

DESCRIPTION

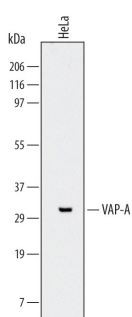
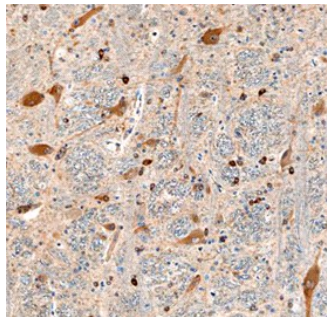
Species Reactivity	Human
Specificity	Detects human VAP-A in direct ELISAs and Western blots. In direct ELISAs, no cross-reactivity with recombinant human VAP-B or recombinant rat VAP-B is observed.
Source	Monoclonal Mouse IgG _{2A} Clone # 604101
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant human VAP-A Ala2-Met132 Accession # Q9P0L0
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	2 µg/mL	See Below
Immunohistochemistry	8-25 µg/mL	See Below

DATA

<p>Western Blot</p>  <p>Detection of Human VAP-A by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line. PVDF Membrane was probed with 2 µg/mL of Mouse Anti-Human VAP-A Monoclonal Antibody (Catalog # MAB5820) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007). A specific band was detected for VAP-A at approximately 33 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1 with 0.05% Tween 20.</p>	<p>Immunohistochemistry</p>  <p>VAP-A in Human Brain. VAP-A was detected in immersion fixed paraffin-embedded sections of human brain using Mouse Anti-Human VAP-A Monoclonal Antibody (Catalog # MAB5820) at 15 µg/mL overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using the Anti-Mouse HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS002) and counterstained with hematoxylin (blue). Specific staining was localized to neuronal cell bodies and processes. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.</p>
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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Vesicle-associated membrane protein (VAMP)-associated protein A (VAP-A; also VAMP-A and VAP-33) is a 33 kDa, ubiquitously expressed, type IV transmembrane protein belonging to the VAP family of proteins (1). It is found in plasma and ER membranes as well as in intracellular vesicles as a homodimer and a heterodimer with VAP-B. Human VAP-A is synthesized as a 249 amino acid (aa) precursor that contains a 227 aa cytoplasmic domain and a 21 aa transmembrane region. The cytoplasmic domain contains a mobile sperm protein (MSP) domain (aa 13-131) and a coiled-coil region (aa 169-205). Human VAP-A is 97% aa identical to mouse and rat VAP-A. VAP-A and VAP-B recruit FFAT (two phenylalanines in an acidic tract)-motif-containing proteins to the cytosolic surface of ER membranes through a conserved region within their MSP domain, and they have been implicated in regulation of membrane transport, phospholipid biosynthesis, and the unfolded protein response (2, 3). Their ability to interact with lipid-transfer/binding proteins (LT/BPs) may affect the lipid composition of certain cellular membranes (2, 4). VAPs play a critical role in maintaining the structural and functional properties of the Golgi complex (2). Knockdown of VAP reduces the levels of phosphatidylinositol-4-phosphate (PI4P), diacylglycerol (DAG), and sphingomyelin (SM) in Golgi membranes and exports pleiotropic effects in Golgi-mediated transport (2). The effects of VAPs are mediated by their interacting FFAT-motif-containing proteins Nir2, OSBP, and CERT (2). VAPs provide a scaffold for these LT/BPs at the ER-Golgi membrane contact sites, thereby affecting the lipid composition of the Golgi membranes and consequently their structural and functional identities (2). VAP-A associates with and regulates the neurite outgrowth-promoting activity of protrudin, a protein that promotes neurite formation (5).

References:

1. Weir, M.L. *et al.* (1998) *Biochem. J.* **333**:247.
2. Peretti, D. *et al.* (2008) *Mol. Biol. Cell* **19**:3871.
3. Kaiser, S.E. *et al.* (2005) *Structure* **13**:1035.
4. Loewen, C.J. *et al.* (2003) *EMBO J.* **22**:2025.
5. Saita, S. *et al.* (2009) *J. Biol. Chem.* **284**:13766.