

#### VPS35 Antibody

VPS35 Antibody, Clone 5A9 Catalog # ASM10692

### Specification

# **VPS35 Antibody - Product Information**

Primary Accession Other Accession Host Clonality **Target/Specificity** VPS35

**0960K1** NP 060676.2 Mouse Monoclonal

**Other Names** 

Vacuolar protein sorting-associated protein 35, MEM3, PARK17, VPS35 retromer complex component, maternal-embryonic 3, vesicle protein sortin 35, TCCCTA00141, FLJ10752

Immunogen Full length recombinant human VSP35

**Purification** Protein G Purified

Storage -20ºC **Storage Buffer** PBS pH 7.4, 50% glycerol, 0.09% Sodium azide \*Storage buffer may change when conjugated

Shipping Temperature Blue Ice or 4°C **Certificate of Analysis** A 1:1000 dilution of SMC-603 was sufficient for detection of VPS35 in 10 µg of SH-SY5Y by ECL immunoblot analysis using Goat Anti-Mouse IgG:HRP as the secondary antibody.

**Cellular Localization** Endosome | Lysosome | Vesicles | Cytoplasm | Membrane

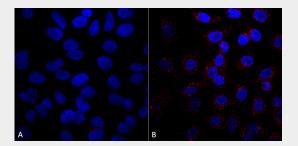
## **VPS35 Antibody - Protocols**

Provided below are standard protocols that you may find useful for product applications.

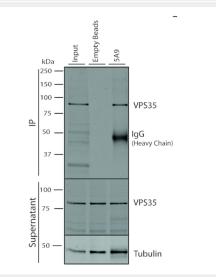
- Western Blot
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

VPS35 Antibody - Images

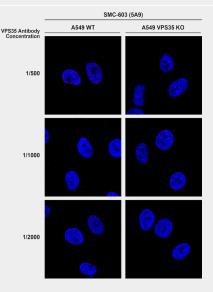




Immunocytochemistry/Immunofluorescence analysis using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Tissue: A549 cells. Species: Human. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692) at 1:5 (tissue culture supernatant). Secondary Antibody: Donkey anti-mouse: Alexa Fluor 594 at 1:4000 in 0.2% BSA PBS. Counterstain: DAPI. Localization: Vesicles. A) VPS35 KO A549 cells B) WT A549 cells. Courtesy of: Dario Alessi Lab, University of Dundee.

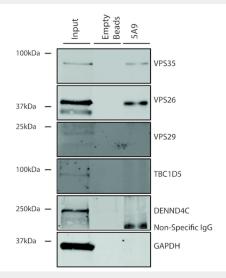


Immunoprecipitation analysis using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Tissue: A549 cells. Species: Human. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692). 500  $\mu$ L cell culture supernatants were incubated with 10  $\mu$ L of Protein A/G resin beads for 1 hour at 4°C. ASM10692 clone 5A9 depletes VPS35 from the A549 cell extract.

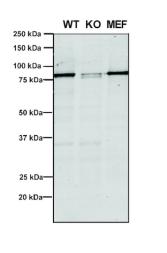




Immunocytochemistry/Immunofluorescence analysis using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Tissue: A549 WT, VPS35 KO cells. Species: Human. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692). Secondary Antibody: Donkey Anti-Mouse AlexaFluor 594. Clone can detect VPS35 at 1/2000 concentration.

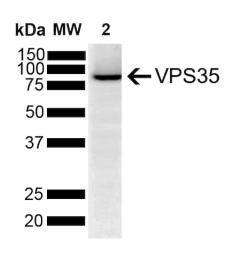


Immunoprecipitation analysis using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Tissue: A549 cells. Species: Human. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692).

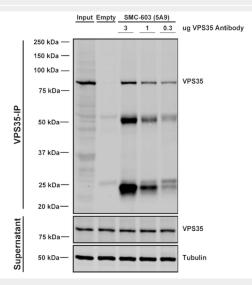


Western Blot analysis of Human, Mouse A549, MEF showing detection of VPS35 protein using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Lane 1: Molecular Weight Ladder. Lane 2: VPS35 KO A549 cells. Lane 3: mouse embryonic fibroblast cells.. Load: 8 µg each A549 and MEF. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692) at 1:5 (tissue culture supernatant). Secondary Antibody: Donkey anti-mouse IRDye 800CW at 1:25000 in TBS-T.



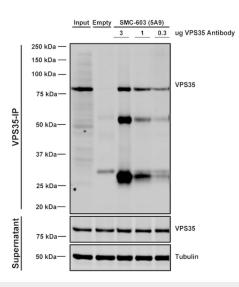


Western Blot analysis of Human SH-SY5Y showing detection of VPS35 protein using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Lane 1: Molecular Weight Ladder. Lane 2: SH-SY5Y (10 ug). Load: 10  $\mu$ g. Block: 5% Skim Milk powder in TBST. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692) at 1:1000 for 2 hours at RT with shaking. Secondary Antibody: Goat anti-mouse IgG:HRP at 1:4000 for 1 hour at RT with shaking. Color Development: Chemiluminescent for HRP (Moss) for 5 min in RT.



Immunoprecipitation analysis using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Tissue: A549 cells. Species: Human. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692). Three amounts of ASM10692 (3, 1 and 0.3 ug) were non-covalently coupled to 10uL of A/G sepharose beads for 1 hour at 4°C and next incubated with 250ug of A549 lysate for 2 hours at 4°C.





Immunoprecipitation analysis using Mouse Anti-VPS35 Monoclonal Antibody, Clone 5A9 (ASM10692). Tissue: embryonic fibroblast. Species: Mouse. Primary Antibody: Mouse Anti-VPS35 Monoclonal Antibody (ASM10692). Three amounts of ASM10692 (3, 1 and 0.3 ug) were non-covalently coupled to 10uL of A/G sepharose beads for 1 hour at 4°C and next incubated with 250ug of MEF lysate for 2 hours at 4°C.

### VPS35 Antibody - Background

Vacuolar Protein Sorter-35 (VPS35) is a component of the retromer complex, which is essential for endosome-to-Golgi retrieval of membrane proteins. VPS35 mutations such as D620N have been linked to Parkinson's Disease (PD) (1,2) and affect retromer function, protein homeostasis, and mitochondria (3).

## VPS35 Antibody - References

- 1. Vilarino-Guell, C. et al. (2011) Am J Hum Genet 89:162-167
- 2. Zimprich, A. et al. (2011) Am J Hum Genet 89:168–175
- 3. Rahman, A.A., Morrison, B.E. (2019) Neurosci 401:1-10.