

# Eap45 (T-20): sc-79931

## BACKGROUND

Vacuolar sorting proteins (VPSs) are required for trafficking normal endocytic and biosynthetic proteins to the vacuole and also play an important role in the budding process of cells. The ESCRT-II (endosomal sorting complex required for transport II) complex, which is involved in endocytosis of ubiquitinated membrane proteins, is formed by Eap45, EAP30, and VPS25. These vacuolar sorting proteins are also involved in a multiprotein complex with RNA polymerase II elongation factor (ELL). Eap45 (ELL-associated protein of 45 kDa), also known as vacuolar protein-sorting-associated protein 36, is a 386 amino acid protein containing a GLUE (GRAM-like ubiquitin-binding in Eap45) domain that mediates the binding to ubiquitin and phosphoinositides. Since Eap45 colocalizes with ubiquitinated proteins on late endosomes, it is likely that Eap45 plays a role in the endosomal sorting of ubiquitinated cargo. There are two isoforms of Eap45 that exist as a result of alternative splicing events.

## REFERENCES

- Martin-Serrano, J., et al. 2003. Divergent retroviral late-budding domains recruit vacuolar protein sorting factors by using alternative adaptor proteins. *Proc. Natl. Acad. Sci. USA* 100: 12414-12419.
- Slagsvold, T., et al. 2005. Eap45 in mammalian ESCRT-II binds ubiquitin via a phosphoinositide-interacting GLUE domain. *J. Biol. Chem.* 280: 19600-19606.
- Teo, H., et al. 2006. ESCRT-I core and ESCRT-II GLUE domain structures reveal role for GLUE in linking to ESCRT-I and membranes. *Cell* 125: 99-111.
- Langelier, C., et al. 2006. Human ESCRT-II complex and its role in human immunodeficiency virus type 1 release. *J. Virol.* 80: 9465-9480.
- Alam, S.L., et al. 2006. Structural basis for ubiquitin recognition by the human ESCRT-II Eap45 GLUE domain. *Nat. Struct. Mol. Biol.* 13: 1029-1030.
- Hirano, S., et al. 2006. Structural basis of ubiquitin recognition by mammalian Eap45 GLUE domain. *Nat. Struct. Mol. Biol.* 13: 1031-1032.

## CHROMOSOMAL LOCATION

Genetic locus: VPS36 (human) mapping to 13q14.3; Vps36 (mouse) mapping to 8 A2.

## SOURCE

Eap45 (T-20) is an affinity purified rabbit polyclonal antibody raised against a peptide mapping within an internal region of Eap45 of human origin.

## PRODUCT

Each vial contains 100 µg IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-79931 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

## STORAGE

Store at 4° C, **\*\*DO NOT FREEZE\*\***. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

## APPLICATIONS

Eap45 (T-20) is recommended for detection of Eap45 of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Eap45 (T-20) is also recommended for detection of Eap45 in additional species, including equine, canine, bovine, porcine and avian.

Suitable for use as control antibody for Eap45 siRNA (h): sc-77215, Eap45 siRNA (m): sc-77216, Eap45 shRNA Plasmid (h): sc-77215-SH, Eap45 shRNA Plasmid (m): sc-77216-SH, Eap45 shRNA (h) Lentiviral Particles: sc-77215-V and Eap45 shRNA (m) Lentiviral Particles: sc-77216-V.

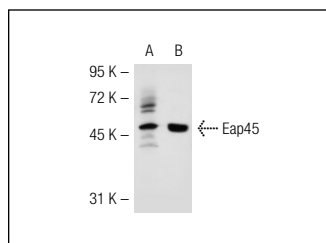
Molecular Weight of Eap45: 45 kDa.

Positive Controls: mouse ovary extract: sc-2404 or mouse embryo extract: sc-364239.

## RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## DATA



Eap45 (T-20): sc-79931. Western blot analysis of Eap45 expression in mouse embryo (A) and mouse ovary (B) tissue extracts.

## SELECT PRODUCT CITATIONS

- Stieler, J.T. and Prange, R. 2014. Involvement of ESCRT-II in hepatitis B virus morphogenesis. *PLoS ONE* 9: e91279.

## RESEARCH USE

For research use only, not for use in diagnostic procedures.