# abcam

## Product datasheet

## Anti-TRIM25/EFP antibody [EPR7315] ab167154



ייבע RabMAb

★★★★★ 1 Abreviews 26 References 画像数 12

製品の概要

製品名 Anti-TRIM25/EFP antibody [EPR7315]

製品の詳細 Rabbit monoclonal [EPR7315] to TRIM25/EFP

由来種 Rabbit

アプリケーション 適用あり: Flow Cyt (Intra), WB, IHC-P, ICC/IF, IP

種交差性 交差種: Mouse, Rat, Human

免疫原 Synthetic peptide within Human TRIM25/EFP aa 100-200. The exact sequence is proprietary.

Database link: Q14258

ポジティブ・コントロール 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.

特記事項 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**.

製品の特性

製品の状態

保存方法 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.

バッファー Preservative: 0.01% Sodium azide

Constituents: 40% Glycerol, 0.05% BSA

精製度 Protein A purified

ポリ/モノ モノクローナル クローン名 **EPR7315** 

### アプリケーション

**The Abpromise guarantee** <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab167154の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/60 - 1/90.
WB	*** <u>*</u> (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.

### ターゲット情報

機能 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.

組織特異性 Ubiquitous.

パスウェイ Protein modification; protein ubiquitination.

配列類似性 Contains 1 B30.2/SPRY domain.

Contains 1 RING-type zinc finger.

ドメイン The RING-type zinc finger is important for ISG15 E3 ligase activity and autoISGylation.

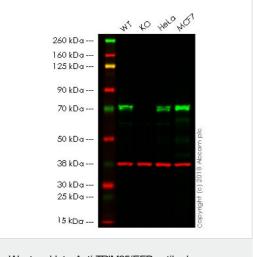
AutoISGylation negatively regulates ISG15 E3 ligase activity.

The C-terminal B30.2/SPRY domain interacts with the first N-terminal CARD domain of DDX58.

翻訳後修飾 Auto-ISGylated.

**細胞内局在** Cytoplasm. Colocalized with DDX58 at cytoplasmic perinuclear bodies.

#### 画像



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

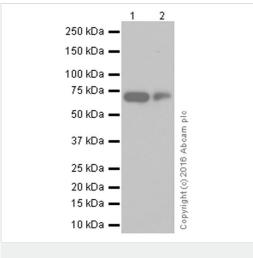
Lane 3 : HeLa whole cell lysate
Lane 4 : MCF7 whole cell lysate

Lysates/proteins at 20 µg per lane.

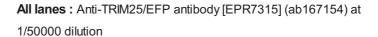
Predicted band size: 71 kDa

**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 <a href="mailto:ab9484">ab9484</a> (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 lgG H&L (IRDye® 800CW) preabsorbed <a href="mailto:ab216773">ab216773</a> and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed <a href="mailto:ab216776">ab216776</a> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



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



**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

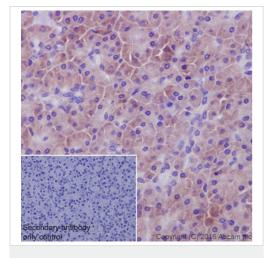
Lysates/proteins at 20 µg per lane.

## **Secondary**

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

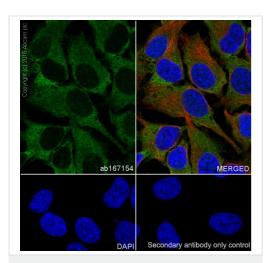
Predicted band size: 71 kDa





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

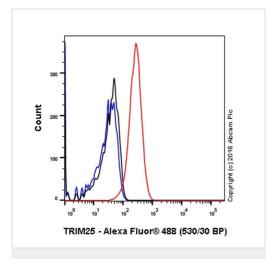
ab167154 staining TRIM25/EFP in human pancreas tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffinembedded sections). Tissue was fixed with paraformaldehyde and antigen retrieval was by heat mediation in a EDTA buffer. Samples were incubated with primary antibody at a dilution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



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

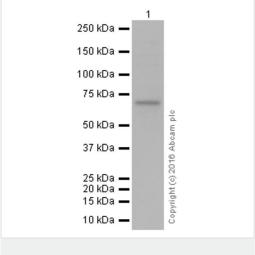
ab167154 staining TRIM25/EFP in HeLa (human cervix adenocarcinoma) cells by ICC/IF

(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 lgG (Alexa Fluor®488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal lgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



Anti-TRIM25/EFP antibody [EPR7315] (ab167154) at 1/2000 dilution + SP2/0 (mouse spleen) whole cell lysate at 15  $\mu$ g

## Secondary

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

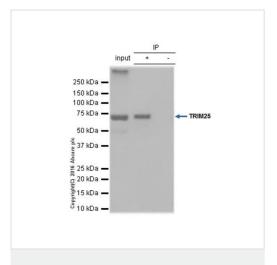
Predicted band size: 71 kDa

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

these extra bands.

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

Blocking and diluting buffer: 5% NFDM /TBST

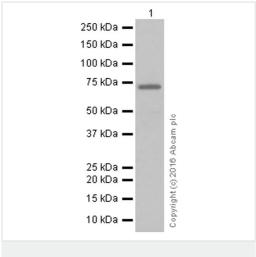


ab167154 immunoprecipitating TRIM25/EFP. 10µg of cell lysate was incubated with primary antibody at a dilution of 1/40 and VeriBlot for IP Detection Reagent (HRP) (ab131366) at a dilution of 1/1000.

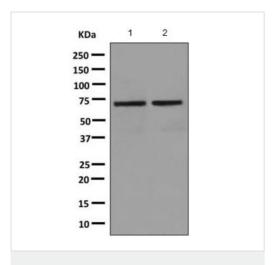
**Lane 1:** HeLa (human cervix adenocarcinoma) whole cell lysate (10ug)

Lane 2: HeLa (human cervix adenocarcinoma) whole cell lysate
Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab167154
in HeLa (Human epithelial cell line from cervix adenocarcinoma)
whole cell lysate.

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



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



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

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

## **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

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

Lane 1 : HeLa cell lysate

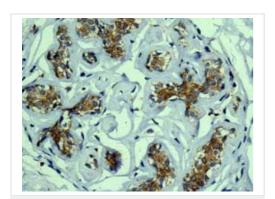
Lane 2 : K562 cell lysate

Lysates/proteins at 10 µg per lane.

## Secondary

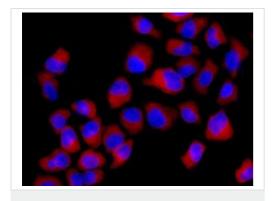
All lanes: HRP labeled goat anti-rabbit at 1/2000 dilution

Predicted band size: 71 kDa



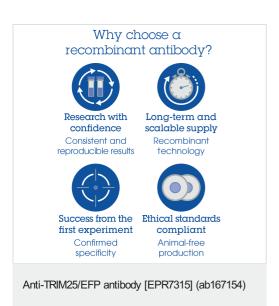
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TRIM25/EFP antibody
[EPR7315] (ab167154)

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



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

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



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