

Anti-VAMP8 Antibody [JF0963]

ET1702-10



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 11 kDa
Clone number:	JF0963

Description: Vesicle-associated membrane protein 8 is a protein that in humans is encoded by the VAMP8 gene. Synaptobrevins/VAMPs, syntaxins, and the 25-kD synaptosomal-associated protein SNAP25 are the main components of a protein complex involved in the docking and/or fusion of synaptic vesicles with the presynaptic membrane. The protein encoded by this gene is a member of the vesicle-associated membrane protein (VAMP)/synaptobrevin family. It is associated with the perinuclear vesicular structures of the early endocytic compartment. It has been found that VAMP8 interacts specifically with the soluble NSF-attachment protein (alpha-SNAP), most likely through an VAMP8-containing SNARE complex. Phosphorylation of VAMP8 inside the conserved SNARE-domain can suppress vesicle fusion.

Immunogen: Synthetic peptide within Human VAMP8 aa1-29 / 100.

Positive control: Hela cell lysate, mouse kidney tissue lysate, U937 cell lysates, SW480, 293T, Hela, human kidney tissue, mouse kidney tissue, rat kidney tissue.

Subcellular location: Lysosome membrane, Early endosome membrane, Late endosome membrane, Cell membrane.

Database links: SwissProt: Q9BV40 Human | O70404 Mouse | Q9WUF4 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IF-Cell	1:50-1:100
IF-Tissue	1:1,000
IHC-P	1:5,000
FC	1:1,000
IP	Use at an assay dependent concentration.

Storage Buffer: 1*PBS (pH7.4), 0.1% BSA, 40% Glycerol, 0.2% Proclean 950.

Storage Instruction: Shipped at 4°C. Store at +4°C short term (1-2 weeks). Store at -20°C long term.

Purity: Protein A affinity purified.

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Orders: 0086-571-88062880

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Applications: WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

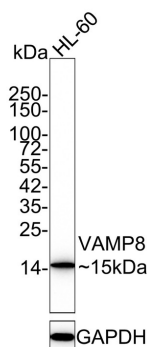


Fig1: Western blot analysis of VAMP8 on HL-60 cell/tissue lysates with Rabbit anti-VAMP8 antibody (ET1702-10) at 1/2,000 dilution.

Lysates/proteins at 20 µg/Lane.

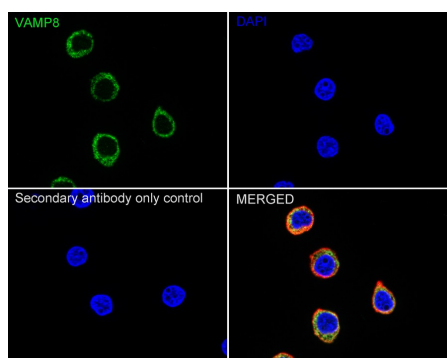
Predicted band size: 11 kDa

Observed band size: 15 kDa

Exposure time: 1 minutes 55 seconds; ECL: K1801;
4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1702-10) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

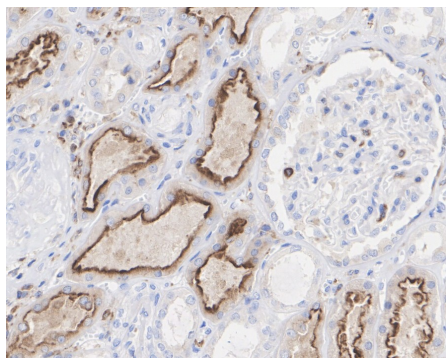
Fig2: Immunocytochemistry analysis of HL-60 cells labeling VAMP8 with Rabbit anti-VAMP8 antibody (ET1702-10) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-VAMP8 antibody (ET1702-10) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Fig3: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-VAMP8 antibody (ET1702-10) at 1/5,000 dilution.



The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1702-10) at 1/5,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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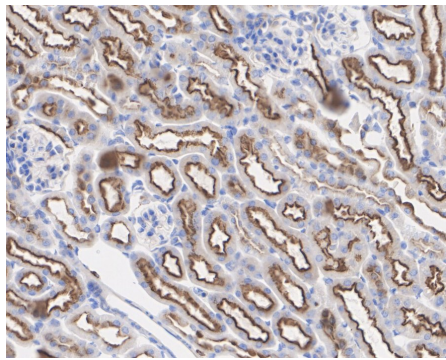


Fig4: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-VAMP8 antibody (ET1702-10) at 1/5,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1702-10) at 1/5,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

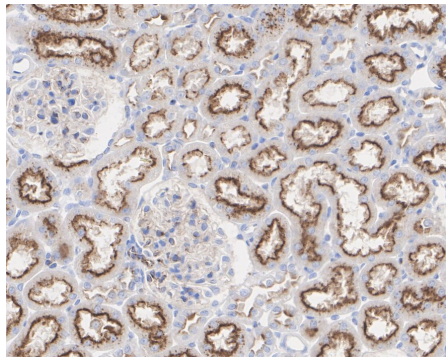


Fig5: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-VAMP8 antibody (ET1702-10) at 1/5,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1702-10) at 1/5,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

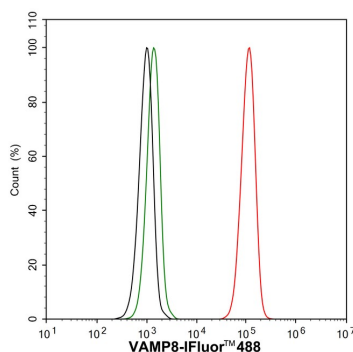


Fig6: Flow cytometric analysis of HL-60 cells labeling VAMP8.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1702-10, 1µg/mL) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Fig7: Western blot analysis of VAMP8 on different lysates with Rabbit anti-VAMP8 antibody (ET1702-10) at 1/2,000 dilution.

Lane 1: Mouse kidney tissue lysate

Lysates/proteins at 20 µg/Lane.

Exposure time: 41 seconds; ECL: K1801

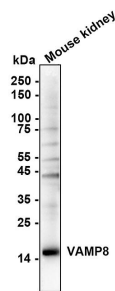
Blocking: 5% NFDM/TBST, 1 hour at room temperature

Primary antibody: ET1702-10, 1/2,000 in primary antibody dilution buffer (K1803), overnight at 4 °C

Secondary antibody: Goat anti-Rabbit IgG-HRP (HA1001), 1/50,000 in 5% NFDM/TBST, 1 hour at room temperature

Predicted band size: 11 kDa

Observed band size: 15 kDa



Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

1. Xie X et al. Deep vein thrombosis is accurately predicted by comprehensive analysis of the levels of microRNA-96 and plasma D-dimer. *Exp Ther Med* 12:1896-1900 (2016).
2. Pirooz SD et al. UVRAG is required for virus entry through combinatorial interaction with the class C-Vps complex and SNAREs. *Proc Natl Acad Sci U S A* 111:2716-21 (2014).

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