Datasheet for ABIN5540294

anti-DFFA antibody (AA 1-331)

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Overview

Quantity:	0.1 mg
Target:	DFFA
Binding Specificity:	AA 1-331
Reactivity:	Human
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This DFFA antibody is un-conjugated
Application:	Western Blotting (WB)

Product Details

Immunogen:	Recombinant full-length human DFF45 (1-331 a.a.)
Clone:	6B8
Isotype:	IgG1
Specificity:	This antibody reacts with DFF45/ICAD. The antibody detects DFF45/ICAD-L (45 kDa) and DFF45/ICAD-S (40 kDa) by Western blot.
Purification:	Protein A agarose

Target Details

Target:	DFFA
Alternative Name:	dffa,icad (DFFA Products)

Target Details

Background:	In the apoptotic pathway, various stimuli activate a protease cas cade of caspases, eventually causing the degradation of chromosomal DNA. DFF45/ICAD is a downstream regulator of DNA degradation. DFF45 is human homologue of mouse ICAD that was identified as an inhibitor of caspase-activated deoxyribonuclease (CAD). CAD is apoptotic DNase with high specific activity that ex ists as an inactive complex with DFF45/ICAD in living cells. Upon apoptosis, caspase-3 is activated and cleaves DFF45/ICAD, which releases CAD from DFF45/ICAD. CAD can then enter the nucleus to degrade the chromosomal DNA. DFF45/ICAD may inhibit not only the DNase activity of CAD, but also its translocation into the nucleus. Furthermore, functional CAD can only be synthesized in the presence of DFF45/ICAD in a cell-free system, suggesting DFF45/ICAD functions as a chaperone for CAD during its sythesis.
UniProt:	O00273
Pathways:	Apoptosis, Caspase Cascade in Apoptosis

Application Details

Application Notes:	Western blot: 1 µg/mL for chemiluminescence detection system.
Protocol:	SDS-Page & Western Blotting 1) Wash the 1x10 7 cells 3 times with PBS and suspend with 1 mL of Laemmli's sample buffer. 2) Boil the samples for 2 minutes and centrifuge. Load 20 µ L of the sample per lane in a 1 mm thick SDS-polyacrylamide gel for electrophoresis. 3) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm 2 for 1 hour in a semi-dry transfer system (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20 % MeOH). See the manufacture's manual for specific transfer procedure. 4) To reduce nonspecific binding, soak the membrane in 5 % skimmed milk (in PBS, pH 7.2) for 1 hour at room temperature, or overnight at 4 o C. 5) Incubate the membrane with primary antibody diluted with PBS, pH 7.2 containing 1 % skimmed milk as suggested in the APPLICATIONS for 1 hour at room temperature. (The optimal antibody concentration will depend on the experimental conditions.) 6) Wash the membrane with PBS-T [0.05 % Tween-20 in PBS] (5 minutes x 3 times). 7) Incubate the membrane with the 1:10,000 HRP-conjugated anti-mouse IgG diluted with 1 % skimmed milk (in PBS, pH 7.2) for 1 hour at room temperature. 8) Wash the membrane with PBS-T (5 minutes x 3 times). 9) Wipe excess buffer from the membrane, then incubate it with appropriate chemiluminescence reagents for 1 minute. Remove extra reagent from the membrane by dabbing with a paper towel, and seal it in plastic wrap. Expose to an X-ray film in a dark room for 1 minute. Develop the film as usual. The conditions for exposure and development may vary. (Positive controls for Western blotting Jurkat, Raji, A431, MRC-5, ZR-75-1, Hep-G2)

Application Details

Restrictions:	For Research Use only

Handling

Format:	Lyophilized
Buffer:	PBS (pH 7.2)/ 1 % sucrose. Contains no preservatives.
Preservative:	Without preservative
Storage:	4 °C,-20 °C
Storage Comment:	Prior to reconstitution store at 2-8°C. Following reconstitution store undiluted at -20°C. Avoid repeated freezing and thawing. Shelf life: one year from despatch.
Expiry Date:	12 months