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DATASHEET

Annexin V Apoptosis Detection Kit (FITC)

Catalogue No.:abx290014

Annexin V Apoptosis Detection Kit is an Apoptosis Detection Kit conjugated to FITC.

Target: Annexin V

Tested Applications: FCM

Form: Liquid

Conjugation: FITC

Storage: Store in the dark at 2-8 °C

GeneID: 308

Buffer: The conjugate is provided in liquid form in buffer containing Antibody Stabilizer, PBS, pH 7.4.

Directions for use: FITC Absorption/Emission Maximum: 495/519 nm. Recommended Band Pass Filter: 530/30 nm.

Protocol for staining cells:

1. Prepare the Annexin V Binding Buffer: 10 mM Hepes/NaOH (pH 7.4), 140 mM NaCl, 2.5 mM $CaCl_2$.
2. Induce apoptosis in cells using the desired method. A negative control should be prepared using untreated cells to define the basal level of apoptotic and necrotic (dead) cells.
3. Harvest the cells after apoptosis induction and wash in PBS at room temperature.
4. Wash cells twice with PBS at room temperature and resuspend cells in 1X Annexin Binding buffer at a concentration 1×10^6 cells/ml.
5. Add 5 µl of the Annexin V-FITC reagent to each 100 µl of cell suspension (up 1×10^5 cells).
6. Incubate the cells at room temperature for 15 minutes in the dark.
7. After incubation period, add 400 µl of 1X Annexin Binding buffer. Analyze by flow cytometry within one hour.

Troubleshooting:

• No Annnexin V-FITC fluoresence observed: apoptosis may not have been induced in the cells.
• Elevated Annexin V-FITC staining: apoptosis is an ongoing process, so cells that are stained with Annexin V should not be kept for prolonged periods of time before taking measurements.
• Adherents cells may be released from their substrate by using trypsin. Trypsinized cells can be affected by the integrity of the plasma membrane. For adherent cells, it is recommended to remove supernatant with floating cells and replace media before adding any substances of interest, or remove culture medium from cells, and immerse slide into cold (2-8 °C) 1X PBS.

Note: This product is for research use only.