Technical Data Sheet

V450 Rabbit Anti-Active Caspase-3

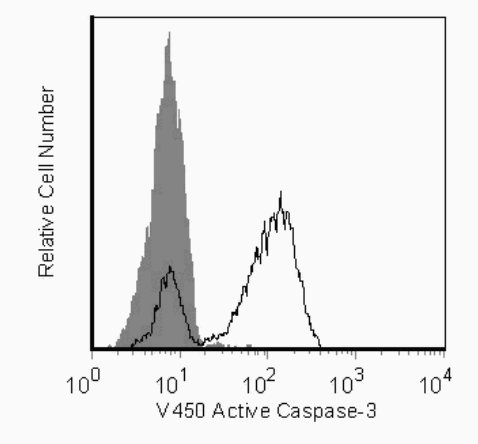
Product Information

Material Number:	**560627**
Alternate Name:	CPP32; Yama; Apopain
Size:	50 tests
Vol. per Test:	5 µl
Clone:	C92-605
Immunogen:	Human Active Caspase-3 Fragment
Isotype:	Rabbit IgG
Reactivity:	QC Testing: Human Predicted: Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer 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.

The antibody is conjugated to BD Horizon™ V450, which has been developed for use in multicolor flow cytometry experiments and is available exclusively from BD Biosciences. It is excited by the **V**iolet laser Ex max of 406 nm and has an Em Max at **450** nm. Conjugates with BD Horizon™ V450 can be used in place of Pacific Blue™ conjugates.



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 (shaded) or were treated with 4-12 µM of camptothecin (Sigma-Aldrich Cat. No. C9911) for 4-6 hr to induce apoptosis (unshaded). Cells were washed once in 1X PBS, then fixed and permeabilized using the BD Cytofix/Cytoperm™ Kit (Cat. No. 554714) followed by staining with the BD Horizon™ V450 Rabbit Anti-Active Caspase-3 antibody. Histograms were derived from gated events based on light scattering characteristics for Jurkat cells. Flow cytometry was performed on a BD™ LSR II flow cytometry system.*

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with BD Horizon™ V450 under optimum conditions, and unreacted BD Horizon™ V450 was removed.

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)

BD Biosciences

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Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10^6 cells in a 100-µl experimental sample (a test).
2. BD Horizon™ V450 has a maximum absorption of 406 nm and maximum emission of 450 nm. Before staining with this reagent, please confirm that your flow cytometer is capable of exciting the fluorochrome and discriminating the resulting fluorescence.
3. 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.
4. Pacific Blue™ is a trademark of Molecular Probes, Inc., Eugene, OR.
5. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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