# abcam

### **Product datasheet**

## Anti-CLASP1 antibody [EPR3409] ab108620

KO 評価済 リコンピナント RabMAb

★★★★★★★ 1 Abreviews 11 References 画像数 12

#### 製品の概要

製品名	Anti-CLASP1 antibody [EPR3409]
製品の詳細	Rabbit monoclonal [EPR3409] to CLASP1
由来種	Rabbit
アプリケーション	適用あり: Flow Cyt (Intra), ICC/IF, WB, IHC-P 適用なし: IP
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: U87-MG, HeLa, T47-D and C6 cell lysates and rat brain tissue lysate. IHC-P: Human and mouse kidney and human cerebral cortex tissues. ICC/IF: HeLa cells. Flow Cyt (intra): HeLa cells.
特記事項	<ul> <li>This product is a recombinant monoclonal antibody, which offers several advantages including:</li> <li>High batch-to-batch consistency and reproducibility</li> <li>Improved sensitivity and specificity</li> <li>Long-term security of supply</li> <li>Animal-free production</li> <li>For more information <u>see here</u>.</li> <li>Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <u>RabMAb<sup>®</sup> patents</u>.</li> </ul>

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. Stable for 12 months at -20°C.
バッファー	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 40% Glycerol (glycerin, glycerine), 59% PBS, 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	EPR3409

#### 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ab108620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。

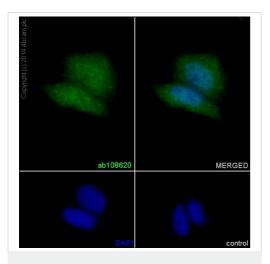
Abreviews	特記事項
	1/100. <u>ab172730</u> - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
	1/100.
★★☆☆☆ <u>(1)</u>	1/5000 - 1/50000. Detects a band of approximately 160 kDa (predicted molecular weight: 169 kDa).
	1/250 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

#### 追加情報

Is unsuitable for IP.

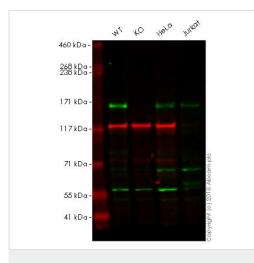
#### ターゲット情報

機能	Microtubule plus-end tracking protein that promotes the stabilization of dynamic microtubules. Involved in the nucleation of noncentrosomal microtubules originating from the trans-Golgi network (TGN). Required for the polarization of the cytoplasmic microtubule arrays in migrating cells towards the leading edge of the cell. May act at the cell cortex to enhance the frequency of rescue of depolymerizing microtubules by attaching their plus-ends to cortical platforms composed of ERC1 and PHLDB2. This cortical microtubule stabilizing activity is regulated at least in part by phosphatidylinositol 3-kinase signaling. Also performs a similar stabilizing function at the kinetochore which is essential for the bipolar alignment of chromosomes on the mitotic spindle.
配列類似性	Belongs to the CLASP family. Contains 7 HEAT repeats.
翻訳後修飾	Phosphorylated upon DNA damage, probably by ATM or ATR.
細胞内局在	Cytoplasm > cytoskeleton. Cytoplasm > cytoskeleton > centrosome. Chromosome > centromere > kinetochore. Cytoplasm > cytoskeleton > spindle. Golgi apparatus > trans-Golgi network. Localizes to microtubule plus ends. Localizes to centrosomes, kinetochores and the mitotic spindle from prometaphase. Subsequently localizes to the spindle midzone from anaphase and to the midbody from telophase. In migrating cells localizes to the plus ends of microtubules within the cell body and to the entire microtubule lattice within the lamella. Localizes to the cell cortex and this requires ERC1 and PHLDB2.



Immunocytochemistry/ Immunofluorescence - Anti-CLASP1 antibody [EPR3409] (ab108620) Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling CLASP1 (green) with ab108620 at 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

Control: primary antibody (1/100) and secondary antibody, <u>**ab150120**</u> Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).



Western blot - Anti-CLASP1 antibody [EPR3409] (ab108620) **All lanes :** Anti-CLASP1 antibody [EPR3409] (ab108620) at 1/10000 dilution

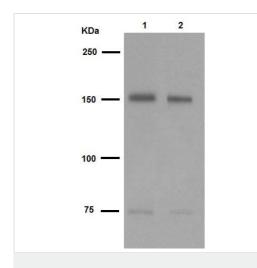
Lane 1 : Wild-type HAP1 whole cell lysate Lane 2 : CLASP1 knockout HAP1 whole cell lysate Lane 3 : HeLa whole cell lysate Lane 4 : Jurkat whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 169 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab108620 observed at 169 kDa. Red - loading control, <u>ab130007</u>, observed at 130 kDa.

ab108620 was shown to recognize CLASP1 in wild-type HAP1 cells as signal was lost at the expected MW in CLASP1 knockout cells. Additional cross-reactive bands were observed in the wildtype and knockout cells. Wild-type and CLASP1 knockout samples were subjected to SDS-PAGE. Ab108620 and <u>ab130007</u> (Mouse anti-Vinculin 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<sup>®</sup> 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> dilution for 1 hour at room temperature before imaging.



Western blot - Anti-CLASP1 antibody [EPR3409] (ab108620) **All lanes :** Anti-CLASP1 antibody [EPR3409] (ab108620) at 1/20000 dilution (purified)

Lane 1 : U87-MG cell lysate
Lane 2 : HeLa cell lysate

Lysates/proteins at 10 µg per lane.

#### Secondary

**All lanes :** Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Predicted band size: 169 kDa Observed band size: 160 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

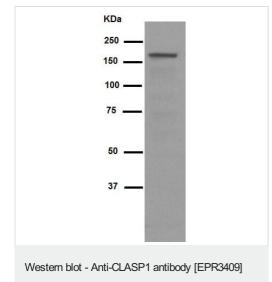
Anti-CLASP1 antibody [EPR3409] (ab108620) at 1/50000 dilution (purified) + Rat brain tissue lysate at 10 μg

#### Secondary

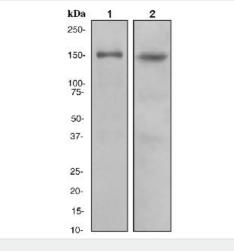
Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 169 kDa Observed band size: 160 kDa

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



(ab108620)



Western blot - Anti-CLASP1 antibody [EPR3409]

(ab108620)

Lysates/proteins at 10 µg per lane.

Predicted band size: 169 kDa

1/10000 dilution (unpurified)

Lane 1 : T47-D cell lysate

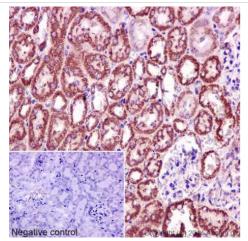
Lane 2 : C6 cell lysate

Negative control

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CLASP1 antibody [EPR3409] (ab108620)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cerebral cortex tissue labelling CLASP1 with purified ab108620 at 1/250. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

All lanes : Anti-CLASP1 antibody [EPR3409] (ab108620) at



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CLASP1 antibody [EPR3409] (ab108620)

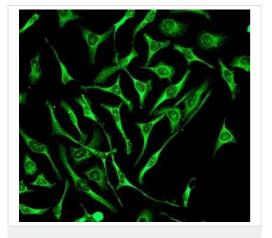


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CLASP1 antibody [EPR3409] (ab108620)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue labeling CLASP1 with purified ab108620 at 1/250. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

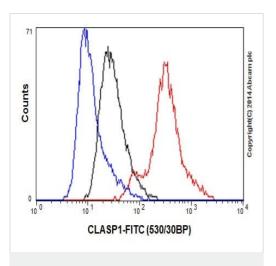
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human kidney tissue labelling CLASP1 with unpurified ab108620 at 1/250.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

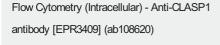


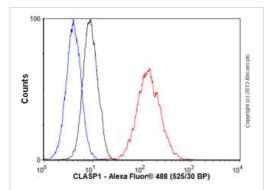
Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling CLASP1 with unpurified ab108620 at 1/100.

Immunocytochemistry/ Immunofluorescence - Anti-CLASP1 antibody [EPR3409] (ab108620)



Intracellular Flow Cytometry analysis of HeLa cells labelling CLASP1 with purified ab108620 at 1/50 (red). Cells were fixed with 80% methanol. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.





Flow Cytometry (Intracellular) - Anti-CLASP1 antibody [EPR3409] (ab108620) Overlay histogram showing HeLa cells stained with unpurified ab108620 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab108620, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluorr<sup>®</sup> 488 goat anti-rabbit IgG (H&L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) ( $0.1\mu$ g/1x10<sup>6</sup> cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



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