abcam

Product datasheet

Anti-RCN1/RCN antibody [EPR17163-117] ab210404



ייבע RabMAb

1 References 画像数 14

製品の概要

製品名 Anti-RCN1/RCN antibody [EPR17163-117]

製品の詳細 Rabbit monoclonal [EPR17163-117] to RCN1/RCN

由来種 Rabbit

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

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: Human placenta and fetal kidney lysates; HEK-293, SH-SY5Y, HeLa, Neuro-2a, C6, PC-12

> and NIH/3T3 whole cell lysates; E14 mouse embryo lysate; E14 rat embryo lysate; Mouse brain, heart, kidney and spleen lysates; Rat brain, heart, kidney and spleen lysates. IHC-P: Human cerebrum and lung adenocarcinoma tissues; Mouse cerebral cortex tissue; Rat hippocampus tissue. ICC/IF: SH-SY5Y, Neuro-2a and NIH/3T3 cells. Flow Cyt (intra): NIH/3T3 and SH-SY5Y

cells. IP: 293T whole cell 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 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 EPR17163-117

アイソタイプ lgG

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ab210404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)		1/700.
IP		1/50.
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/2000. Detects a band of approximately 44, 46 kDa (predicted molecular weight: 39 kDa).
ICC/IF		1/500.

ターゲット情報

機能 May regulate calcium-dependent activities in the endoplasmic reticulum lumen or post-ER

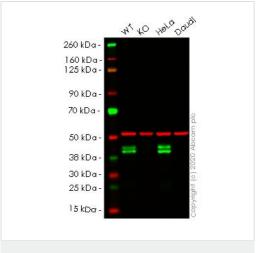
compartment.

配列類似性 Belongs to the CREC family.

Contains 6 EF-hand domains.

細胞内局在 Endoplasmic reticulum lumen.

画像



Western blot - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)

All lanes : Anti-RCN1/RCN antibody [EPR17163-117] (ab210404) at 1/2000 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: RCN1 knockout HEK-293T cell lysate

Lane 3 : HeLa cell lysate

Lane 4 : Daudi cell lysate

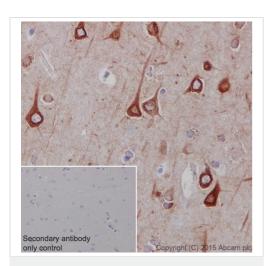
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 39 kDa **Observed band size:** 45 kDa

Lanes 1-4: Merged signal (red and green). Green - ab210404 observed at 45 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) observed at 50 kDa.

ab210404 was shown to react with RCN1 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266098 (knockout cell lysate ab258172) was used. Wild-type HEK-293T and RCN1 knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab210404 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) overnight at 4°C at a 1 in 2000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RCN1/RCN antibody
[EPR17163-117] (ab210404)

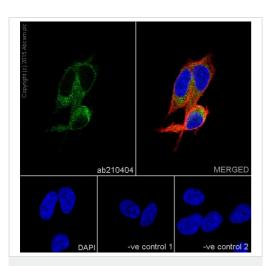
Immunohistochemical analysis of paraffin-embedded Human cerebrum tissue labeling RCN1/RCN with ab210404 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Cytoplasm staining on neurons of Human cerebrum is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is **ab97051** at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized SH-SY5Y (Human neuroblastoma cell line from bone marrow) cells labeling RCN1/RCN with ab210404 at 1/500 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing cytoplasmic staining on SH-SY5Y cell line.

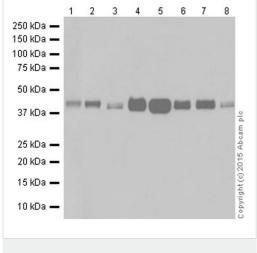
The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse lgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab210404 at 1/500 dilution followed by $\underline{ab150120}$ at 1/1000 dilution.

-ve control 2: <u>ab7291</u> at 1/1000 dilution followed by <u>ab150077</u> at 1/1000 dilution.



Western blot - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404) **All lanes :** Anti-RCN1/RCN antibody [EPR17163-117] (ab210404) at 1/5000 dilution

Lane 1: Human placenta lysate

Lane 2: Human fetal kidney lysate

Lane 3: HEK-293 (Human epithelial cell line from embryonic

kidney) whole cell lysate

Lane 4: SH-SY5Y (Human neuroblastoma cell line from bone

marrow) whole cell lysate

Lane 5: HeLa (Human epithelial cell line from cervix

adenocarcinoma) whole cell lysate

Lane 6: E14 mouse embryo lysate

Lane 7: E14 rat embryo lysate

Lane 8: Neuro-2a (Mouse neuroblastoma cell line) whole cell

lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/100000 dilution

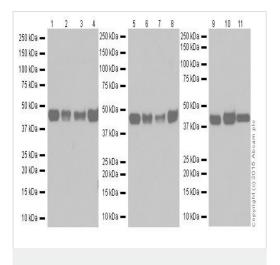
Predicted band size: 39 kDa **Observed band size:** 44,46 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been described in the literature (PMID: 8416973).

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Western blot - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)

All lanes : Anti-RCN1/RCN antibody [EPR17163-117] (ab210404) at 1/2000 dilution

Lane 1: Mouse brain lysate

Lane 2: Mouse heart lysate

Lane 3: Mouse kidney lysate

Lane 4: Mouse spleen lysate

Lane 5: Rat brain lysate

Lane 6: Rat heart lysate

Lane 7: Rat kidney lysate

Lane 8: Rat spleen lysate

Lane 9: C6 (Rat glial tumor cell line) whole cell lysate

Lane 10: PC-12 (Rat adrenal gland pheochromocytoma cell line)

whole cell lysate

Lane 11: NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell

lysate

Lysates/proteins at 10 µg per lane.

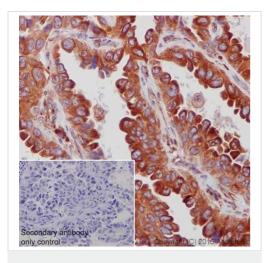
Secondary

All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/100000 dilution

Predicted band size: 39 kDa **Observed band size:** 44,46 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1-8: 1 minute; Lane 9,10 and 11: 10 seconds.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RCN1/RCN antibody
[EPR17163-117] (ab210404)

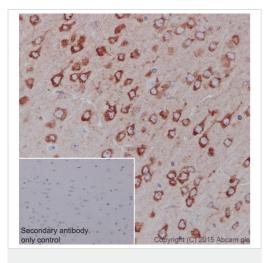
Immunohistochemical analysis of paraffin-embedded Human lung adenocarcinoma tissue labeling RCN1/RCN with ab210404 at 1/2000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Cytoplasm staining on tumor cells of Human lung adenocarcinoma is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is **ab97051** at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RCN1/RCN antibody
[EPR17163-117] (ab210404)

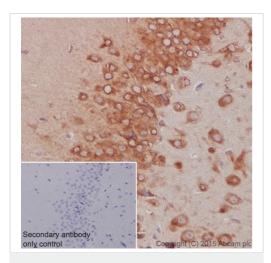
Immunohistochemical analysis of paraffin-embedded
Mouse cerebral cortex tissue labeling RCN1/RCN with ab210404 at
1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP)
(ab97051) at 1/500 dilution.

Cytoplasm staining on neurons of mouse cerebral cortex is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is **ab97051** at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RCN1/RCN antibody
[EPR17163-117] (ab210404)

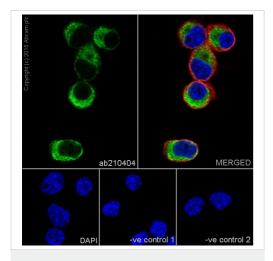
Immunohistochemical analysis of paraffin-embedded Rat hippocampus tissue labeling RCN1/RCN with ab210404 at 1/2000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Cytoplasm staining on neurons of rat hippocampus is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is **ab97051** at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Neuro-2a (Mouse neuroblastoma cell line) cells labeling RCN1/RCN with ab210404 at 1/500 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

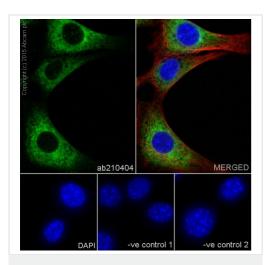
Confocal image showing cytoplasmic staining on Neuro-2a cell line.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse lgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

- -ve control 1: ab210404 at 1/500 dilution followed by **ab150120** at 1/1000 dilution.
- -ve control 2: $\underline{ab7291}$ at 1/1000 dilution followed by $\underline{ab150077}$ at 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling RCN1/RCN with ab210404 at 1/500 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor[®] 488) (**ab150077**) secondary antibody at 1/1000 dilution (green).

Confocal image showing cytoplasmic staining on NIH/3T3 cell line.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse lgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

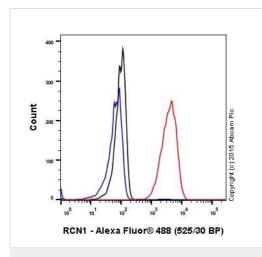
The negative controls are as follows:-

-ve control 1: ab210404 at 1/500 dilution followed by <u>ab150120</u> at 1/1000 dilution.

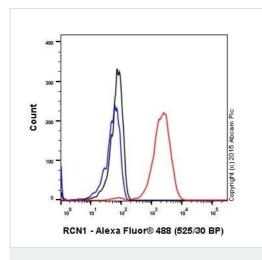
-ve control 2: $\underline{ab7291}$ at 1/1000 dilution followed by $\underline{ab150077}$ at 1/1000 dilution.

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling RCN1/RCNwith ab210404 at 1/700 dilution (red) compared with a Rabbit lgG,monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti Rabbit lgG (Alexa Fluor® 488) at 1/500 dilution was used as the secondary antibody.

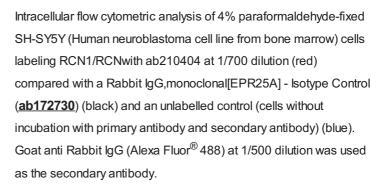
Cells were permeabilised with 90% methanol (diluted with 1X PBS).



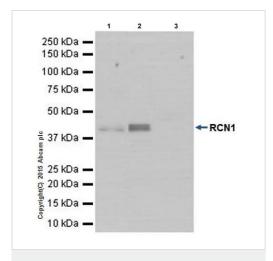
Flow Cytometry (Intracellular) - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)



Flow Cytometry (Intracellular) - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)



Cells were permeabilised with 90% methanol (diluted with 1X PBS).



Immunoprecipitation - Anti-RCN1/RCN antibody [EPR17163-117] (ab210404)

RCN1/RCN was immunoprecipitated from 1mg of 293T (Human epithelial cell line from embryonic kidney) whole cell lysate with ab210404 at 1/50 dilution.

Western blot was performed from the immunoprecipitate using ab210404 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/10000 dilution.

Lane 1: 293T whole cell lysate, 10µg (Input).

Lane 2: ab210404 IP in 293T whole cell lysate.

Lane 3: Rabbit IgG,monoclonal [EPR25A] - Isotype Control (ab172730) instead of ab210404 in 293T whole cell lysate.

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

Exposure time: 1 second.



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