

Annexin V-PE-Cy5 Reagent

CATALOG #:	1015-1000
AMOUNT:	1000 assays (1 ml)
STORAGE CONDITIONS:	Store at 4°C. Do not freeze.
SHELF LIFE:	1 year under proper storage conditions

DESCRIPTION:

Annexin V-PE-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, Ca²⁺-dependent affinity for PS and therefore serves as a probe for detecting apoptosis. The one-step staining procedure takes only 10 minutes. In addition, the assay can be directly performed on live cells. Annexin V-PE-Cy5 stained cells can be detected by flow cytometry or fluorescence microscopy.

ASSAY PROTOCOL:

A. Incubation of cells with Annexin V-PE:

1. Induce apoptosis by desired methods.
2. Collect 1 x 10⁵ 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-PE-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 Annexin V-PE-Cy5 binding by flow cytometry (Ex = 488 nm; Em = 670 nm).

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

C. Detection by Fluorescence Microscopy

1. Place the cell suspension from Step A.5 on a glass slide. Cover the cells with a glass coverslip.

For analyzing adherent cells, grow cells directly on a coverslip. Following incubation (A.5), invert coverslip on glass slide and visualize cells. The cells can also be washed and fixed in 2% paraformaldehyde before visualization. (Cells must be incubated with Annexin V-PE-Cy5 before fixation since 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 a PE-Cy5 filter.
Cells which have bound Annexin V-PE-Cy5 will show bright purple staining in the plasma membrane.

STORAGE AND STABILITY:

Store at 4°C. Stable for one year under proper storage conditions.

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 Inhibitors
- 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 Kit
- 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

Quality Antibodies for Apoptosis and Signal Transduction Molecules