DAPI [4,6-Diamidino-2-phenylindole, dihydrochloride]

Ordering Information Storage Conditions

Product Numbers: 17507 (2 mL), 17510 (10 mg), Keep at -20 °C and desiccated 17511 (100 mg), 17513 (25 mg) Expiration date is 6 months from the date of receipt

Chemical and Physical Properties

Molecular Weight: 350.25

Solvent: water

Spectral Properties: Excitation = 358 nm; Fluorescence = 461 nm.

Biological Applications

DAPI is a fluorescent stain that binds strongly to DNA. It is used extensively in fluorescence microscopy. Since DAPI passes through an intact cell membrane, it can be used to stain live cells besides fixed cells. For fluorescence microscopy, DAPI is excited with ultraviolet light. When bound to double-stranded DNA, its absorption maximum is at 358 nm and its emission maximum is at 461 nm. One drawback of DAPI is that its emission is fairly broad. DAPI also binds to RNA although it is not as strongly fluorescent as it is when it binds to DNA. Its emission shifts to around 500 nm when bound to RNA. DAPI's blue emission is convenient for multiplexing assays since there is very little fluorescence overlap between DAPI and green-fluorescent molecules like fluorescein and green fluorescent protein (GFP), or red-fluorescent stains like Texas Red. Besides labeling cell nuclei, DAPI is also used for the detection of mycoplasma or virus DNA in cell cultures.

Sample Protocol for Staining Cells

Use the fixation protocol appropriate for your sample. DAPI staining is normally performed after all other staining.

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.

Pellet cells by centrifugation and resuspend the cells in buffered salt solutions or media, with optimal dye binding at pH 7.4. Adherent cells in culture may be stained *in situ* on cover slips or in the cell culture wells. Add DAPI stain using the concentrations between 0.5 and 5 μ M and incubate it for 15 to 60 minutes as a guide. 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.

References

- 1. Belonogova NM, Karamysheva TV, Biltueva LS, Perepelov EA, Minina JM, Polyakov AV, Zhdanova NS, Rubtsov NB, Searle JB, Borodin PM. (2006) Identification of all pachytene bivalents in the common shrew using DAPI-staining of synaptonemal complex spreads. Chromosome Res., 14, 673.
- Gichner T, Mukherjee A, Veleminsky J. (2006) DNA staining with the fluorochromes EtBr, DAPI and YOYO-1 in the comet assay with tobacco plants after treatment with ethyl methanesulphonate, hyperthermia and DNase-I. Mutat Res. 605, 17.
- Suda J, Travnicek P. (2006) Reliable DNA ploidy determination in dehydrated tissues of vascular plants by DAPI flow cytometry--new prospects for plant research. Cytometry A, 69, 273.
- Chen JM, Hong YH, Wang YP, Bowley S, Wan JM. (2006) [Physical localization of ribosomal genes and chromosome DAPI banding by in situ hybridization in Medicago sativa L]. Yi Chuan, 28, 184.
- 5. Samatadze TE, Muravenko OM, Bol'sheva NL, Amosova AB, Gostimsckii SA, Zelenin AV. (2005) [Investigation of chromosomes in varieties and translocation lines of pea Pisum sativum L. by FISH, Ag-NOR, and differential DAPI staining]. Genetika, 41, 1665.
- 6 Krishan A, Dandekar PD. (2005) DAPI fluorescence in nuclei isolated from tumors. J Histochem Cytochem, 53, 1033.
- Barcellona ML, Gammon S, Hazlett T, Digman MA, Gratton E. (2004) Polarized fluorescence correlation spectroscopy of DNA-DAPI complexes. Microsc Res Tech, 65, 205.
- Pancheva EV, Volkova VN, Kamzolkina OV. (2004) [DNA quantification in nuclei of cultivated mushroom with DAPI staining]. Tsitologiia, 46, 381.
- Daniel B, DeCoster MA. (2004) Quantification of sPLA2-induced early and late apoptosis changes in neuronal cell cultures using combined TUNEL and DAPI staining. Brain Res Brain Res Protoc, 13, 144.
- Li M, Wu RS, Tsai JS. (2003) DAPI derivative: a fluorescent DNA dye that can be covalently attached to biomolecules. Bioorg Med Chem Lett, 13, 4351.
- 11 Kubota Y, Kubota K, Tani S. (2000) DNA binding properties of DAPI (4',6-diamidino-2-phenylindole) analogs having an imidazoline ring or a tetrahydropyrimidine ring: groovebinding and intercalation. Nucleic Acids Symp Ser, 53.
- 12 Knobloch J, Kunz W, Grevelding CG. (2002) Quantification of DNA synthesis in multicellular organisms by a combined DAPI and BrdU technique. Dev Growth Differ, 44, 559.
- De Castro LF, Zacharias M. (2002) DAPI binding to the DNA minor groove: a continuum solvent analysis. J Mol Recognit, 15, 209.

Disclaimer: This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact our technical service representative for more information.