



CELL TECHNOLOGY, INC.

APO ACTIVE 3™ PE™

Antibody Specific Active Caspase 3 Phycoerythrin (PE) Detection Kit

Key Benefits

- **Highly Specific:** Cell Technology's APO-ACTIVE 3 Kit is highly specific for active human and murine caspase 3. Other assays require the utilization of peptide based (DEVD) reagents that tend to cross-react with caspase 7 and other caspases.
- **Compatible with Multiple Fluorometric Modalities:**
 - Flow cytometry
 - 96 well plate reader
 - Fluorescence microscope
- **Reliable:** Yields both quantitative and qualitative results. Gives a strong positive signal. The kit can be used in conjunction with other antibodies or stains.
- **Free Technical Support:** Cell Technology offers free technical support to help the researcher optimize the application of the APO-ACTIVE detection kit.
- **Easy to Use:** No need to make cell lysates or run western blots. Cells can be fixed and analyzed later.

Background

Apoptosis is an evolutionarily conserved form of cell suicide, which follows a specialized cellular process. The central component of this process is a cascade of proteolytic enzymes called caspases. These enzymes participate in a series of reactions that are triggered in response to pro-apoptotic signals and result in cleavage of protein substrates, causing the disassembly of the cell (1).

Caspases have been identified in organisms ranging from *C. elegans* to humans. The mammalian caspases play distinct roles in apoptosis and inflammation. In apoptosis, caspases are responsible for proteolytic cleavages that lead to cell disassembly (effector caspases), and are involved in upstream regulatory events (initiator caspases). An active caspase consists of two large (~20 kD) and two small (~10 kD) subunits to form two heterodimers which associate in a tetramer (2-4). As is common with other proteases, caspases are synthesized as precursors that undergo proteolytic maturation, either autocatalytically or in a cascade by enzymes with similar specificity (5). Caspase 3, also known as CPP-32, Apopain or Yama, is a key effector caspase in the apoptotic pathway (6). It is present in many different cell lineages and is responsible for the cleavage of a variety of molecules such as poly ADP-ribose polymerase (PARP), protein kinase Cd, actin and DNA-dependent protein kinase (7,8).

Cross Reactivity

Human, Mouse and Rat

Assay Principle

Cell Technology's APO ACTIVE CASPASE 3 Kit utilizes a rabbit affinity purified polyclonal antibody raised against amino acid 163-175 of murine caspase 3 (9). This neo epitope is present on the p20 subunit of cleaved caspase 3 (9). Cells undergoing apoptosis are fixed and permeabilized prior to the addition of the antibody. A secondary PE labeled goat anti rabbit antibody is used to visualize the bound rabbit anti caspase 3 polyclonal antibody.

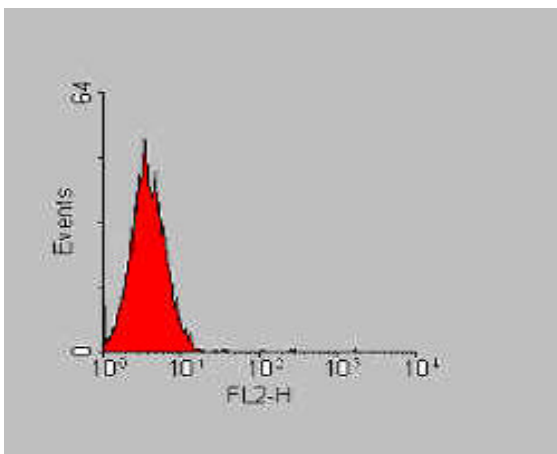


Figure A

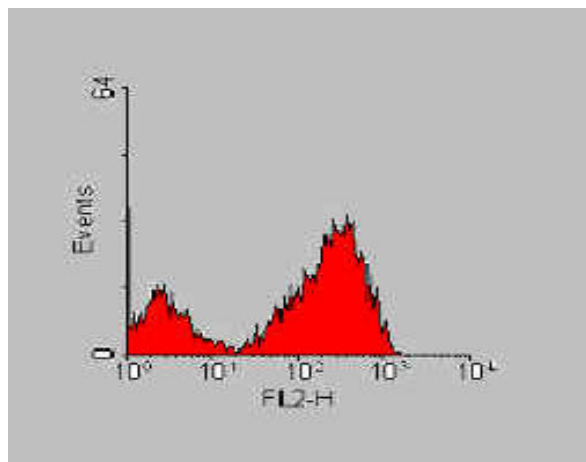


Figure B

Jurkat cells treated with (A) DMSO or (B) staurosporin for 2 hours. The cells were washed and fixed for 15 minutes, permeabilized with .1% Triton X-100 and stained with Rabbit anti caspase 3 antibody for 1 hour. The samples were then stained with PE labeled Goat anti Rabbit antibody.

Kit Content

- 1) 1 vial of Rabbit anti cleaved caspase 3 affinity purified polyclonal antibody.
- 2) 1 vial of PE labeled Goat anti Rabbit affinity purified polyclonal antibody.
- 3) 1 vial of 10X fixative.

Ordering Information

PE APO ACTIVE CASPASE 3 Kit.

Catalog #	Size	Price (US\$)
PAB200-1	25 Tests	145
PAB200-2	100 Tests	425

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