Annexin V-Cy5 Reagent

CATALOG #: 1013-200 200 assays
1013-1000 1000 assays

STORAGE CONDITIONS: Store at 4˚C. Do not freeze.

SHELF LIFE: 1 year under proper storage conditions

DESCRIPTION:

Annexin V-Cy5 is a bright fluorescent reagent for detecting of the early stages of apoptosis. During apoptosis, phosphatidylserine (PS) is translocated from the cytoplasmic face of the plasma membrane to the cell surface. Annexin V has a strong, Ca^{2+}-dependent affinity for PS and therefore serves as a probe for detecting apoptosis. Cy5 fluorescent dye produces an intense signal in the far-red region of the spectrum and therefore it is very useful for multiple labeling of cells with green and red colored fluorescent probes. Cy5 yields fluorescence with a λ_{max} emission of 670 nm.

ASSAY PROTOCOL:

A. Incubation of cells with Annexin V-Cy5:

1. Induce apoptosis by desired methods.
2. Collect 1 x 10^5 cells by centrifugation.
3. Resuspend cells in 500 µl of 1X Annexin V Binding Buffer (Cat.#1035-100).
4. Add 1 µl of Annexin V-Cy5.
5. Incubate at room temperature for 5 min in the dark.

   Proceed to B or C below depending on method of analysis.

B. Quantification by Flow Cytometry:

Analyze cells by flow cytometry (Ex = 649 nm; Em = 670 nm) using Helium-Neon Laser.

For adherent cells, trypsinize and gently wash cells with serum-containing medium before incubation with Annexin V-Cy5 (A.3-5).

C. Detection by Fluorescence Microscopy:

1. Place the cell suspension from Step A.5 on a glass slide, and cover with a glass coverslip.

   For analyzing adherent cells, grow cells directly on a coverslip. Following incubation (A.5), invert coverslip on a glass slide and visualize cells. The cells can also be washed with 1X Annexin V Binding Buffer and fixed in 2% formaldehyde before visualization. (Cells must be incubated with Annexin V-Cy5 before fixation because any cell membrane disruption can cause nonspecific binding of annexin V to PS on the inner surface of the cell membrane.)

2. Observe the cells under a fluorescence microscope using Cy5 filter or a FITC/Cy3/Cy5 triple band filter set (Chroma Technology) if you perform triple labeling with these dyes, or detect cells using CCD camera.

   Cells that have bound Annexin V-Cy5 will show bright red-blue staining on the plasma membrane.

FOR RESEARCH USE ONLY! Not to be used in humans!

RELATED PRODUCTS:

Apoptosis Detection Kits & Reagents
- Annexin V Kits & Bulk Reagents
- Caspase Assay Kits & Reagents
- Mitochondrial Apoptosis Kits & Reagents
- Nuclear Apoptosis Kits & Reagents
- Apoptosis Inducers and Set
- Apoptosis siRNA Vectors

Cell Fractionation System
- Mitochondria/Cytosol Fractionation Kit
- Nuclear/Cytosol Fractionation Kit
- Membrane Protein Extraction Kit
- Cytosol/Particulate Rapid Separation Kit
- Mammalian Cell Extraction Kit
- FractionPREP Fractionation System

Cell Proliferation & Senescence
- Quick Cell Proliferation Assay Kit
- Senescence Detection Kit
- High Throughput Apoptosis/Cell Viability Assay Kits
- LDH-Cytotoxicity Assay Kit
- Bioluminescence Cytotoxicity Assay Kit
- Live/Dead Cell Staining Kit

Cell Damage & Repair
- HDAC Fluorometric & Colorimetric Assays & Drug Discovery Kits
- HAT Colorimetric Assay Kit & Reagents
- DNA Damage Quantification Kit
- Glutathione & Nitric Oxide Fluorometric & Colorimetric Assay Kits

Signal Transduction
- cAMP & cGMP Assay Kits
- Akt & JNK Activity Assay Kits
- Beta-Secretase Activity Assay Kit

Adipocyte & Lipid Transfer
- Recombinant Adiponectin, Survivin, & Leptin
- CETP Activity Assay & Drug Discovery Kits
- PLTP Activity Assay & Drug Discovery Kits
- Total Cholesterol Quantification Kit

Molecular Biology & Reporter Assays
- siRNA Vectors
- Cloning Insert Quick Screening Kit
- Mitochondrial & Genomic DNA Isolation Kits
- 5 Minutes DNA Ligation Kit
- 20 Minutes Gel Staining/Destaining Kit
- β -Galactosidase Staining Kit & Luciferase Reporter Assay Kit

Growth Factors and Cytokines

Monoclonal and Polyclonal Antibodies

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