

#### **UK Office**

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# 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.