

DAPI

1 mg



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

Applications
 IF-F, IF-IC, IF-P, F

Species Cross-Reactivity
 All

Description: DAPI is supplied as a lyophilized powder in 1 mg units. It can be used to examine cellular DNA in fluorescent microscopy and cytometry applications.

Background: 4', 6-diamidino-2-phenylindole, dihydrochloride (DAPI) is a blue fluorescent DNA dye that targets double-stranded AT clusters in the DNA minor groove (1). One molecule of dye binds to each 3 base pairs of dsDNA and yields an approximate 20-fold fluorescent enhancement (2). The level of DAPI-DNA fluorescence is proportional to DNA content (3).

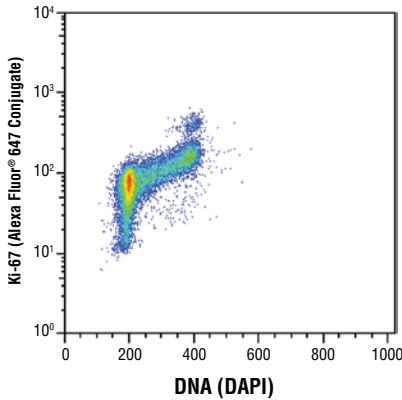
Fluorescent Properties:

- Free dye excitation maximum = 340 nm
- Free dye emission maximum = 488 nm
- DNA complex excitation maximum = 364 nm
- DNA complex emission maximum = 454 nm

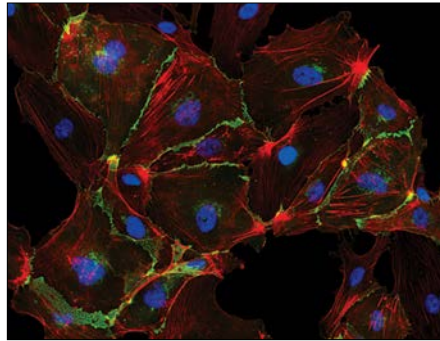
Molecular Formula: C₁₆H₁₅N₅2(HCl)

Background References:

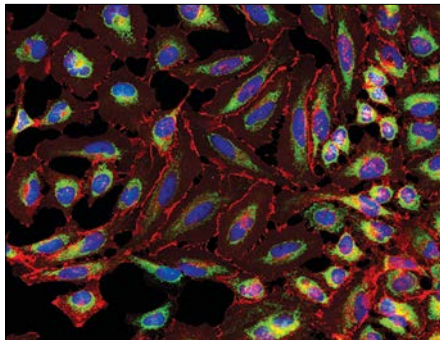
- (1) Portugal, J. and Waring, M.J. (1988) *Biochim Biophys Acta* 949, 158-68.
- (2) Kapuscinski, J. (1995) *Biotech Histochem* 70, 220-33.
- (3) Manzini, G. et al. (1983) *Nucleic Acids Res* 11, 8861-76.



Flow cytometric analysis of Jurkat cells using Ki-67 (D3B5) Rabbit mAb (Alexa Fluor® 647 Conjugate) #12075 and DAPI (DNA content).



Immunofluorescent analysis of HUVE cells using VE-Cadherin (D87F2) XP® Rabbit mAb #2500 (green) and DAPI (blue). Actin filaments were labeled with DyLight™ 554 Phalloidin #13054 (red).



Immunofluorescent analysis of HeLa cells using COX IV (3E11) Rabbit mAb #4850 (green), β-Catenin (L54E2) Mouse mAb (IF Preferred) #2677 (red) and DAPI (blue).

Storage: Store lyophilized or in solution at 4°C, desiccated. Protect from light. In lyophilized form, the chemical is stable for 24 months. Once in solution, use within 2-3 weeks to prevent loss of potency. For long-term storage, aliquot and store at -20°C to avoid multiple freeze/thaw cycles.

Directions for Use: DAPI is supplied as a lyophilized powder. For a 20 mg/ml stock, reconstitute the 1 mg in 50 µl deionized water or dimethylformamide. Please follow CST's recommended IF and Flow protocols. For both applications, following secondary detection:

Immunofluorescence: Counterstain with DAPI as the final step in your staining procedure. Rinse samples twice in PBS for five min each. Dilute DAPI stock solution to a concentration between 1-0.1 µg/ml in PBS and incubate for 5 min at room temperature in the dark. Rinse samples once in PBS and then prepare for imaging. Examine immediately using appropriate excitation wavelength.

Alternatively, dilute DAPI stock solution to a concentration between 1-0.1 µg/ml in mounting media, apply to cells, and prepare for imaging. Examine immediately using appropriate excitation wavelength.

Flow Cytometry: Rinse samples once in Incubation Buffer. Dilute DAPI stock solution to a concentration between 1.60-0.400 µg/ml in PBS and incubate for 15 min at room temperature in the dark before analyzing cells on flow cytometer.

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Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine
 Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—horse All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.