abcam

Product datasheet

Anti-GEF H1 antibody [EPR17963] - C-terminal ab201687

KO 評価済 リコンビナント RabMAb

4 References 画像数 7

製品の概要

製品名	Anti-GEF H1 antibody [EPR17963] - C-terminal
製品の詳細	Rabbit monoclonal [EPR17963] to GEF H1 - C-terminal
由来種	Rabbit
アプリケーション	**適用あり:** ICC/IF, WB, Flow Cyt (Intra)
種交差性	**交差種:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	WB: HeLa, HEK293, C6 and NIH/3T3 cell lysate. ICC/IF: HeLa and MCF-7 cells. Flow Cyt (intra): HEK293 cells.
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	Liquid
保存方法	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
バッファー	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 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	EPR17963
アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab201687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
ICC/IF		1/500.
WB		1/2000. Detects a band of approximately 112 kDa (predicted molecular weight: 112 kDa).
Flow Cyt (Intra)		1/100. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

ターゲット情報

機能

Activates Rho-GTPases by promoting the exchange of GDP for GTP. May be involved in epithelial barrier permeability, cell motility and polarization, dendritic spine morphology, antigen presentation, leukemic cell differentiation, cell cycle regulation, and cancer. Binds Rac-GTPases, but does not seem to promote nucleotide exchange activity toward Rac-GTPases, which was uniquely reported in PubMed:9857026. May stimulate instead the cortical activity of Rac. Inactive toward CDC42, TC10, or Ras-GTPases. Forms an intracellular sensing system along with NOD1 for the detection of microbial effectors during cell invasion by pathogens. Required for RHOA and RIP2 dependent NF-kappaB signaling pathways activation upon S.flexneri cell invasion. Involved not only in sensing peptidoglycan (PGN)-derived muropeptides through NOD1 that is independent of its GEF activity, but also in the activation of NF-kappaB by Shigella effector proteins (IpgB2 and OspB) which requires its GEF activity and the activation of RhoA.

配列類似性

Contains 1 DH (DBL-homology) domain.
Contains 1 PH domain.
Contains 1 phorbol-ester/DAG-type zinc finger.

ドメイン

The DH (DBL-homology) domain interacts with and promotes loading of GTP on RhoA.
The PH (pleckstrin-homology) domain is involved in microtubule binding and targeting to tight junctions.

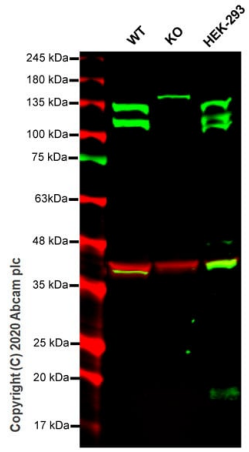
翻訳後修飾

Phosphorylation of Ser-886 by PAK1 induces binding to protein 14-3-3 zeta, promoting its relocation to microtubules and the inhibition of its activity. Phosphorylated by STK6 and CDK1 during mitosis, which negatively regulates its activity. Phosphorylation by MAPK1 or MAPK3 increases nucleotide exchange activity. Phosphorylation by PAK4 releases GEF-H1 from the microtubules.

細胞内局在

Cytoplasm. Cell junction > tight junction. Golgi apparatus. Cytoplasm > cytoskeleton > spindle. Cell projection > ruffle membrane. Localizes to the tips of cortical microtubules of the mitotic spindle during cell division, and is further released upon microtubule depolymerization. Recruited into membrane ruffles induced by S.flexneri at tight junctions of polarized epithelial cells.

画像



Western blot - Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687)

All lanes : Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : ARHGEF knockout HeLa cell lysate
Lane 3 : HEK-293T cell lysate

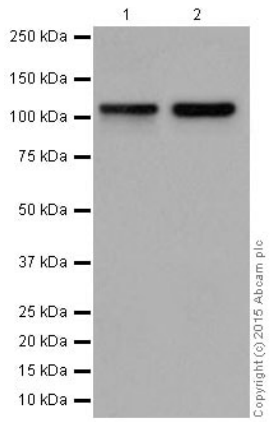
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 112 kDa
Observed band size: 112 kDa

Lanes 1-3: Merged signal (red and green). Green - ab201687 observed at 112 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab201687 Anti-GEF H1 antibody [EPR17963] - C-terminal was shown to specifically react with GEF H1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265797** (knockout cell lysate **ab257223**) was used. Wild-type and GEF H1 knockout samples were subjected to SDS-PAGE. ab201687 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687)

All lanes : Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687) at 1/10000 dilution

Lane 1 : HEK293 (Human embryonic kidney) lysate
Lane 2 : HEK293 (Human epithelial cells from cervix adenocarcinoma) lysate

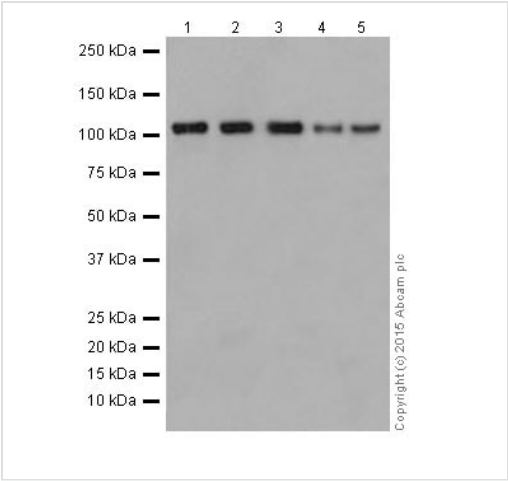
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 112 kDa
Observed band size: 112 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687)

All lanes : Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687) at 1/2000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Rat brain lysate
Lane 3 : Raw264.7 (Mouse macrophage cells transformed with Abelson murine leukemia virus) lysate
Lane 4 : C6 (Rat glial tumor cells) lysate
Lane 5 : NIH/3T3 (Mouse embyro fibroblast cells)

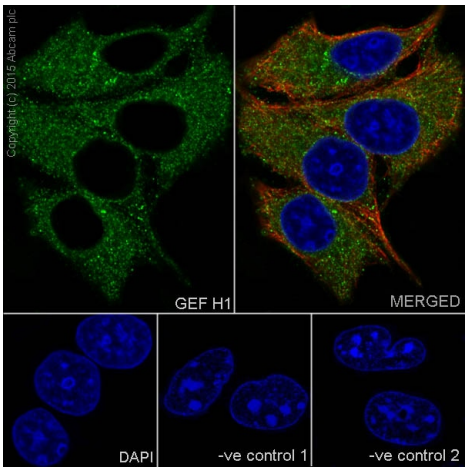
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 112 kDa
Observed band size: 112 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



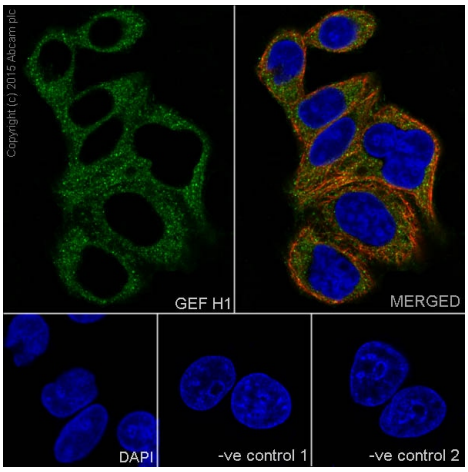
Immunocytochemistry/ Immunofluorescence - Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling GEF H1 with ab201687 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/500 dilution (green).

Confocal image showing cytoplasmic staining on HeLa cell line is observed.

The nuclear counter stain is DAPI (blue).

Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows;
1. ab201687 at 1/500 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
2. **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



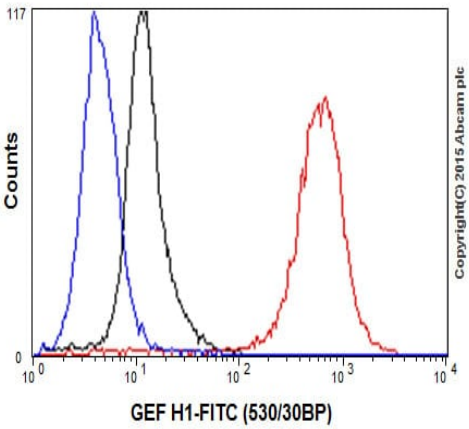
Immunocytochemistry/ Immunofluorescence - Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized MCF7 (Human breast adenocarcinoma cell line) cells labeling GEF H1 with ab201687 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/500 dilution (green).

Confocal image showing cytoplasmic staining on MCF7 cell line is observed.

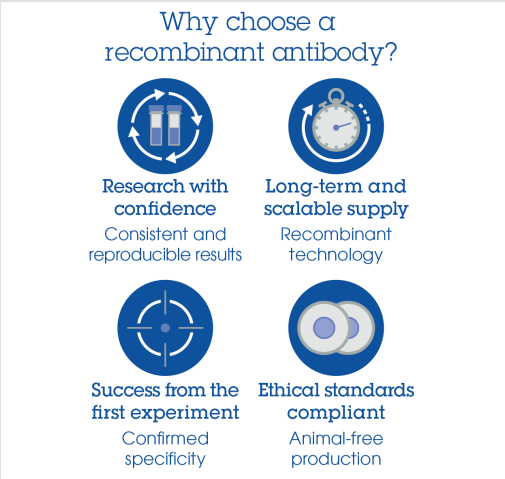
The nuclear counter stain is DAPI (blue).

Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows;
1. ab201687 at 1/500 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
2. **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



Flow Cytometry (Intracellular) - Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687)

Intracellular flow cytometric analysis of 2% paraformaldehyde-fixed HEK293 (human embryonic kidney) cells labeling GEF H1 with ab201687 at 1/100 dilution (red) compared with a rabbit monoclonal IgG isotype control (black) and a unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (FITC) at 1/150 dilution was used as the secondary antibody.



Anti-GEF H1 antibody [EPR17963] - C-terminal (ab201687)

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