Technical Data Sheet

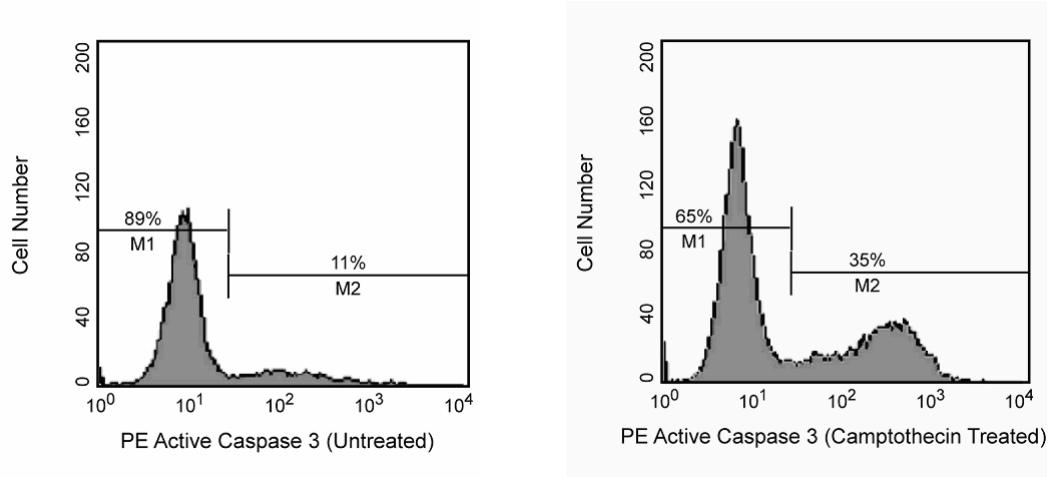
PE Rabbit Anti- Active Caspase-3

Product Information

Material Number:	**550821**
Alternate Name:	CPP32; Yama; Apopain
Size:	100 tests
Vol. per Test:	20 µl
Clone:	C92-605
Isotype:	Rabbit IgG
Reactivity:	QC Testing: Human Tested in Development: Mouse
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The caspase family of cysteine proteases plays a key role in apoptosis and inflammation. Caspase-3 is a key protease that is activated during the early stages of apoptosis and, like other members of the caspase family, is synthesized as an inactive pro-enzyme that is processed in cells undergoing apoptosis by self-proteolysis and/or cleavage by another protease. The processed forms of caspases consist of large (17-22 kDa) and small (10-12 kDa) subunits which associate to form an active enzyme. Active caspase-3, a marker for cells undergoing apoptosis, consists of a heterodimer of 17 and 12 kDa subunits which is derived from the 32 kDa pro-enzyme. Active caspase-3 proteolytically cleaves and activates other caspases, as well as relevant targets in the cytoplasm, e.g., D4-GDI and Bcl-2, and in the nucleus (e.g. PARP). This antibody has been reported to specifically recognize the active form of caspase-3 in human and mouse cells. It has not been reported to recognize the pro-enzyme form of caspase-3.



Flow cytometric analysis of apoptotic and non-apoptotic populations for active caspase-3. *Jurkat cells (Human T-cell leukemia; ATCC TIB-152) were left untreated (left panel) or treated with 4 µM of camptothecin for 4 hr to induce apoptosis (right panel). Cells were washed once in PBS, then fixed and permeabilized using the BD Cytofix/Cytoperm™ Kit (Cat. No. 554714) for 20 min at room temperature (RT), pelleted and washed with BD Perm/Wash™ buffer (component of Cat. No. 554714). Cells were subsequently stained with the PE rabbit anti- active caspase-3 antibody (clone C92-605). Cells were then washed and resuspended in BD Perm/Wash™ buffer before analyzing by flow cytometry. The results show that untreated cells were primarily negative for active caspase-3 (left panel, M1); whereas over one third of the treated cells were positive for active caspase-3 staining (right panel, M2).*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
The antibody was conjugated with R-PE under optimum conditions, and unconjugated antibody and free PE were removed.
Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

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Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
554714	BD Cytofix/Cytoperm™ Fixation/Permeablization Kit	250 tests	(none)

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10e6 cells in a 100-µl experimental sample (a test).
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/pharmingen/colors.
4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
5. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Dai C, Krantz SB. Interferon gamma induces upregulation and activation of caspases 1, 3, and 8 to produce apoptosis in human erythroid progenitor cells. *Blood.* 1999; 93(10):3309-3316.(Biology)

Fujita N, Tsuruo T. Involvement of Bcl-2 cleavage in the acceleration of VP-16-induced U937 cell apoptosis. *Biochem Biophys Res Commun.* 1998; 246(2):484-488.(Biology)

Thornberry NA, Lazebnik Y. Caspases: enemies within. *Science.* 1998; 281(5381):1312-1316.(Biology)