

Anti-Robo1 antibody [EPR23699-159] ab256791

KO 評価済 リコンビナント RabMAb

2 References 画像数 9

製品の概要

製品名	Anti-Robo1 antibody [EPR23699-159]
製品の詳細	Rabbit monoclonal [EPR23699-159] to Robo1
由来種	Rabbit
アプリケーション	適用あり: WB, Flow Cyt, IP 適用なし: ICC/IF, IHC-Fr or IHC-P
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: HeLa, HAP1 and HepG2 whole cell lysates; Mouse E14.5 brain and C57 E18 brain tissue lysates; Rat E14 brain tissue lysate; His-tagged human Robo1 recombinant protein (aa660-897(Q883R)). IP: Mouse E14.5 brain tissue lysate and HepG2 whole cell lysate. Flow cyt: HeLa cells.
特記事項	<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>

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: 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 EPR23699-159
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ab256791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確認ください。

アプリケーション	Abreviews	特記事項
WB		1/1000. Detects a band of approximately 120, 130, 250 kDa (predicted molecular weight: 181 kDa).
Flow Cyt		1/500.
IP		1/30.

追加情報 Is unsuitable for ICC/IF, IHC-Fr or IHC-P.

ターゲット情報

機能 Receptor for SLIT1 and SLIT2 which are thought to act as molecular guidance cue in cellular migration, including axonal navigation at the ventral midline of the neural tube and projection of axons to different regions during neuronal development. In axon growth cones, the silencing of the attractive effect of NTN1 by SLIT2 may require the formation of a ROBO1-DCC complex. May be required for lung development.

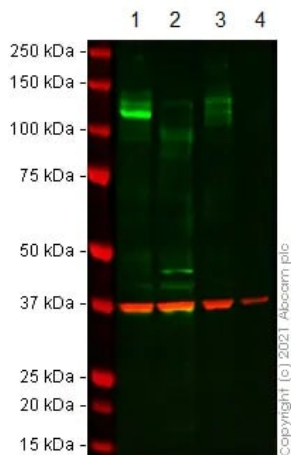
組織特異性 Widely expressed, with exception of kidney.

配列類似性 Belongs to the immunoglobulin superfamily. ROBO family.
Contains 3 fibronectin type-III domains.
Contains 5 Ig-like C2-type (immunoglobulin-like) domains.

翻訳後修飾 Ubiquitinated. May be deubiquitinated by USP33.

細胞内局在 Membrane.

画像



Western blot - Anti-Robo1 antibody [EPR23699-159] (ab256791)

All lanes : Anti-Robo1 antibody [EPR23699-159] (ab256791) at 1/1000 dilution

Lane 1 : Wild-type HAP1 cell lysate

Lane 2 : ROBO1 CRISPR/Cas9 edited HAP1 cell lysate

Lane 3 : HeLa cell lysate

Lane 4 : MCF7 cell lysate

Lysates/proteins at 20 µg per lane.

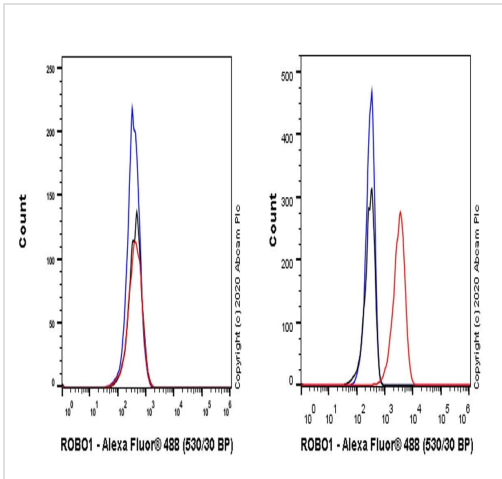
Performed under reducing conditions.

Predicted band size: 181 kDa

Observed band size: 120 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab256791 observed at 120 kDa. Red - loading control, **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab256791 was shown to react with Robo1 in western blot. The band observed in the CRISPR/Cas9 edited lysate lane below 120 kDa is likely to represent a truncated form. This has not been investigated further. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween[®]) before incubation with ab256791 and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated 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 in 20000 dilution for 1 h at room temperature before imaging.



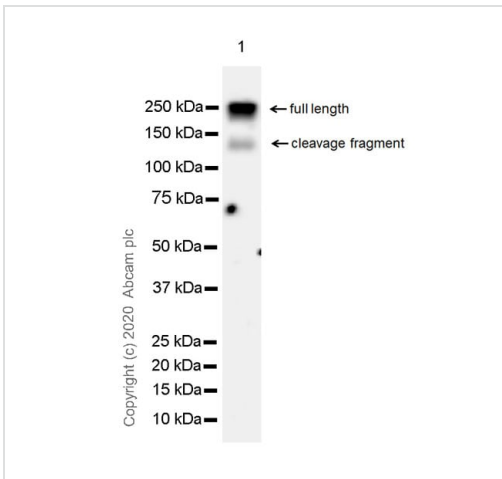
Flow Cytometry - Anti-Robo1 antibody [EPR23699-159] (ab256791)

Flow cytometric analysis of LoVo (Human colorectal adenocarcinoma epithelial cell) (Left) / HeLa (Human cervix adenocarcinoma epithelial cell) (Right) cells labelling Robo1 with ab256791 at 1/500 dilution (0.1 µg) (Red) compared with a Rabbit monoclonal IgG (**ab172730**) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).

Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

Negative control: LoVo(PMID: 16740745).

Gated on viable cells.



Western blot - Anti-Robo1 antibody [EPR23699-159] (ab256791)

Anti-Robo1 antibody [EPR23699-159] (ab256791) at 1/1000 dilution + HeLa whole cell lysate at 20 µg

Secondary

VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at 1/1000 dilution

Predicted band size: 181 kDa

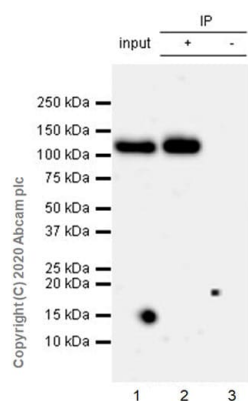
Observed band size: 130,250 kDa

Exposure time: 92 seconds

Buffer and concentration: 5% NFDM/TBST

250 kDa full length band and 130 kDa cleavage fragment band were observed.

Freshly prepared lysate was used in this blot.



Immunoprecipitation - Anti-Robo1 antibody
[EPR23699-159] (ab256791)

Robo1 was immunoprecipitated from 0.35 mg HepG2 whole cell lysate with ab256791 at 1/30 dilution (2ug in 0.35mg lysates).

Western blot was performed on the immunoprecipitate using ab256791 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)([ab131366](#)) was used at 1/5000 dilution.

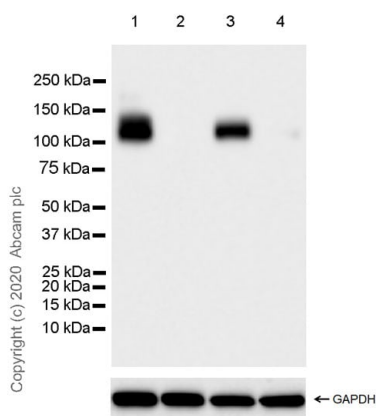
Lane 1: HepG2 whole cell lysate 10 ug.

Lane 2: ab256791 IP in HepG2 whole cell lysate.

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab256791 in HepG2 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDm/TBST.

Exposure time: 93 seconds.



Western blot - Anti-Robo1 antibody [EPR23699-159]
(ab256791)

All lanes : Anti-Robo1 antibody [EPR23699-159] (ab256791) at 1/1000 dilution

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

Lane 2 : LoVo (human colorectal adenocarcinoma epithelial cell) whole cell lysate

Lane 3 : HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate

Lane 4 : MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated ([ab97051](#)) at 1/50000 dilution

Predicted band size: 181 kDa

Observed band size: 120-130 kDa

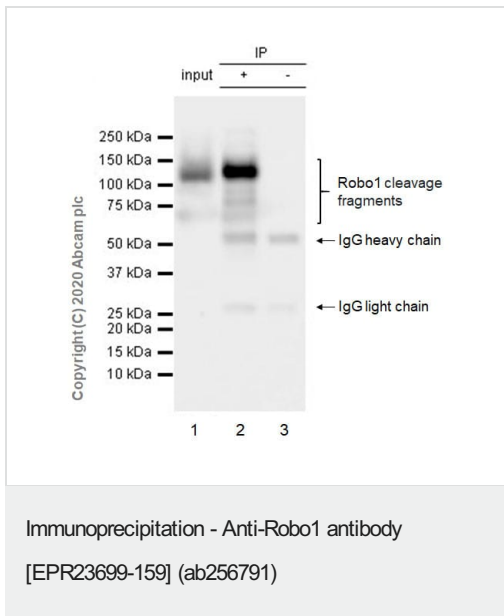
Blocking and diluting buffer and concentration: 5% NFDm/TBST.

The molecular weight observed is consistent with what has been described in the literature (PMID: 20471383, 16740745). The 120/130-kDa bands correspond to the Robo1 cleavage fragments from metalloprotease and/or gamma secretase activities (PMID: 20570941, 20471383).

Negative control: LoVo (PMID: 16740745); **Low expression:** MCF7 (PMID: 16740745).

Lysates used in this blot have experienced a freeze-thaw cycle.

Exposure time: 10 seconds.



Robo1 was immunoprecipitated from 0.35 mg Mouse E14.5 brain tissue lysate with ab256791 at 1/30 dilution (2ug in 0.35mg lysates).

Western blot was performed on the immunoprecipitate using ab256791 at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP)([ab131366](#)) was used at 1/1000 dilution.

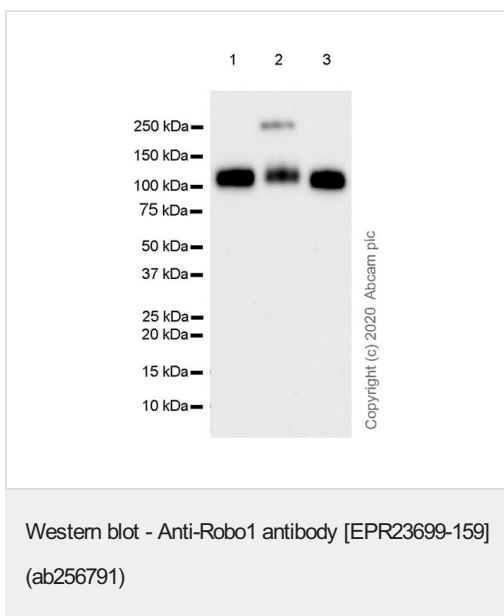
Lane 1: Mouse E14.5 brain tissue lysate 10 ug.

Lane 2: ab256791 IP in Mouse E14.5 brain tissue lysate.

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab256791 in mouse E14.5 brain tissue lysate.

Blocking and dilution buffer and concentration: 5% NFDm/TBST.

Exposure time: 1 second.



All lanes : Anti-Robo1 antibody [EPR23699-159] (ab256791) at 1/1000 dilution

Lane 1 : Mouse E14.5 brain tissue lysate

Lane 2 : C57 E18 brain tissue lysate

Lane 3 : Rat E14 brain tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated ([ab97051](#)) at 1/50000 dilution

Predicted band size: 181 kDa

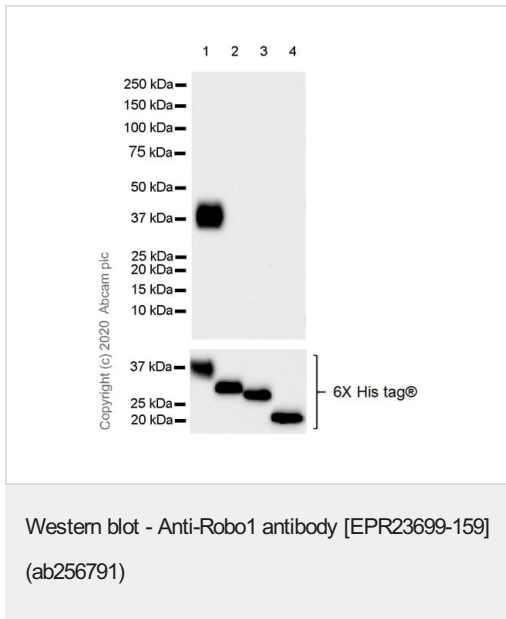
Observed band size: 120,250 kDa

Blocking and diluting buffer and concentration: 5% NFDm/TBST.

The 120 kDa band corresponds to the Robo1 cleavage fragments from metalloprotease and/or gamma secretase activities (PMID: 20570941, 20471383).

Lysates used in this blot have experienced a freeze-thaw cycle.

Exposure time: 70 seconds.



All lanes : Anti-Robo1 antibody [EPR23699-159] (ab256791) at 1/1000 dilution

Lane 1 : His-tagged human Robo1 recombinant protein (aa660-897(Q883R))

Lane 2 : His-tagged human Robo2 recombinant protein (aa621-859)

Lane 3 : His-tagged human Robo3 recombinant protein (aa655-892)

Lane 4 : His-tagged human Robo4 recombinant protein (aa301-463)

Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (**ab97051**) at 1/100000 dilution

Predicted band size: 181 kDa


Observed band size: 38 kDa

Blocking and diluting buffer and concentration: 5% NFDm/TBST.

These recombinant proteins were made in house. The sample loaded onto lane 1 was purified Robo1 recombinant protein expressed from a mammalian - HEK-293 expression system. The samples loaded onto lanes 2-4 were E.coil extracts containing Robo2, Robo3 or Robo4 recombinant protein respectively.

Exposure time: 26 seconds.

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-Robo1 antibody [EPR23699-159] (ab256791)

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

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