

## **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

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# EB06268-T - Goat Anti-VPS35 / MEM3 Antibody -

**Trial** Size: 20µg specific antibody in 40µl



## **Target Protein**

Principal Names: VPS35, MEM3, FLJ10752, FLJ13588, FLJ20388, DKFZp434E1211, DKFZp434P1672, vacuolar protein sorting 35 (yeast), maternal-embryonic 3, vacuolar protein sorting 35 homolog (S. cerevisiae), vacuolar protein sorting 35 Official Symbol: VPS35 Accession Number(s): NP\_060676.2 Human GenelD(s): 55737 Non-Human GenelD(s): 65114 (mouse) Important Comments: Note there is a hypothetical protein called similar to vacuolar protein sorting 35 (XP\_040192.1), which is virtually identical.

#### Immunogen

Peptide with sequence C-SPESEGPIYEGLIL, from the C Terminus of the protein sequence according to NP\_060676.2.

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 1:128000.

**Western blot:** Approx 90kDa band observed in Human (Cerebellum), Mouse and Rat Brain lysates and in lysates of cell line HepG2, and approx. 85kDa observed in lysates of cell line HEK293 (calculated MW of 91.7kDa according to Human NP\_060676.2, Mouse NP\_075373.1). Recommended concentration: 0.03-0.1µg/ml. Primary incubation 1 hour at room temperature.

IHC: Paraffin embedded Human Prostate. Recommended concentration: 8µg/ml.

**Immunofluorescence:** Strong expression of the protein seen in the cytoplasm/vesicles of HEK293 and U2OS cells. Recommended concentration: 10µg/ml.

## **Species Reactivity**

Tested: Human, Mouse, Rat Expected from sequence similarity: Human, Mouse, Rat, Cow

## **Specific References**

This antibody has been successfully used in ICC and In situ PLA on Human: Anne Mette G Jensen, Yu Kitago, Elnaz Fazeli, Christian B Vægter, Scott A Small, Gregory A Petsko, Olav M Andersen Dimerization of the Alzheimer's disease pathogenic receptor SORLA regulates its association with retromer Proc Natl Acad Sci U S A. 2023 Jan 24;120(4):e2212180120. PMID: 36652482

## This antibody (previous batch) has been successfully used in the following paper:

Krzysztof Sikorski, Adi Mehta, Marit Inngjerdingen, Flourina Thakor, Simon Kling, Tomas Kalina, Tuula A. Nyman, Maria Ekman Stensland, Wei Zhou, Gustavo A. De Souza, Lars Holden, Jan Stuchly, Markus Templin and Fridtjof Lund-Johansen A high-throughput pipeline for validation of antibodies

Nat Methods. 2018 Nov;15(11):909-912

PMID: 30377371

#### This antibody (previous batch) has been successfully used in ICC on Human:

Kanatsu K, Hori Y, Ebinuma I, Chiu YW, Tomita T.

Retrograde transport of  $\gamma$ -secretase from endosomes to the trans-Golgi network regulates A $\beta$ 42 production.

J Neurochem. 2018 May 31.

PMID: 29851073

#### This antibody (previous batch) has been successfully used in IF on Human:

Takatori S, Tatematsu T, Cheng J, Matsumoto J, Akano T, Fujimoto T.

Phosphatidylinositol 3,5-Bisphosphate-Rich Membrane Domains in Endosomes and Lysosomes

Traffic. 2016 Feb;17(2):154-67 PMID: 26563567

#### This antibody (previous batch) has been successfully used in IF on CHO cells:

Lee S, Uchida Y, Emoto K, Umeda M, Kuge O, Taguchi T, Arai H.

Impaired retrograde membrane traffic through endosomes in a mutant CHO cell defective in phosphatidylserine synthesis.

Genes Cells. 2012 Aug;17(8):728-36. doi: 10.1111/j.1365-2443.2012.01622.x. PMID: 22747682

EB06268 (0.03µg/ml) staining of Human (A) Mouse (B) and Rat (C) Brain lysate (35µg protein in RIPA buffer). Detected by chemiluminescence.

EB06268 (0.03µg/ml) staining of HepG2 (A) and HEK293 (B) cell lysate (35µg protein in RIPA buffer). Detected by chemiluminescence.

EB06268 Immunofluorescence analysis of paraformaldehyde fixed HEK293 cells, permeabilized with 0.15% Triton. Primary incubation 1hr (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml), showing cytoplasmic/vesicle staining. The nuclear stain is DAPI (blue). Negative control: Unimmunized goat IgG (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml).

EB06268 Immunofluorescence analysis of paraformaldehyde fixed U2OS cells, permeabilized with 0.15% Triton. Primary incubation 1hr (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml), showing cytoplasmic/vesicle staining. The nuclear stain is DAPI (blue). Negative control: Unimmunized goat IgG (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml).

EB06268 (8µg/ml) staining of paraffin embedded Human Prostate. Heat induced antigen retrieval with citrate buffer pH 6, HRP-staining.

EB06268 Negative Control showing staining of paraffin embedded Human Prostate, with no primary antibody.