using 7-Aminoactinomycin D. Cytometry, 49, 8.