

Product datasheet

Anti-TRIM25/EFP antibody [EPR7315] ab167154

KO **VALIDATED** Recombinant RabMAb

★★★★★ [1 Abreviews](#) [26 References](#) [12 Images](#)

Overview

Product name	Anti-TRIM25/EFP antibody [EPR7315]
Description	Rabbit monoclonal [EPR7315] to TRIM25/EFP
Host species	Rabbit
Tested applications	Suitable for: Flow Cyt (Intra), WB, IHC-P, ICC/IF, IP
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide within Human TRIM25/EFP aa 100-200. The exact sequence is proprietary. Database link: Q14258
Positive control	Recombinant Human TRIM25/EFP protein (ab152814) can be used as a positive control in WB. HeLa and K562 cell lysates; Human breast tissue. Human pancreas tissue. Rat brain lysate. SP2/0 (mouse spleen) whole cell lysate.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

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. Stable for 12 months at -20°C.
Storage buffer	Preservative: 0.01% Sodium azide Constituents: 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR7315

Isotype

IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab167154 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
Flow Cyt (Intra)		1/60 - 1/90.
WB	★★★★★ (1)	1/2000 - 1/50000. Predicted molecular weight: 71 kDa.
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/500 - 1/1000.
IP		1/40 - 1/100.

Target

Function

Functions as an ubiquitin E3 ligase and as an ISG15 E3 ligase. Involved in innate immune defense against viruses by mediating ubiquitination of DDX58. Mediates 'Lys-63'-linked polyubiquitination of the DDX58 N-terminal CARD-like region which is crucial for triggering the cytosolic signal transduction that leads to the production of interferons in response to viral infection. Promotes ISGylation of 14-3-3 sigma (SFN), an adapter protein implicated in the regulation of a large spectrum signaling pathway. Mediates estrogen action in various target organs.

Tissue specificity

Ubiquitous.

Pathway

Protein modification; protein ubiquitination.

Sequence similarities

Contains 1 B30.2/SPRY domain.
Contains 1 RING-type zinc finger.

Domain

The RING-type zinc finger is important for ISG15 E3 ligase activity and autoISGylation. AutoISGylation negatively regulates ISG15 E3 ligase activity.
The C-terminal B30.2/SPRY domain interacts with the first N-terminal CARD domain of DDX58.

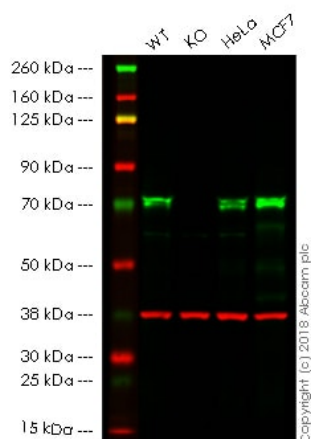
Post-translational modifications

Auto-ISGylated.

Cellular localization

Cytoplasm. Colocalized with DDX58 at cytoplasmic perinuclear bodies.

Images



Western blot - Anti-TRIM25/EFP antibody
[EPR7315] (ab167154)

All lanes : Anti-TRIM25/EFP antibody [EPR7315] (ab167154) at 1/10000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate

Lane 2 : TRIM25/EFP knockout HAP1 whole cell lysate

Lane 3 : HeLa whole cell lysate

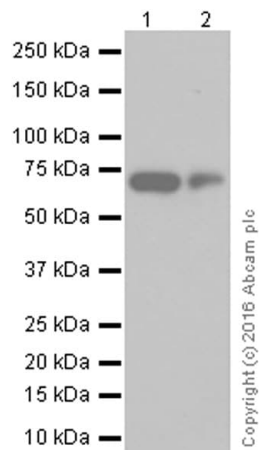
Lane 4 : MCF7 whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 71 kDa

Lanes 1 -4: Merged signal (red and green). Green - ab167154 observed at 71 kDa. Red - loading control, **ab9484**, observed at 37 kDa.

ab167154 was shown to specifically react with TRIM25/EFP in wild-type HAP1 cells as signal was lost in TRIM25/EFP knockout cells. Wild-type and TRIM25/EFP knockout samples were subjected to SDS-PAGE. Ab167154 and **ab9484** (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/10000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-TRIM25/EFP antibody
[EPR7315] (ab167154)

All lanes : Anti-TRIM25/EFP antibody [EPR7315] (ab167154) at 1/50000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 2 : K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate

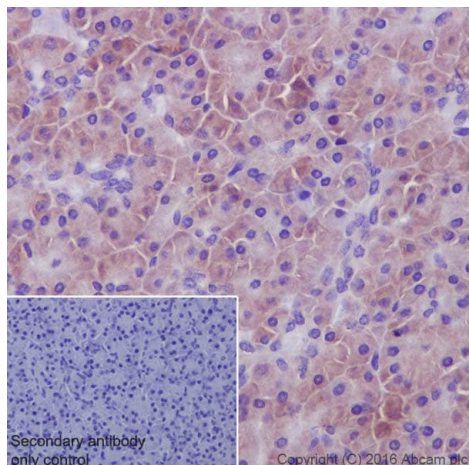
Lysates/proteins at 20 µg per lane.

Secondary

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

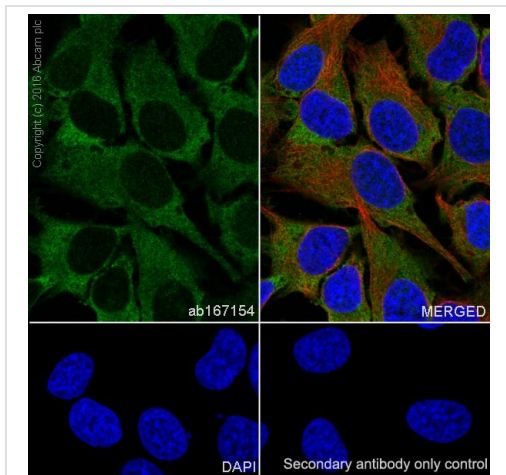
Predicted band size: 71 kDa

Blocking and diluting buffer: 5% NFDM/TBST



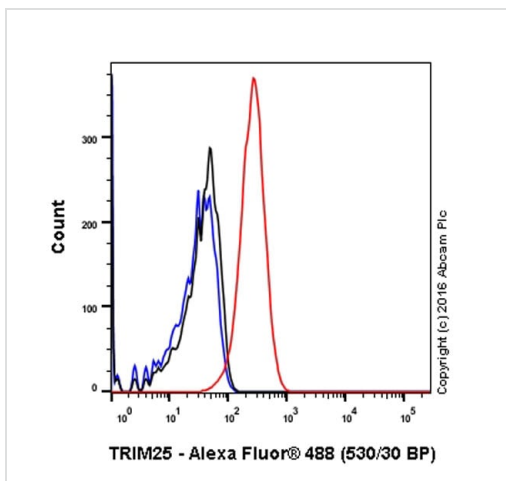
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TRIM25/EFP antibody
[EPR7315] (ab167154)

ab167154 staining TRIM25/EFP in human pancreas tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with paraformaldehyde and antigen retrieval was by heat mediation in a EDTA buffer. Samples were incubated with primary antibody at a dilution of 1/100. A goat anti-rabbit IgG H&L (HRP) [ab97051](#) was used as the secondary antibody at a dilution of 1/500.



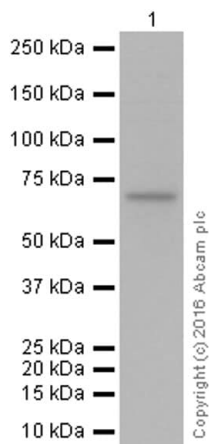
Immunocytochemistry/ Immunofluorescence - Anti-TRIM25/EFP antibody [EPR7315] (ab167154)

ab167154 staining TRIM25/EFP in HeLa (human cervix adenocarcinoma) cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. Samples were incubated with primary antibody at a dilution of 1/500. A goat anti rabbit IgG (Alexa Fluor® 488) ([ab150077](#)) was used as the secondary antibody at a dilution of 1/1000. [ab195889](#) was used as a tubulin counterstain at a dilution of 1/200 and DAPI was used as a nuclear counterstain.



Flow Cytometry (Intracellular) - Anti-TRIM25/EFP antibody [EPR7315] (ab167154)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling TRIM25/EFP with purified ab167154 at 1/90 dilution (10ug/mL) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor®488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



Western blot - Anti-TRIM25/EGFP antibody
[EPR7315] (ab167154)

Anti-TRIM25/EGFP antibody [EPR7315] (ab167154) at 1/2000 dilution + SP2/0 (mouse spleen) whole cell lysate at 15 µg

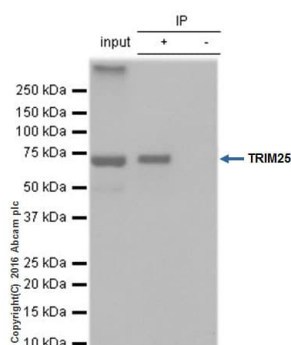
Secondary

Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 71 kDa

Additional bands at: 71 kDa. We are unsure as to the identity of these extra bands.

Blocking and diluting buffer: 5% NFDM /TBST



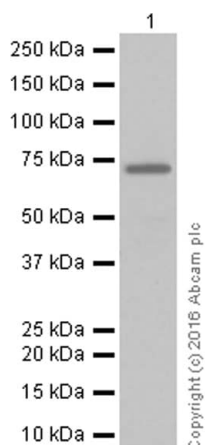
Immunoprecipitation - Anti-TRIM25/EGFP antibody
[EPR7315] (ab167154)

ab167154 immunoprecipitating TRIM25/EGFP. 10µg of cell lysate was incubated with primary antibody at a dilution of 1/40 and VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) at a dilution of 1/1000.

Lane 1: HeLa (human cervix adenocarcinoma) whole cell lysate (10ug)

Lane 2: HeLa (human cervix adenocarcinoma) whole cell lysate

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab167154 in HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate.



Western blot - Anti-TRIM25/EFP antibody
[EPR7315] (ab167154)

Anti-TRIM25/EFP antibody [EPR7315] (ab167154) at 1/10000 dilution + Rat brain lysate at 15 µg

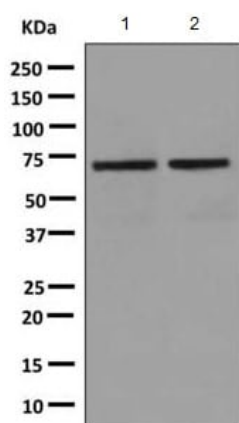
Secondary

Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 71 kDa

Additional bands at: 71 kDa. We are unsure as to the identity of these extra bands.

Blocking and diluting buffer: 5% NFDm/TBST



Western blot - Anti-TRIM25/EFP antibody
[EPR7315] (ab167154)

All lanes : Anti-TRIM25/EFP antibody [EPR7315] (ab167154) at 1/10000 dilution

Lane 1 : HeLa cell lysate

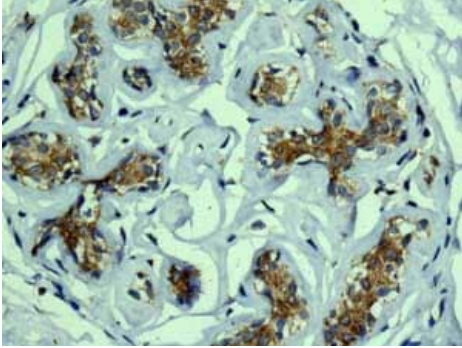
Lane 2 : K562 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

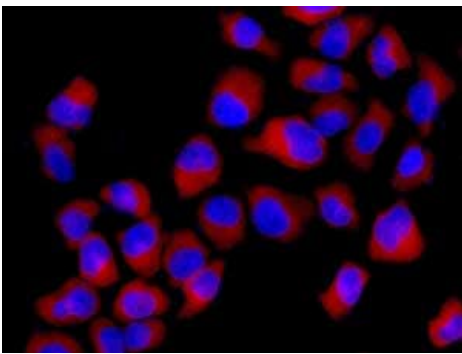
All lanes : HRP labeled goat anti-rabbit at 1/2000 dilution

Predicted band size: 71 kDa



Immunohistochemical analysis of paraffin-embedded Human breast tissue labeling TRIM25/EFP with ab167154 at 1/100 dilution.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TRIM25/EFP antibody [EPR7315] (ab167154)



Immunofluorescent analysis of HeLa cells labeling TRIM25/EFP with ab167154 at 1/100 dilution.

Immunocytochemistry/ Immunofluorescence - Anti-TRIM25/EFP antibody [EPR7315] (ab167154)

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-TRIM25/EFP antibody [EPR7315] (ab167154)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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