

Product Information

ANTI-DFF45/ICAD, C-TERMINAL

Developed in Rabbit, IgG Fraction of Antiserum

Product Number **D 3313**

Product Description

Anti-DFF45/ICAD is developed in rabbit using a synthetic peptide corresponding to amino acids 313-331 at C-terminus of human DFF45¹ as immunogen.

Anti-DFF45/ICAD recognizes DFF45 by immunoblotting (45 kDa)² and immunoprecipitation.

DFF45 and DFF40 (also termed ICAD and CAD) are two subunits that make up the heterodimeric protein caspase-activated DNase or DNA Fragmentation Factor (DFF) that triggers DNA fragmentation during apoptosis.¹

DFF exists as an inactive cytoplasmic protein until activated by apoptotic signals. DFF45 functions as both a chaperone, mediating the correct folding of DFF40, and an inhibitor of DFF40.³ In response to apoptotic signals, DFF45 is cleaved by caspase-3 at two sites. This releases active nuclease, DFF40.^{2,4-7} DFF40 seems to oligomerize, forming a large, functional complex which breaks down DNA by introducing double-strand breaks. Furthermore, DFF40 appears to interact directly with histone H1 that may stimulate its activity.⁸

Reagents

Anti-DFF45/ICAD, C-Terminal is supplied as 0.5 mg/ml of IgG fraction of antiserum in phosphate buffered saline, 0.02% sodium azide.

Precautions and Disclaimer

Due to the sodium azide content a material safety data sheet (MSDS) has been sent to the attention of the safety officer at your institution. Consult the MSDS for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8°C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working

dilution samples should be discarded if not used within 12 hours.

Product Profile

Recommended working concentration is approximately 0.25 µg/ml (1:1,000-1:2,000 dilution) by immunoblotting using a total HeLa cell lysate. A 45 kDa band should be detected in non-apoptotic cells.

Note: In order to obtain best results and assay sensitivities in different techniques and preparations, we recommend determining optimal working dilutions by titration test.

References

1. Liu X., et al., DFF, a heterodimeric protein that functions downstream of caspase-3 to trigger DNA fragmentation during apoptosis. *Cell*, **89**, 175-184 (1997).
2. Tang D., and Kidd, V.J., Cleavage of DFF-45/ICAD by multiple caspases is essential for its function during apoptosis. *J. Biol. Chem.*, **273**, 28549-28552 (1998).
3. McCarty, J.S., et al., Study of DFF45 in its role of chaperone and inhibitor: two independent inhibitory domain of DFF40 nuclease activity. *Biochem. Biophys. Res. Commun.*, **264**, 176-180 (1999).
4. Enari M., et al., A caspase-activated Dnase that degrades DNA during apoptosis, and its inhibitor ICAD. *Nature*, **391**, 43-50 (1998).
5. Sakahira H, et al., Cleavage of CAD inhibitor in CAD activation and DNA degradation during apoptosis. *Nature*, **391**, 96-99 (1998).
6. Wolf, B.B., et al., Caspase-3 is the primary activator of apoptotic DNA fragmentation via DNA fragmentation factor-45/inhibitor of caspase-activated Dnase inactivation. *J. Biol. Chem.*, **274**, 30651-30656 (1999).
7. Wohrl, W., and Hacker, G., Extent and limitation of the control of nuclear apoptosis by DNA-fragmenting factor. *Biochem. Biophys. Res. Commun.*, **254**, 552-558 (1999).
8. Lui Z, et al., Activation of the apoptotic endonuclease DFF40 (caspase-activated DNase or nuclease). *J. Biol. Chem.*, **274**, 13836-13840 (1999).

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