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Product datasheet

Anti-Thrombospondin 1 antibody [EPR22927-54] ab267388

אילשעבע RabMAb

★★★★★★ 1 Abreviews 7 References 画像数 15

製品の概要

制口小柱井

製品名	Anti-Thrombospondin 1 antibody [EPR22927-54]	
製品の詳細	Rabbit monoclonal [EPR22927-54] to Thrombospondin 1	
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	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.	
ポジティブ・コントロール	WB: 3T3-L1 starved with 0.4% serum for 24 hours, then cultivated with 15% serum for 6 hours, whole cell lysate. HUVEC and mouse platelet lysates and rat platelet whole cell lysate. IHC-P: Human spleen, human bone marrow, human cervical carcinoma and mouse spleen tissues. ICC/IF: HUVEC cells and PC-12 cells. Flow Cyt (intra): HUVEC, 3T3-L1 and PC-12 cells. IP: HUVEC and mouse platelets 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 <u>see here</u>. Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <u>RabMAb[®] patents</u>. 	

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.
パッファー	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine)
精製度	Protein A purified
ポリ/モノ	モノクローナル

アプリケーション

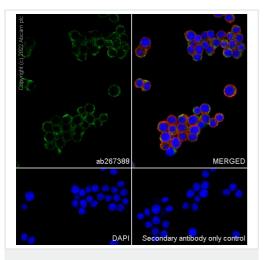
The Abpromise guarantee Abpromise保証は、次のテスト済みアプリケーションにおけるab267388の使用に適用されます

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。

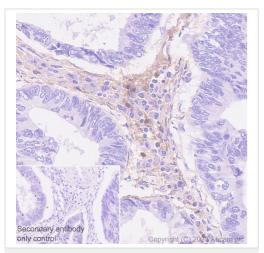
アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		1/50 - 1/500.
WB		1/1000. Predicted molecular weight: 129 kDa.
IHC-P		1/5000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/100.
IP		1/30.

ターゲット情報

機能	Adhesive glycoprotein that mediates cell-to-cell and cell-to-matrix interactions. Binds heparin. May play a role in dentinogenesis and/or maintenance of dentin and dental pulp (By similarity). Ligand for CD36 mediating antiangiogenic properties. Plays a role in ER stress response, via its interaction with the activating transcription factor 6 alpha (ATF6) which produces adaptive ER stress response factors.
配列類似性	Belongs to the thrombospondin family. Contains 2 EGF-like domains. Contains 1 laminin G-like domain. Contains 1 TSP C-terminal (TSPC) domain. Contains 3 TSP type-1 domains. Contains 8 TSP type-3 repeats. Contains 1 VWFC domain.
細胞内局在	Endoplasmic reticulum. Sarcoplasmic reticulum.



Immunocytochemistry/ Immunofluorescence - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)

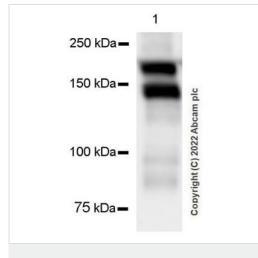
Immunofluorescent analysis of 100% Methanol-fixed, 0.1% TritonX-100 permeabilized PC-12 (rat adrenal gland pheochromocytoma cell) cells labelling Thrombospondin 1 with ab267388 at 1/100 (5.3 µg/ml) dilution, followed by **ab150077** AlexaFluor[®] 488 Goat anti-Rabbit secondary antibody at 1/1000 (2 µg/ml) dilution (Green). Confocal image showing cytoplasmic staining in PC-12 cell line. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8). **ab195889** Anti-alpha Tubulin antibody [DM1A] -Microtubule Marker (Alexa Fluor[®] 594) was used to counterstain tubulin at 1/200 (2.5 µg/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>**ab150077**</u> AlexaFluor[®]488 Goat anti-Rabbit secondary at 1/1000 (2 μ g/ml) dilution.

Immunohistochemical analysis of paraffin-embedded Human cervical carcinoma tissue labelling Thrombospondin 1 with ab267388 at 1/5000 (0.101 µg/ml) followed by a Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) at a Ready to use dilution. Positive staining on extracellular matrix of human cervical carcinoma. The section was incubated with ab267388 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) at Ready to use dilution.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Western blot - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388) Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388) at 1/1000 dilution + Rat platelet whole cell lysate at 20 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 129 kDa Observed band size: 180,140 kDa

Exposure time: 0 second

Blocking buffer and concentration : 5% NFDM/TBST Diluting buffer and concentration : 5% NFDM/TBST

The full-length TSP 1 (180 kDa) and a ~140 kDa band, likely to be a TSP 1 isoform or fragment, are observed.

The molecular weight observed is consistent with what has been described in the literature (PMID:1426766, 27588705).

Exposure time : 5.5 seconds

All lanes : Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388) at 1/1000 dilution

Lane 1 : HUVEC (human umbilical vein endothelial cell) whole cell lysate

Lane 2 : Mouse platelet lysate

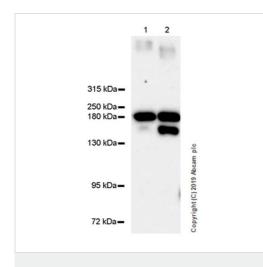
Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 129 kDa

Exposure time: 3 minutes



Western blot - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388) The full-length TSP 1 (180kDa) and a 160-kDa band, likely to be an TSP 1 isoform or fragment, are observed.

The molecular weight observed is consistent with what has been described in the literature (PMID:1426766, 27588705).

Blocking/Dilution buffer: 5% NFDM/TBST.

All lanes : Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388) at 1/1000 dilution

Lane 1: 3T3-L1 (mouse embryonic fibroblast) starved with 0.4% serum for 30 hours whole cell lysate
Lane 2: 3T3-L1 starved with 0.4% serum for 24 hours, then cultivated with 15% serum for 6 hours. whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 129 kDa

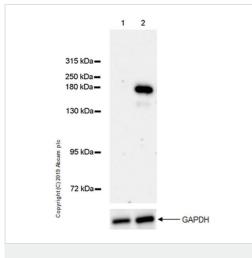
Exposure time: 3 minutes

TSP 1 is a serum-responsive gene. Its express is elevated in quiescent 3T3 with serum addition.

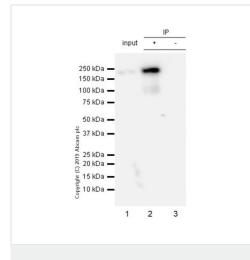
The full-length TSP 1 (180kDa) and a 160-kDa band, likely to be an TSP 1 isoform or fragment, are observed.

The molecular weight observed is consistent with what has been described in the literature (PMID:1426766, 27588705).

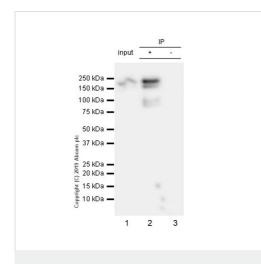
Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)



Immunoprecipitation - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)



Immunoprecipitation - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)

Thrombospondin 1 was immunoprecipitated from 0.35 mg HUVEC (human umbilical vein endothelial cell) whole cell lysate 10µg with ab267388 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab267388 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

Lane 1: HUVEC whole cell lysate 10µg.

Lane 2: ab267388 IP in HUVEC whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab267388 in HUVEC whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 8 seconds.

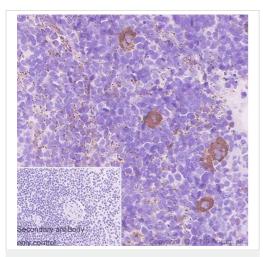
Thrombospondin 1 was immunoprecipitated from 0.35 mg mouse platelets whole cell lysate 10µg with ab267388 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab267388 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

Lane 1: Mouse platelets whole cell lysate 10µg.

Lane 2: ab267388 IP in mouse platelets whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab267388 in mouse platelets whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 8 seconds.

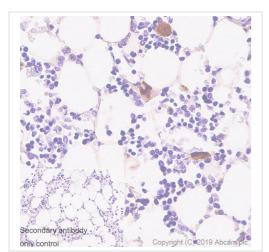


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling Thrombospondin 1 with ab267388 at 1/5000 dilution (0.1 µg/ml) followed by a ready to use Goat Anti-Rabbit lgG H&L (HRP Polymer). Positive staining on the megakaryocytes and platelets in the mouse spleen is observed. The section was incubated with ab267388 for 10 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

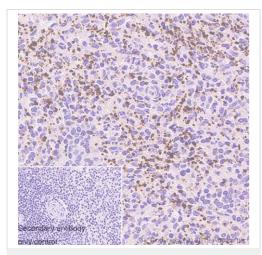


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)

Immunohistochemical analysis of paraffin-embedded human bone marrow tissue labeling Thrombospondin 1 with ab267388 at 1/5000 dilution (0.1 μ g/ml) followed by a ready to use Goat Anti-Rabbit lgG H&L (HRP Polymer). Positive staining on the megakaryocytes in the human bone marrow (PMID: 28239144). The section was incubated with ab267388 for 10 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

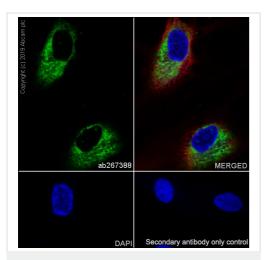


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)

Immunohistochemical analysis of paraffin-embedded human spleen tissue labeling Thrombospondin 1 with ab267388 at 1/5000 dilution (0.1 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer). Positive staining on the platelets in the human spleen (PMID: 28239144). The section was incubated with ab267388 for 10 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

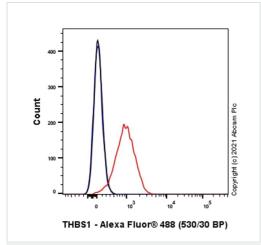


Immunocytochemistry/ Immunofluorescence - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)

Immunofluorescent analysis of 100% methanol-fixed, permeabilized HUVEC (human umbilical vein endothelial cell) cells labeling Thrombospondin 1 with ab267388 at 1/100 dilution (5 µg/ml), followed by **ab150077** AlexaFluor[®]488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in HUVEC cell line is observed. **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) was used to counterstain tubulin at 1/200 dilution (Red). The nuclear counterstain was DAPI (Blue).

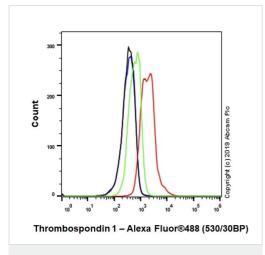
Secondary antibody only control: Secondary antibody is <u>ab150077</u> AlexaFluor[®]488 Goat anti-Rabbit secondary at 1/1000 dilution.

100% methanol fixation is recommended.

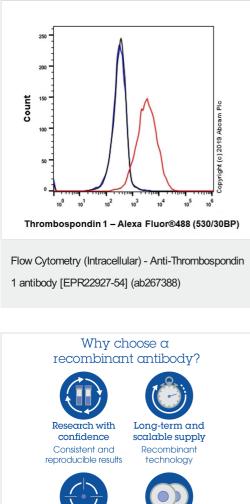


Intracellular Flow Cytometry analysis of PC-12 (Rat adrenal gland pheochromocytoma cell line) cells labeling Thrombospondin 1 with ab267388 at 1/500 dilution (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat Anti-rabbit IgG (Alexa Fluor[®] 488, **ab150081**) secondary antibody was used at 1/5000 dilution. Isotype control - Rabbit monoclonal IgG (Black) (**ab172730**). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

Flow Cytometry (Intracellular) - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)



Flow Cytometry (Intracellular) - Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388) Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized 3T3-L1 (mouse embryonic fibroblast) starved with 0.4% serum for 24h, then cultured with 15% serum for 6h (Red) / Untreated control (Green) cells labeling Thrombospondin 1 with ab267388 at 1/50 (Red) compared with a Rabbit monoclonal IgG (<u>ab172730</u>) / Black isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor[®] 488, <u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.



Success from the Ethical standards first experiment Confirmed specificity

Anti-Thrombospondin 1 antibody [EPR22927-54] (ab267388)

compliant

Animal-free

production

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Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized HUVEC (human umbilical vein endothelial cell) cells labeling Thrombospondin 1 with ab267388 at 1/50 (Red) compared with a Rabbit monoclonal IgG (ab172730) / Black isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor[®] 488, <u>ab150077</u>) at 1/2000 dilution was used as the secondary antibody.

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