

UK Office

Everest Biotech Ltd Cherwell Innovation Centre 77 Heyford Park Upper Heyford Oxfordshire OX25 5HD UK

Enquiries:

info@everestbiotech.com Sales: sales@everestbiotech.com Tech support: support@everestbiotech.com

Tel: +44 (0)1869 238326

www.everestbiotech.com

Research Use Only. Not for diagnostic or therapeutic use.

EB05427 - Goat Anti-DAP3 Antibody

Size: 100µg specific antibody in 200µl



Target Protein

Principal Names: DAP3, death associated protein 3, DAP-3, DKFZp686G12159, MGC126058, MGC126059, MRP-S29, MRPS29, bMRP-10, death-associated protein 3, mitochondrial 28S ribosomal protein S29 Official Symbol: DAP3 Accession Number(s): NP_387506.1; NP_001186779.1; NP_001186780.1 Human GeneID(s): 7818 Important Comments: This antibody is expected to recognise isoform 1 (NP_387506.1), isoform 2 (NP_001186779.1) and isoform 3 (NP_001186780.1). Reported variants represent identical protein (NP_387506.1; NP_004623.1; NP_001186778.1).

Immunogen

Peptide with sequence NPSLLERHCAYL, from the C Terminus of the protein sequence according to NP_387506.1; NP_001186779.1; NP_001186780.1.

Please note the peptide is available for sale.

Purification and Storage

Purified from goat serum by ammonium sulphate precipitation followed by antigen affinity chromatography using the immunizing peptide.

Supplied at 0.5 mg/ml in Tris saline, 0.02% sodium azide, pH7.3 with 0.5% bovine serum albumin.

Aliquot and store at -20°C. Minimize freezing and thawing.

Applications Tested

Peptide ELISA: antibody detection limit dilution 1:16000.

Western blot: Approx 40kDa band observed in lysates of cell line HeLa (calculated MW of 45.6kDa according to NP_387506.1). The band fades in HeLa after si-RNA-mediated knock-down (including the fainter shoulder band at 37kDa). Recommended concentration: $1-3\mu$ g/ml.

IHC: Paraffin embedded Human Tonsil and Brain (Cortex). Recommended concentration: 2.5µg/ml.

Immunocytochemistry: Mitochondrial staining of HeLa after in situ denaturation of the target protein using 3-4M guanidinium thiocyanate (patent pending). The subcellular localisation is in agreement with Morgan et al., Biochem Biophys Res Commun. 2001 Jan 12;280(1):177-81.(PMID: 11162496) and Harada et al., Apoptosis. 2010 Oct;15(10):1247-55.(PMID: 20563667). The signal goes down upon knockdown of the

DAP3 expression.Recommended concentration: 3-10µg/ml. **Immunofluorescence:** Protein expression clearly seen within the nucleus. Recommended concentration: 5µg/ml.

Species Reactivity

Tested: Human Expected from sequence similarity: Human EB05427 staining (3µg/ml) of HeLa lysate (RIPA buffer, 30µg total protein per lane). Primary incubated for 1 hour. Detected by western blot using chemiluminescence.

EB05427 (1µg/ml) staining of HeLa lysate (control in left lane and after si-RNA-mediated DAP3 knock-down expresson in right lane) (35µg protein in RIPA buffer). Level of knock-down relative to Actin expression level was determined by RT-PCR. Primary incubation was 1 hour. Detected by chemiluminescence.

EB05427 (2.5µg/ml) staining of paraffin embedded Human Tonsil. Steamed antigen retrieval with citrate buffer pH 6, AP-staining.

EB05427 (2.5µg/ml) staining of paraffin embedded Human Cortex. Steamed antigen retrieval with citrate buffer pH 6, AP-staining.

EB05427 (0.5ug/ml) staining of guanidinium thiocyanate-treated HeLa before (left) and after (right) si-RNA-mediated DAP3 knock-down expression. Primary incubation 1h at ambient temp. Detection by DyLight 488. Nuclear DAPI stain.

Immunofluorescence staining of MCF7 cells with 5ug/ml EB05427 antibody. Detected with Rabbit anti-goat IgG-Alexafluor488 antibody at 1:1000.