

Datasheet for ABIN2690898

Ac-DEVD-AMC Caspase-3 Fluorogenic Substrate[Go to Product page](#)**2** Publications

Overview

Quantity: 1 mg

Application: Functional Studies (Func)

Product Details

Brand: BD Pharmingen™

Characteristics: Members of the ICE/CED-3 cysteine protease family have key roles in inflammation and mammalian apoptosis. The ICE family member Caspase-3 (also known as CPP32, Yama, apopain) is activated early in apoptosis and appears to be involved in the proteolysis of several important molecules, including poly (ADP ribose) polymerase (PARP). Activated Caspase-3 cleaves PARP from its 116 kDa to an 85 kDa residual fragment. The cleavage site in PARP is C-terminal to Asp-216. The upstream sequence of the cleavage site, DEVD (Asp-Glu-Val-Asp), is utilized as a basis for the highly specific Caspase-3 substrate, Ac (N-acetyl)-DEVD-AMC (7-amino-4-methylcoumarin). Ac-DEVD-AMC is a synthetic tetrapeptide fluorogenic substrate for Caspase-3 (CPP32) and contains the amino acid sequence of the PARP cleavage site at Asp-216. The tetrapeptide substrate can be used to identify and quantify the Caspase-3 activity in apoptotic cells. Caspase-3 cleaves the tetrapeptide between D and AMC, thus releasing the fluorogenic AMC, which can be quantified in a spectrofluorometer. The substrate can also be used to study the inhibition of Caspase-3 by the tetrapeptide aldehyde, Ac-DEVD-CHO or any other inhibitor of Caspase-3. Protease Assay Using Substrate for Caspase-3 (CPP32), Ac-DEVD-AMC (Cat. No. 556449). Non-apoptotic (left panel) and apoptotic HPB-ALL leukemia cell lysates (right panel) were incubated in Ac-DEVD-AMC protease assay buffer at 37 °C for 1 hour. In each assay, the AMC liberated from the fluorogenic substrate was measured in a spectrofluorometer with an excitation wavelength of 380 nm and emission wavelength range of 430-460 nm. The results show that the Ac-DEVD-AMC substrate was cleaved by apoptotic cell lysate but not by

Product Details

the non-apoptotic lysate. In the right panel, cells were induced to undergo apoptosis using an Anti-Human Fas monoclonal antibody, clone DX2 (Cat. No. 555670). Apoptosis was measured after 5 hour of incubation with 2 µg/mL Fas mAb and 1 µg/mL Protein G. We have found that the addition of Protein G can significantly enhance the efficiency of Fas mAb to induce apoptosis.

Application Details

Application Notes:	Optimal working dilution should be determined by the investigator.
Comment:	BD Pharmingen™ Ac-DEVD-AMC Caspase-3 Fluorogenic Substrate - Purified
Restrictions:	For Research Use only

Handling

Format:	Lyophilized
Reconstitution:	Reconstitute in 1 mL DMSO to yield 1 mg/mL peptide in DMSO. Store the reconstituted substrate at -20 °C for up to 1-2 months and avoid repeated freeze-thaw cycles, which greatly alter product stability.
Buffer:	Lyophilized powder
Storage:	-20 °C
Storage Comment:	Store undiluted at -20°C. Reconstitute the substrate before use. Reconstitute in 1 ml DMSO to yield 1 mg/ml peptide in DMSO. Store the reconstituted substrate at -20°C for up to 1-2 months and avoid repeated freeze-thaw cycles, which greatly alter product stability.

Publications

Product cited in:	Thornberry, Lazebnik: "Caspases: enemies within." in: Science (New York, N.Y.) , Vol. 281, Issue 5381, pp. 1312-6, (1998) (PubMed).
	Nicholson, Ali, Thornberry, Vaillancourt, Ding, Gallant, Gareau, Griffin, Labelle, Lazebnik: "Identification and inhibition of the ICE/CED-3 protease necessary for mammalian apoptosis." in: Nature , Vol. 376, Issue 6535, pp. 37-43, (1995) (PubMed).