Product Datasheet
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Annexin V

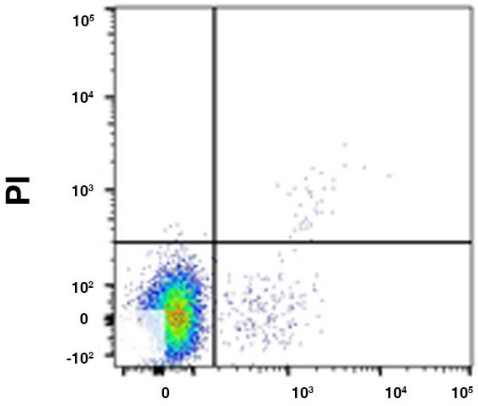
FITC Apoptosis Detection Kit

Apoptosis & Protease Detection Kits

Product Information

Product No.: A432

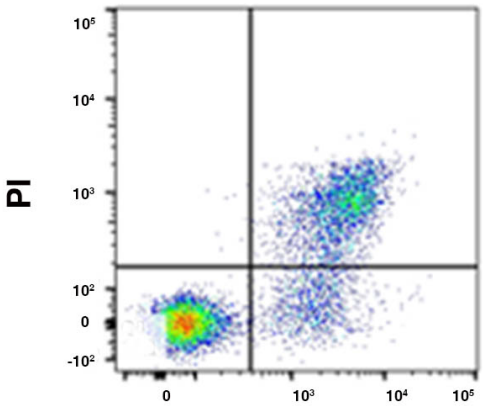
Storage: Sterile 2 to 8°C



Jurkat cells were treated for 6 hours with 1µg/mL anti-Fas anitbody. Cells were spun down, washed with 1x PBS, spun and resuspended in Annexin V binding buffer ($2x10^6$ cells/mL buffer). 100µL of cells ($2x10^5$ cells) were added to FACs tubes and stained with 5µL Annexin V FITC and 5µL propidium iodide. (Leinco Annexin V-FITC Apoptosis Detection Kit, Product # A432)

Bottom right quadrant: Viable cells undergoing early apoptosis.

Top right quadrant: Cells that are in late-stage apoptosis.



Jurkat cells were treated overnight with 1µg/mL anti-Fas anitbody. Cells were spun down, washed with 1x PBS, spun and resuspended in Annexin V binding buffer ($2x10^6$ cells/mL buffer). 100µL of cells ($2x10^5$ cells) were added to FACs tubes and stained with 5µL Annexin V FITC and 5µL propidium iodide. (Leinco Annexin V-FITC Apoptosis Detection Kit, Product # A432)

Bottom right quadrant: Viable cells undergoing early apoptosis.

Top right quadrant: Cells that are in late-stage apoptosis.

HeLa cells were treated with 5uM camptothecin for approximately 24 hours for apoptosis induction. Cells were spun down, washed with 1x ice cold PBS, spun and re-suspended in ice cold Annexin V-FITC buffer and filtered through 80um Nitex to remove clumps. Cells were counted on a Z2 coulter counter and diluted to 1x10^6 cells/ml in Annexin V buffer. 100ul of cells were added into 5ml tubes (1x10^5 cells/tube), and stained with 5ul of Annexin V-FITC along with 10ul Propidium Iodide.

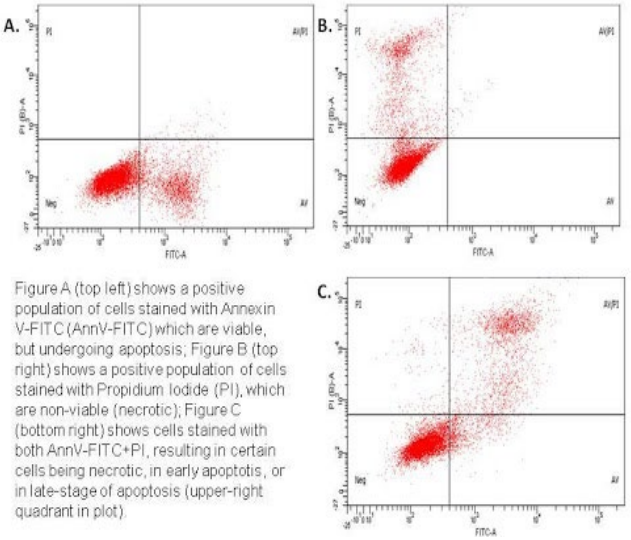


Figure A (top left) shows a positive population of cells stained with Annexin V-FITC (AnnV-FITC) which are viable, but undergoing apoptosis; Figure B (top right) shows a positive population of cells stained with Propidium Iodide (PI), which are non-viable (necrotic); Figure C (bottom right) shows cells stained with both AnnV-FITC+PI, resulting in certain cells being necrotic, in early apoptotis, or in late-stage of apoptosis (upper-right quadrant in plot).

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Product Description

Background:

Apoptosis, the physiological process of cell death, is a naturally occurring phenomenon. It is normally a tightly regulated process; however, if deregulated, tumor growth can result. Apoptotic processes can be measured in the laboratory easily and most effectively by using a FITC-labeled Annexin V protein to stain cells and analyze them via flow cytometry. It is a useful method to screen and analyze cells in a high throughput manner, resulting in a clearer understanding of the cellular response to various stimuli.

When cells are undergoing cell death, phosphatidylserine (PS), a phospholipid which makes up a portion of the cell membrane, will translocate to the outside-facing side of the membrane. This translocation in the cell death mechanism exposes PS to the extracellular environment. Annexin V, a 35-36 kDa phospholipid-binding protein, has a high affinity for PS (KDa=7nmol/l). Once Annexin V is bound to the apoptotic cell it acts as a beacon for cells that have undergone apoptosis. Annexin V is often conjugated to FITC so the amount of signal can be measured and analyzed from labeled cells using a flow cytometer. The translocation of PS to the outside or exposed side of the membrane is an early event in the apoptotic process. Therefore, Annexin V staining is considered a marker for early-stage apoptosis. Often times, Annexin V is paired with Propidium Iodide (PI) stain in order to analyze for late-apoptotic or dead cells. Early-stage apoptotic cells will only take up the Annexin V stain but will remain PI negative, and late-stage apoptotic cells will be positive for both Annexin V and PI.

Country of Origin

USA

References

1) Piazza, GA. *et al.* (2016) *Oncotarget.* 7(5):5353-65.PubMed
2) Gassman, NR. *et al.* (2018) *Environ Toxicol.* 33(3):333-342.PubMed
3) Gelles, JD. and Chipuk, JE. (2016) *Cell Death Dis.* 7(12): e2493.

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