abcam

Product datasheet

Anti-TSG101 antibody [EPR7130(B)] - Low endotoxin, Azide free ab216447



8 Images

Overview

Product name Anti-TSG101 antibody [EPR7130(B)] - Low endotoxin, Azide free

Description Rabbit monoclonal [EPR7130(B)] to TSG101 - Low endotoxin, Azide free

Host species Rabbit

Tested applications Suitable for: WB, ICC/IF, IHC-P, Flow Cyt (Intra)

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control Flow cyt: HeLa cells. ICC/IF: Jurkat cells and HeLa cells. IHC: rat kidney tissue, mouse kidney

tissue, human thyroid cancer tissue, human kidney tissue.

General notes ab216447 is the carrier-free version of ab125011.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar[®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.

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**.

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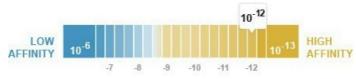
Our <u>Low endotoxin, azide-free formats</u> have low endotoxin level (≤ 1 EU/ml, determined by the LAL assay) and are free from azide, to achieve consistent experimental results in functional assays.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Dissociation constant (K_D) $K_D = 4.00 \times 10^{-12} M$



Learn more about K_D

Storage buffer pH: 7.20

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR7130(B)

Isotype IgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab216447 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		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration. This product gave a positive signal in HeLa fixed with 100% methanol (5 min).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.
Flow Cyt (Intra)		Use at an assay dependent concentration. <u>ab199376</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

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

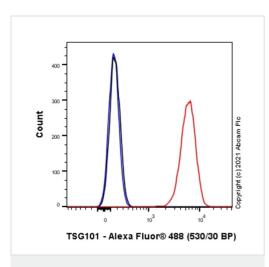
Monoubiquitinated at multiple sites by LRSAM1 and by MGRN1. Ubiquitination inactivates it, possibly by regulating its shuttling between an active membrane-bound protein and an inactive

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.

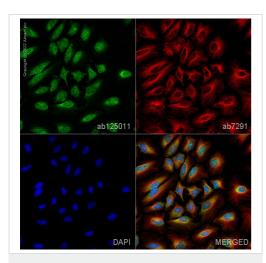
Images



Flow Cytometry (Intracellular) - Anti-TSG101 antibody [EPR7130(B)] - Low endotoxin, Azide free (ab216447)

This data was developed using <u>ab125011</u>, the same antibody clone in a different buffer formulation.

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 lgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1:2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-TSG101 antibody [EPR7130(B)] - Low endotoxin, Azide free (ab216447)

ab125011 MERGED

DAPI

Secondary antibody only control

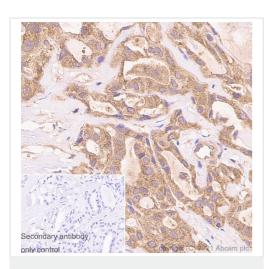
Immunocytochemistry/ Immunofluorescence - Anti-TSG101 antibody [EPR7130(B)] - Low endotoxin, Azide free (ab216447)

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 lgG - H&L (Alexa Fluor[®] 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150119**, Goat polyclonal Secondary Antibody to Mouse lgG - H&L (Alexa Fluor[®] 647), pre-adsorbed at 1/1000 dilution (shown in 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.

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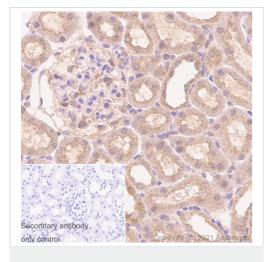
Immunocytochemistry analysis of Jurkat (Human T cell leukemia T lymphocyte) cells labeling TSG101 with Purified $\underline{ab125011}$ at 1:50 dilution (4.5 µg/ml). Cells were fixed in 100% Methanol and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit lgG (Alexa Fluor® 488, $\underline{ab150077}$) 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 paraffinembedded sections) - Anti-TSG101 antibody
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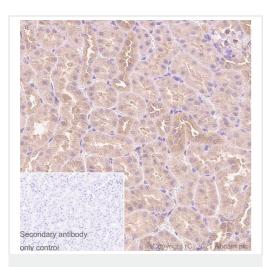
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human thyroid cancer tissue sections labeling TSG101 with Purified <u>ab125011</u> at 1:300 dilution (0.73 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) . Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



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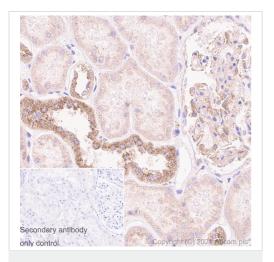
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat kidney tissue sections labeling TSG101 with Purified <u>ab125011</u> at 1:300 dilution (0.73 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution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



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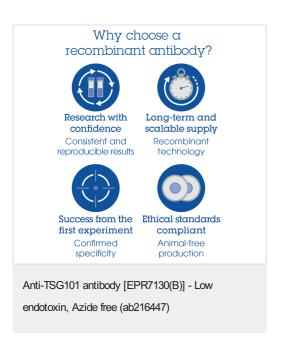
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue sections labeling TSG101 with Purified <u>ab125011</u> at 1:300 dilution (0.73 µg/ml). Heat mediated antigen retrieval was performed using Heat mediated antigen retrieval using Bond™ Epitope Retrieval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



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Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human kidney tissue sections labeling TSG101 with Purified <u>ab125011</u> 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 (<u>ab209101</u>) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



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- We investigate all quality concerns to ensure our products perform to the highest standards

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