

International Office

Everest Biotech Ltd Vector Laboratories, Inc. 6737 Mowry Ave Newark, CA 94560 United States

Customer Service: <u>customerservice@vectorlabs.com</u> Technical Service: <u>technical@vectorlabs.com</u>

Tel: +1 (800) 227-6666

www.everestbiotech.com

Research Use Only. Not for diagnostic or therapeutic use.

EB06268 - Goat Anti-VPS35 / MEM3 Antibody

Size: 100µg specific antibody in 200µl



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): <u>55737</u> 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 γ -secretase from endosomes to the trans-Golgi network regulates A β 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

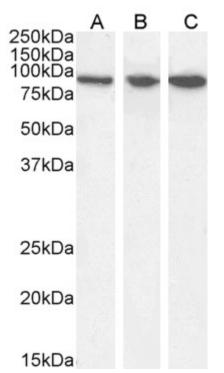
Traffic. 2016 Feb;17(2):154-67 PMID: 26563567

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

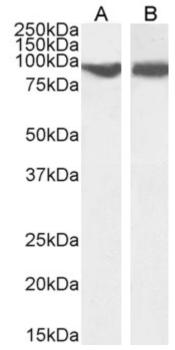
Lee S, Uchida Y, Emoto K, Umeda M, Kuge O, Taguchi T, Arai H.

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

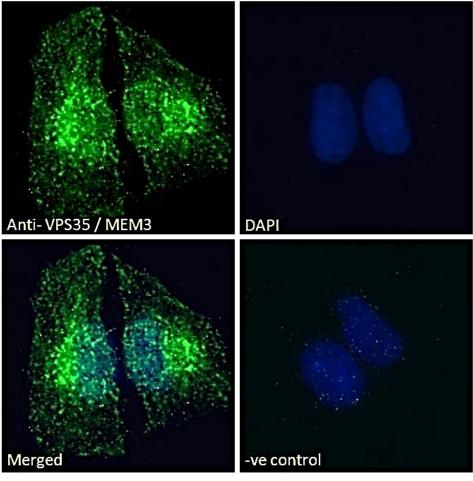
Genes Cells. 2012 Aug;17(8):728-36. doi: 10.1111/j.1365-2443.2012.01622.x. PMID: 22747682



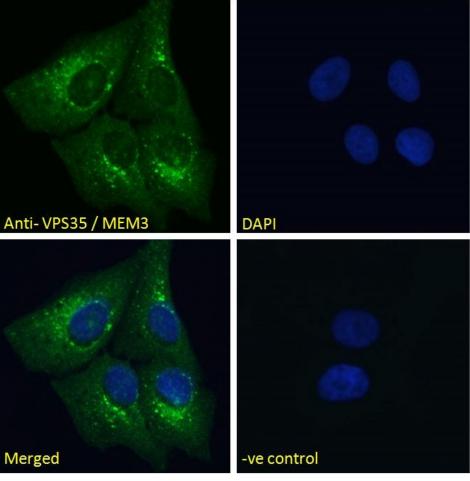
EB06268 (0.03µg/ml) staining of Human (A) Mouse (B) and Rat (C) Brain lysate (35µg protein in RIPA buffer). Detected by chemiluminescence.



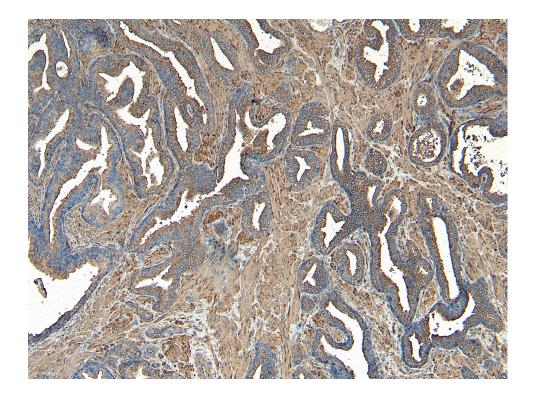
EB06268 (0.03µg/ml) staining of HepG2 (A) and HEK293 (B) cell lysate (35µg protein in RIPA buffer). Detected by chemiluminescence.



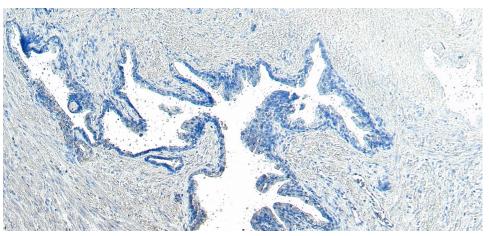
EB06268 Immunofluorescence analysis of paraformaldehyde fixed HEK293 cells, permeabilized with 0.15% Triton. Primary incubation 1hr (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml), showing cytoplasmic/vesicle staining. The nuclear stain is DAPI (blue). Negative control: Unimmunized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