abcam

Product datasheet

Anti-DAB2 antibody [EP2297Y] ab76253

יעלאעבע RabMAb

★★★★★ 1 Abreviews 3 References 画像数6

製品の概要

製品名 Anti-DAB2 antibody [EP2297Y]

製品の詳細 Rabbit monoclonal [EP2297Y] to DAB2

由来種 Rabbit

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

種交差性 交差種: 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: PC3 and HeLa whole cell lysates; ICC/IF: HeLa cells; IP: HeLa whole cell lysate; Flow Cyt

(intra): HeLa cells.

特記事項 Our RabMAb® technology is a patented hybridoma-based technology for making rabbit

monoclonal antibodies. For details on our patents, please refer to **RabMAb**® patents.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

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: 7.20

Preservative: 0.01% Sodium azide

Constituents: 0.05% BSA, 40% Glycerol (glycerin, glycerine), 59% PBS

精製度 Protein A purified

ポリ/モノ モノクローナル **ウローン名** EP2297Y **I I G**

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ab76253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)		1/100. For unpurified use at 1/50. <u>ab172730</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/1000. Detects a band of approximately 105 kDa (predicted molecular weight: 83 kDa). For unpurified use at 1/2000 - 1/10000.
IP		1/20.
ICC/IF		1/100. For unpurified use at 1/100 - 1/250.

ターゲット情報

機能

Adapter protein that functions as clathrin-associated sorting protein (CLASP) required for clathrinmediated endocytosis of selected cargo proteins. Can bind and assemble clathrin, and binds simultaneously to phosphatidylinositol 4,5-bisphosphate (Ptdlns(4,5)P2) and cargos containg nonphosphorylated NPXY internalization motifs, such as the LDL receptor, to recruit them to clathrincoated pits. Can function in clathrin-mediated endocytosis independently of the AP-2 complex. Involved in endocytosis of integrin beta-1; this function seems to redundant with the AP-2 complex and seems to require DAB2 binding to endocytosis accessory EH domain-containing proteins such as EPS15, EPS15L1 and ITSN1. Involved in endocytosis of cystic fibrosis transmembrane conductance regulator/CFTR. Involved in endocytosis of megalin/LRP2 lipoprotein receptor during embryonal development. Required for recycling of the TGF-beta receptor. Involved in CFTR trafficking to the late endosome. Involved in several receptor-mediated signaling pathways. Involved in TGF-beta receptor signaling and facilitates phosphorylation of the signal transducer SMAD2. Mediates TFG-beta-stimulated JNK activation. May inhibit the canoniocal Wnt/betacatenin signaling pathway by stabilizing the beta-catenin destruction complex through a competing association with axin preventing its dephosphorylation through protein phosphatase 1 (PP1). Sequesters LRP6 towards clathrin-mediated endocytosis, leading to inhibition of Wnt/beta-catenin signaling. May activate non-canonical Wnt signaling. In cell surface growth factor/Ras signaling pathways proposed to inhibit ERK activation by interrupting the binding of GRB2 to SOS1 and to inhibit SRC by preventing its activating phosphorylation at 'Tyr-419'. Proposed to be involved in modulation of androgen receptor (AR) signaling mediated by SRC activation; seems to compete with AR for interaction with SRC. Plays a role in the CSF-1 signal transduction pathway. Plays a role in cellular differentiation. Involved in cell positioning and formation of visceral endoderm (VE) during embryogenesis and proposed to be required in the VE to respond to Nodal signaling coming from the epiblast. Required for the epithelial to

mesenchymal transition, a process necessary for proper embryonic development. May be involved in myeloid cell differentiation and can induce macrophage adhesion and spreading. May act as a tumor suppressor.

組織特異性 Expressed in deep invaginations, inclusion cysts and the surface epithelial cells of the ovary. Also

expressed in breast epithelial cells, spleen, thymus, prostate, testis, macrophages, fibroblasts, lung epithelial cells, placenta, brain stem, heart and small intestine. Expressed in kidney proximal

tubular epithelial cells (at protein level).

配列類似性 Contains 1 PID domain.

ドメイン The PID domain binds to predominantly non-phosphorylated NPXY internalization motifs present

in members of the LDLR and APP family; it also mediates simultaneous binding to

phosphatidylinositol 4,5-bisphosphate.

The Asn-Pro-Phe (NPF) motifs, which are found in proteins involved in the endocytic pathway,

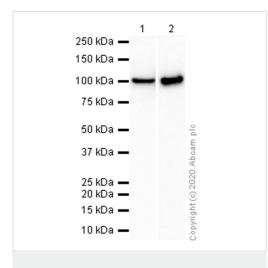
mediate the interaction with the EH domain of EPS15, EPS15R and ITSN1.

翻訳後修飾 Phosphorylated. Phosphorylation during mitosis is leading to membrane displacement.

細胞内局在 Cytoplasm. Cytoplasmic vesicle, clathrin-coated vesicle membrane. Membrane, clathrin-coated pit. Colocalizes with large insert-containing isoforms of MYO6 at clathrin-coated pits/vesicles.

During mitosis is progressively displaced from the membrane and translocated to the cytoplasm.

画像



Western blot - Anti-DAB2 antibody [EP2297Y] (ab76253)

All lanes : Anti-DAB2 antibody [EP2297Y] (ab76253) at 1/1000 dilution (purified)

Lane 1 : PC-3 (Human prostate adenocarcinoma epithelial cell)

whole cell lysate

Lane 2: HeLa (Human cervix adenocarcinoma epithelial cell)

whole cell lysate

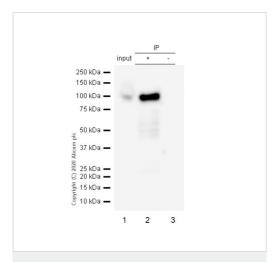
Lysates/proteins at 20 µg per lane.

Secondary

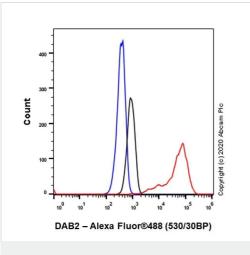
 $\textbf{All lanes:} \ \ \text{Goat Anti-Rabbit lgG (HRP) with minimal cross-reactivity}$

with human IgG at 1/2000 dilution

Predicted band size: 83 kDa **Observed band size:** 105 kDa



Immunoprecipitation - Anti-DAB2 antibody [EP2297Y] (ab76253)



Flow Cytometry (Intracellular) - Anti-DAB2 antibody [EP2297Y] (ab76253)

Purified ab76253 at 1/20 dilution (0.5 μ g) immunoprecipitating DAB2 in HeLa whole cell lysate.

Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10 μg

Lane 2 (+): ab76253 + HeLa whole cell lysate.

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab76253 in HeLa whole cell lysate.

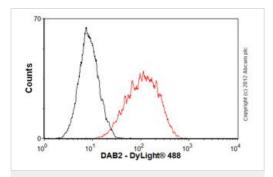
VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) (1/1000 dilution) was used for Western blotting.

Blocking Buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.

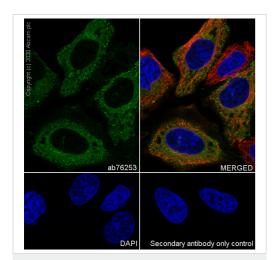
Observed band size: 105 kDa

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling DAB2 with purified ab76253 at 1/100 dilution (1µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluorr[®]488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



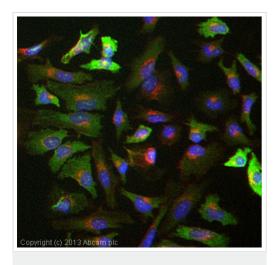
Flow Cytometry (Intracellular) - Anti-DAB2 antibody [EP2297Y] (ab76253)

Overlay histogram showing HeLa cells stained with unpurified ab76253 (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 (ab76253, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monclonal) (1 μ g/1x106 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Immunocytochemistry/ Immunofluorescence - Anti-DAB2 antibody [EP2297Y] (ab76253)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling DAB2 with purified ab76253 at 1/100 dilution (1.1 μ g/mL). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% TritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 μ g/mL). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 μ g/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunocytochemistry/ Immunofluorescence - Anti-DAB2 antibody [EP2297Y] (ab76253)

ab76253 (unpurified) stained HeLa cells. The cells were 4% formaldehyde fixed for 10 minutes at room temperature and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1hour at room temperature to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (unpurified ab76253 at 1/100 dilution) overnight at +4°C. The secondary antibody (pseudocolored green) was Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) used at a 1/1000 dilution for 1hour at room temperature. Alexa Fluor® 594 WGA was used to label plasma membranes (pseudo-colored red) at a 1/200 dilution for 1hour at room temperature. DAPI was used to stain the cell nuclei (pseudo-colored blue) at a concentration of 1.43μM for 1hour at room temperature.

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