abcam

Product datasheet

Anti-TSG101 antibody [EPR7130(B)] ab125011

Recombinant RabMAb

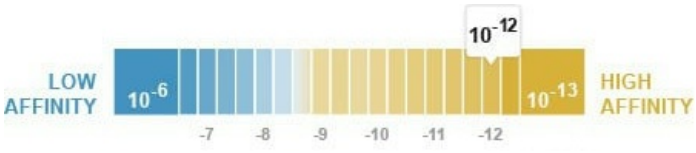
10 Abreviews 426 References 10 Images

Overview

Product name	Anti-TSG101 antibody [EPR7130(B)]
Description	Rabbit monoclonal [EPR7130(B)] to TSG101
Host species	Rabbit
Tested applications	**Suitable for:** WB, Flow Cyt (Intra), ICC/IF, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human TSG101 aa 1-100. The exact sequence is proprietary.
Positive control	WB: Mouse heart tissue lysates. K-562, Jurkat, SH-SY5Y, NIH/3T3 and PC-12 whole cell lysate. Mouse and rat heart and brain lysate. Flow Cyt: HeLa cells. ICC/IF: Jurkat cells and HeLa cells. IHC-P: Rat kidney tissue, mouse kidney tissue, human thyroid cancer tissue, human kidney tissue.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Dissociation constant (K_D)	$K_D = 4.00 \times 10^{-12}$ M



Learn more about K_D

Storage buffer	pH: 7.20

	Preservative: 0.01% Sodium azide Constituents: 40% Glycerol, 0.05% BSA, 59% PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR7130(B)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab125011 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

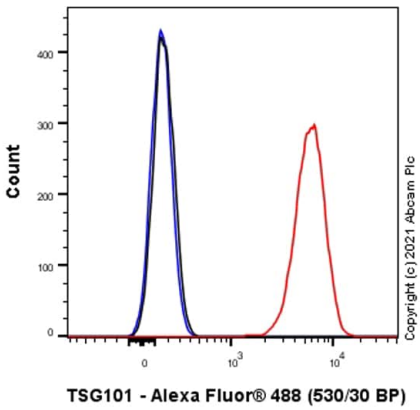
Application	Abreviews	Notes
WB	★★★★☆ (7)	1/1000 - 1/10000. Detects a band of approximately 47, 52 kDa (predicted molecular weight: 44 kDa).
Flow Cyt (Intra)		1/20. For unpurified use at 1/3. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF	★★★★★ (3)	Use a concentration of 1 µg/ml. **For unpurified use at 1/10.** **This product gave a positive signal in HeLa fixed with 100% methanol (5 min).**
IHC-P		1/300. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. **For unpurified use at 1/30.**

Target

Function	Component of the ESCRT-I complex, a regulator of vesicular trafficking process. Binds to ubiquitinated cargo proteins and is required for the sorting of endocytic ubiquitinated cargos into multivesicular bodies (MVBs). Mediates the association between the ESCRT-0 and ESCRT-I complex. Required for completion of cytokinesis; the function requires CEP55. May be involved in cell growth and differentiation. Acts as a negative growth regulator. Involved in the budding of many viruses through an interaction with viral proteins that contain a late-budding motif P-[ST]-A-P. This interaction is essential for viral particle budding of numerous retroviruses.
Tissue specificity	Heart, brain, placenta, lung, liver, skeletal, kidney and pancreas.
Sequence similarities	Belongs to the ubiquitin-conjugating enzyme family. UEV subfamily. Contains 1 SB (steadiness box) domain. Contains 1 UEV (ubiquitin E2 variant) domain.
Domain	The UEV domain is required for the interaction of the complex with ubiquitin. It also mediates the interaction with PTAP/PSAP motifs of HIV-1 P6 protein and human spumaretrovirus Gag protein. The coiled coil domain may interact with stathmin.

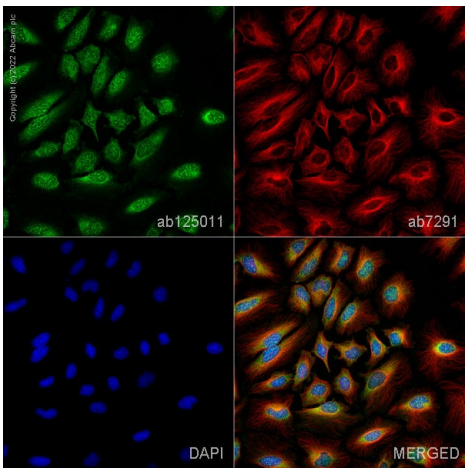
	The UEV domain binds ubiquitin and P-[ST]-A-P peptide motif independently.
Post-translational modifications	Monoubiquitinated at multiple sites by LRSAM1 and by MGRN1. Ubiquitination inactivates it, possibly by regulating its shuttling between an active membrane-bound protein and an inactive soluble form. Ubiquitination by MGRN1 requires the presence of UBE2D1.
Cellular localization	Cytoplasm. Membrane. Nucleus. Late endosome membrane. Mainly cytoplasmic. Membrane-associated when active and soluble when inactive. Depending on the stage of the cell cycle, detected in the nucleus. Colocalized with CEP55 in the midbody during cytokinesis.

Images



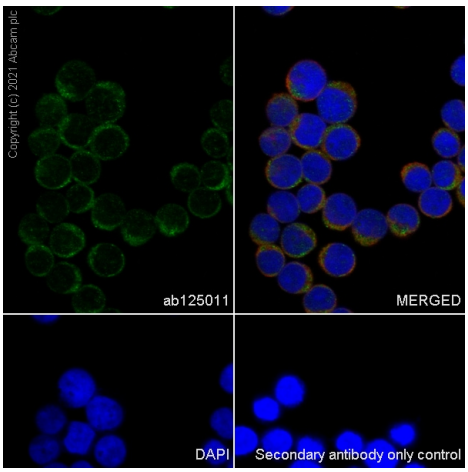
Flow Cytometry (Intracellular) - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling TSG101 with Purified ab125011 at 1:20 dilution (10 µg/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1:2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



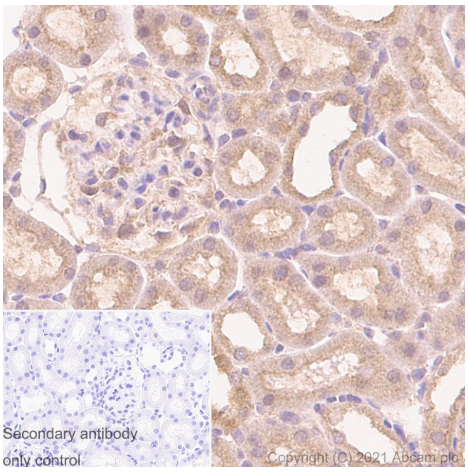
Immunocytochemistry/ Immunofluorescence - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

ab125011 staining TSG101 in HeLa cells. The cells were fixed with 100% methanol (5 min), permeabilised with 0.1% Triton x-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab125011 at 1 µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 µg/ml. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150119**, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



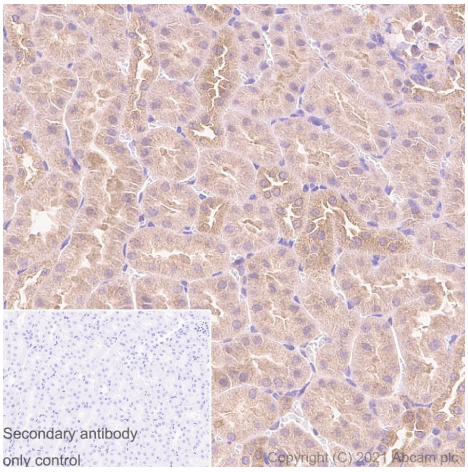
Immunocytochemistry/ Immunofluorescence - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

Immunocytochemistry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling TSG101 with Purified ab125011 at 1:50 dilution (4.5 µg/ml). Cells were fixed in 100% Methanol and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



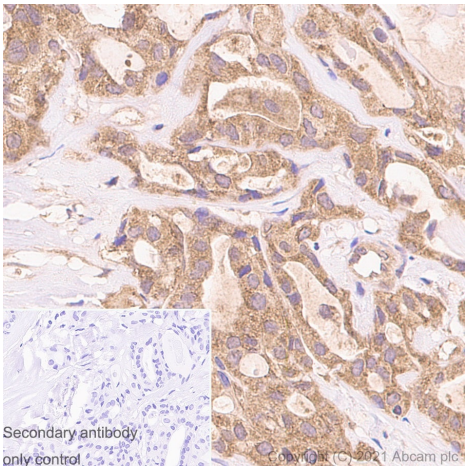
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat kidney tissue sections labeling TSG101 with Purified ab125011 at 1:300 dilution (0.73 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



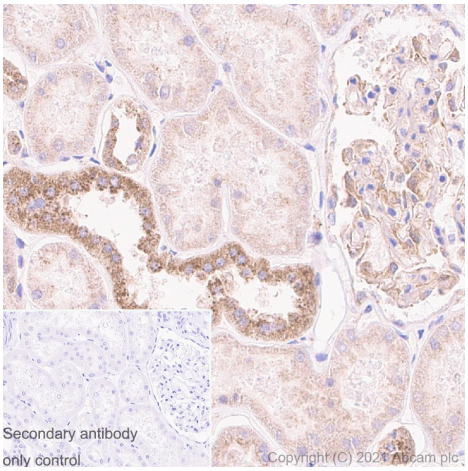
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue sections labeling TSG101 with Purified ab125011 at 1:300 dilution (0.73 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



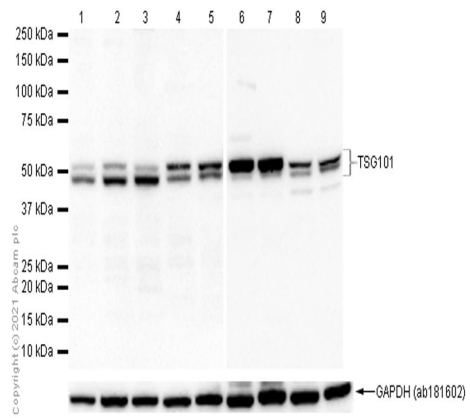
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human thyroid cancer tissue sections labeling TSG101 with Purified ab125011 at 1:300 dilution (0.73 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human kidney tissue sections labeling TSG101 with Purified ab125011 at 1:300 dilution (0.73 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



Western blot - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

All lanes : Anti-TSG101 antibody [EPR7130(B)] (ab125011) at 1/1000 dilution

Lane 1 : K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysate
Lane 2 : Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate
Lane 3 : SH-SY5Y (Human neuroblastoma epithelial cell) whole cell lysate
Lane 4 : NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate
Lane 5 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysate
Lane 6 : Mouse heart lysate
Lane 7 : Rat heart lysate
Lane 8 : Mouse brain lysate
Lane 9 : Rat brain lysate

Lysates/proteins at 20 µg per lane.

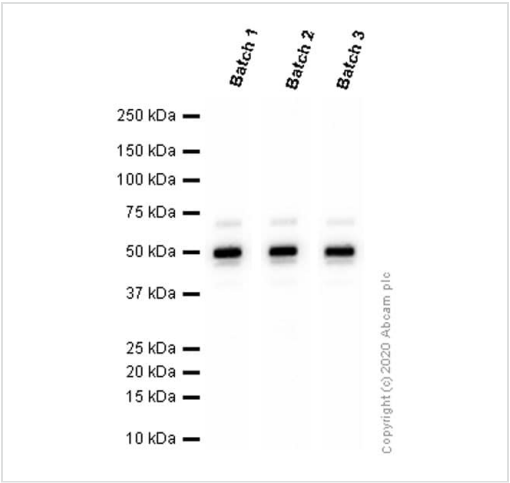
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 44 kDa
Observed band size: 47, 52 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

The doublet are consistent with what have been described in the literature (PMID: 27882925, 31684046, 32932791, 11427703).

Image produced using the purified version.



Western blot - Anti-TSG101 antibody [EPR7130(B)] (ab125011)

Different batches of ab125011 were tested on Mouse heart lysate at 1.0 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 45 kDa.

This data was generarted using the unpurified format.

Anti-TSG101 antibody [EPR7130(B)] (ab125011)

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