SANTA CRUZ BIOTECHNOLOGY, INC.

ICAD (B-6): sc-17818

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

The CED/ICE family of cysteine proteases plays a pivotal role in mediating apoptosis through the proteolysis of specific targets. Among the targets are poly (ADP-ribose) polymerase (PARP), gelsolin, DFF-45/ICAD and the nuclear lamins. PARP is a nuclear protein that is specifically cleaved by CPP32 and Mch2, but not by ICE, into a signature apoptotic fragment. Gelsolin is cleaved by CPP32 to an active form that severs actin filaments in a Ca^{2+}-independent manner. In addition to binding Actin, gelsolin can form complexes with fibronectin, which may be important for localizing gelsolin to inflammatory sites. DFF-45/ICAD, the subunit of DNA fragmentation factor, is cleaved by CPP32 to generate an active factor that induces DNA fragmentation. The nuclear Lamin A is cleaved by Mch2, but not CPP32. Nuclear Lamin B is fragmented as a consequence of apoptosis by an unidentified member of the ICE family.

REFERENCES

1. Lind, S.E., et al. 1984. Human plasma gelsolin binds to Fibronectin. J. Biol. Chem. 259: 13262-13266.
2. Fernandes-Alnemri, T., et al. 1995. Mch3, a novel human apoptotic cysteine protease highly related to CPP32. Cancer Res. 55: 6045-6052.
3. Takahashi, A., et al. 1996. Cleavage of lamin A by Mch2α but not CPP32: multiple interleukin 1 β-converting enzyme-related proteases with distinct substrate recognition properties are active in apoptosis. Proc. Natl. Acad. Sci. USA 93: 8395-8400.
4. Rao, L., et al. 1996. Lamin proteolysis facilitates nuclear events during apoptosis. J. Cell Biol. 135: 1441-1455.
5. Liu, X., et al. 1997. DFF, a heterodimeric protein that functions downstream of caspase-3 to trigger DNA fragmentation during apoptosis. Cell 89: 175-184.
6. Salvesen, G.S., et al. 1997. Caspases: intracellular signaling by proteolysis. Cell 91: 443-446.
7. Kothakota, S., et al. 1997. Caspase-3-generated fragment of gelsolin: effector of morphological change in apoptosis. Science 278: 294-298.

CHROMOSOMAL LOCATION

Genetic locus: DFFA (human) mapping to 1p36.22.

SOURCE

ICAD (B-6) is a mouse monoclonal antibody raised against amino acids 1-331 representing full length ICAD of human origin.

PRODUCT

Each vial contains 200 µg IgG_{2b} kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

APPLICATIONS

ICAD (B-6) is recommended for detection of ICAD of human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:500), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for ICAD siRNA (h): sc-35624, ICAD shRNA Plasmid (h): sc-35624-SH and ICAD shRNA (h) Lentiviral Particles: sc-35624-V.

Molecular Weight of DFF-45 splice variant: 45 kDa.

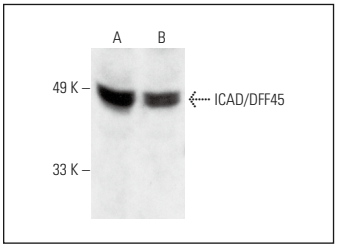
Molecular Weight of DFF-35 splice variant: 35 kDa.

Positive Controls: U-937 cell lysate: sc-2239, Jurkat whole cell lysate: sc-2204 or HeLa whole cell lysate: sc-2200.

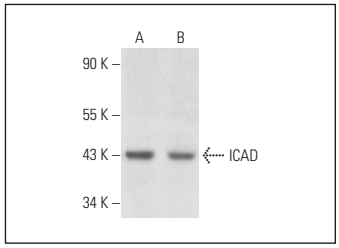
RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

DATA



ICAD (B-6): sc-17818. Western blot analysis of ICAD/DFF expression in HeLa (**A**) and U-937 (**B**) whole cell lysates.



ICAD (B-6): sc-17818. Western blot analysis of ICAD expression in Jurkat (**A**) and HeLa (**B**) whole cell lysates.

SELECT PRODUCT CITATIONS

1. Ali, M., et al. 2022. Small-molecule targeted therapies induce dependence on DNA double-strand break repair in residual tumor cells. Sci. Transl. Med. 14: eabc7480.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.