abcam

Product datasheet

Anti-RBX1 antibody [EPR20185] ab221548



重组 RabMAb

16 图像

概述

产品名称 Anti-RBX1抗体[EPR20185]

描述 兔单克隆抗体[EPR20185] to RBX1

宿主 Rabbit

适用于: IHC-P, IP, ICC/IF, WB, Flow Cyt (Intra) 经测试应用

种属反应性 与反应: Mouse, Rat, Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: HeLa, HepG2, HT-1080, HEK-293, RAW 264.7, PC-12 and NIH/3T3 whole cell lysates;

> Human fetal heart and fetal kidney lysates; Mouse heart, kidney and spleen lysates; Rat brain, kidney and spleen lysates. IHC-P: Human colon, colon carcinoma, lung, lung carcinoma, gastric carcinoma and bladder cancer tissues; Mouse stomach tissue; Rat colon tissue. ICC/IF: HeLa

and NIH/3T3 cells. Flow Cyt (intra): HeLa and NIH/3T3 cells. IP: HeLa whole cell lysate.

常规说明 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**.

性能

形式 Liquid

存放说明 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.

存储溶液 pH: 7.2

Preservative: 0.01% Sodium azide

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

纯度 Protein A purified

单克隆 克隆

克隆编号 EPR20185

同种型 IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab221548于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
IHC-P		1/4000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		1/40.
ICC/IF		1/500.
WB		1/1000. Detects a band of approximately 12,11 kDa (predicted molecular weight: 12 kDa).
Flow Cyt (Intra)		1/60.

靶标

功能 E3 ubiquitin ligase component of multiple cullin-RING-based E3 ubiquitin-protein ligase

complexes which mediate the ubiquitination and subsequent proteasomal degradation of target proteins, including proteins involved in cell cycle progression, signal transduction, transcription and transcription-coupled nucleotide excision repair. The functional specificity of the E3 ubiquitin-protein ligase complexes depends on the variable substrate recognition components. As a component of the CSA complex promotes the ubiquitination of ERCC6 resulting in proteasomal degradation. Through the RING-type zinc finger, seems to recruit the E2 ubiquitination enzyme, like CDC34, to the complex and brings it into close proximity to the substrate. Probably also stimulates CDC34 autoubiquitination. May be required for histone H3 and histone H4 ubiquitination in response to ultraviolet and for subsequent DNA repair. Promotes the neddylation

of CUL1, CUL2, CUL4 and CUL4 via its interaction with UBE2M.

组织特异性 Widely expressed.

通路 Protein modification; protein ubiquitination.

序列相似性 Belongs to the RING-box family.

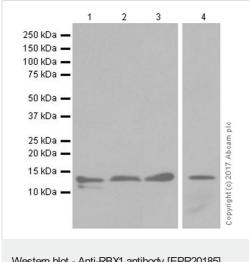
Contains 1 RING-type zinc finger.

结构域 The RING-type zinc finger domain is essential for ubiquitin ligase activity. It coordinates an

additional third zinc ion.

细胞定位 Cytoplasm. Nucleus.

图片



Western blot - Anti-RBX1 antibody [EPR20185] (ab221548)

All lanes : Anti-RBX1 antibody [EPR20185] (ab221548) at 1/5000 dilution

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

Lane 2: HepG2 (human liver hepatocellular carcinoma cell line) whole cell lysate

Lane 3: HT1080 (human fibrosarcoma cell line) whole cell lysateLane 4: HEK-293 (human epithelial cell line from embryonic kidney) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit $\lg G \ H\&L \ (HRP) \ (\underline{ab97051})$ at 1/100000 dilution

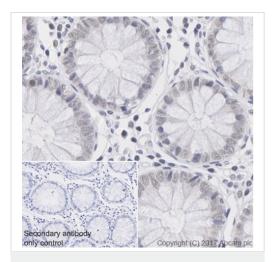
Developed using the ECL technique.

Predicted band size: 12 kDa **Observed band size:** 11,12 kDa

Exposure time: Lanes 1-3: 30 seconds; Lane 4: 10 seconds.

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been described in the literature (PMID: 22822056).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody
[EPR20185] (ab221548)

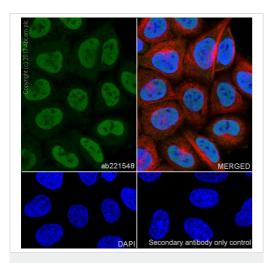
Immunohistochemical analysis of paraffin-embedded human colon tissue labeling RBX1 with ab221548 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear staining is observed on human colon tissue section.

As documented in the literature RBX1 has lower expression level in normal tissue compared to cancerous tissue (PMID:19509229).

Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

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

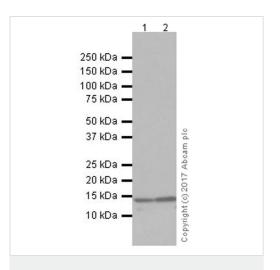


Immunocytochemistry/ Immunofluorescence - Anti-RBX1 antibody [EPR20185] (ab221548)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cells labeling RBX1 with ab221548 at 1/500 dilution followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear and weak cytoplasmic staining on HeLa cell line.

The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) (ab195889) (red) at 1/200 dilution.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution.



Western blot - Anti-RBX1 antibody [EPR20185] (ab221548)

All lanes : Anti-RBX1 antibody [EPR20185] (ab221548) at 1/1000 dilution

Lane 1 : Human fetal heart lysate

Lane 2: Human fetal kidney 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 1/4000 dilution

Developed using the ECL technique.

Predicted band size: 12 kDa Observed band size: 12 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

250 kDa -250 kDa -250 kDa -150 kDa -150 kDa -150 kDa -100 kDa -100 kDa -100 kDa -75 kDa -75 kDa -75 kDa -50 kDa -50 kDa -37 kDa -37 kDa -37 kDa -25 kDa 🕳 25 kDa -20 kDa -20 kDa -20 kDa -15 kDa 🕳 15 kDa -10 kDa -10 kDa -10 kDa -

Western blot - Anti-RBX1 antibody [EPR20185] (ab221548)

All lanes: Anti-RBX1 antibody [EPR20185] (ab221548) at 1/1000 dilution

Lane 1: Mouse heart lysate

Lane 2: Mouse kidney lysate

Lane 3: Mouse spleen lysate

Lane 4: Rat brain lysate

Lane 5: Rat kidney lysate

Lane 6: Rat spleen lysate

Lane 7: RAW 264.7 (mouse macrophage cell line transformed

with Abelson murine leukemia virus) whole cell lysate

Lane 8: PC-12 (rat adrenal gland pheochromocytoma cell line)

whole cell lysate

Lane 9: NIH/3T3 (mouse embyro fibroblast cell line) whole cell

lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at

1/100000 dilution

Developed using the ECL technique.

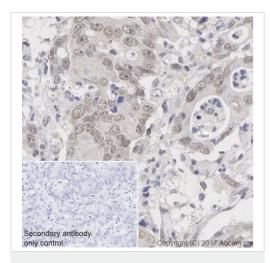
Predicted band size: 12 kDa Observed band size: 11,12 kDa

Exposure time: Lanes 1-6: 3 seconds; Lanes 7-8: 5 seconds.

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been

described in the literature (PMID: 22822056).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody
[EPR20185] (ab221548)

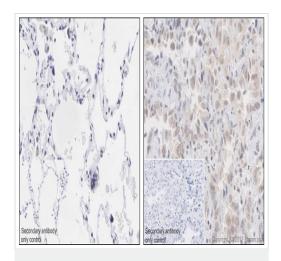
Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue labeling RBX1 with ab221548 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear and cytoplasmic staining is observed on human colon carcinoma tissue sections.

As documented in the literature RBX1 has higher expression level in cancerous tissue compared to normal tissue (PMID:19509229).

Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

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-RBX1 antibody
[EPR20185] (ab221548)

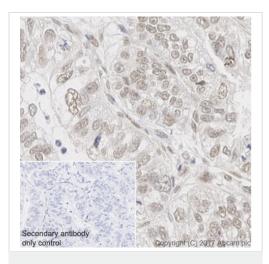
Immunohistochemical analysis of paraffin-embedded human lung and lung carcinoma tissues labeling RBX1 with ab221548 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Weak nuclear staining of RBX1 on the epithelium cells of human lung (left) compared to strong staining in human lung carcinoma (right).

As documented in the literature ROC1 has higher expression level in cancerous tissue compared to normal tissue (PMID:19509229).

Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

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



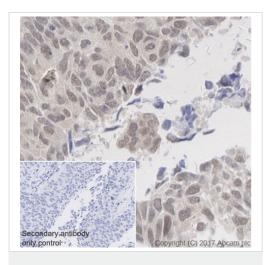
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody
[EPR20185] (ab221548)

Immunohistochemical analysis of paraffin-embedded human gastric carcinoma tissue labeling RBX1 with ab221548 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear and cytoplasmic staining on human gastric carcinoma tissue sections. The staining pattern observed is consistent with what has been described in the literature (PMID:24292229).

Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

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



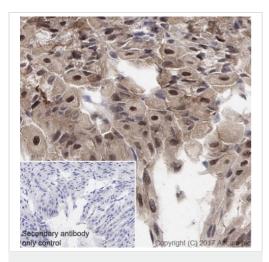
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody
[EPR20185] (ab221548)

Immunohistochemical analysis of paraffin-embedded human bladder cancer tissue labeling RBX1 with ab221548 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear and cytoplasmic staining on human bladder carcinoma tissue sections. The staining pattern observed is consistent with what has been described in the literature (PMID:23667514).

Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

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



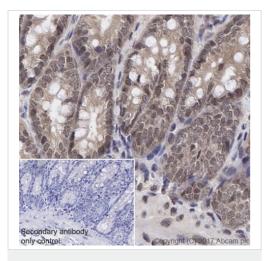
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody
[EPR20185] (ab221548)

Immunohistochemical analysis of paraffin-embedded mouse stomach tissue labeling RBX1 with ab221548 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear and cytoplasmic staining on mouse stomach tissue sections.

Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

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



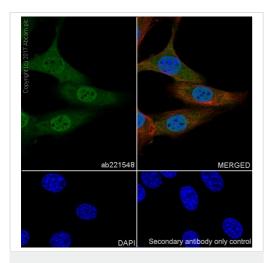
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RBX1 antibody
[EPR20185] (ab221548)

Immunohistochemical analysis of paraffin-embedded rat colon tissue labeling RBX1 with ab221548 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear and cytoplasmic staining on rat colon tissue sections.

Counter stained with hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

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

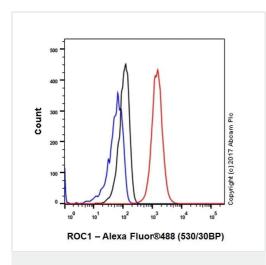


Immunocytochemistry/ Immunofluorescence - Anti-RBX1 antibody [EPR20185] (ab221548)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (mouse embyro fibroblast cell line) cells labeling RBX1 with ab221548 at 1/500 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (<u>ab150077</u>) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear and weak cytoplasmic staining on NIH/3T3 cell line.

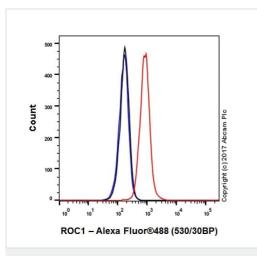
The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) (red) at 1/200 dilution.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution.



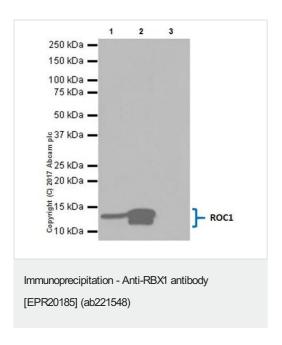
Flow Cytometry (Intracellular) - Anti-RBX1 antibody [EPR20185] (ab221548)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cell line labeling RBX1 with ab221548 at 1/600 dilution (red) compared with a Rabbit lgG, monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-RBX1 antibody [EPR20185] (ab221548)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized NIH/3T3 (mouse embyro fibroblast cell line) cell line labeling RBX1 with ab221548 at 1/60 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



RBX1 was immunoprecipitated from 0.35 mg of HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab221548 at 1/40 dilution. Western blot was performed from the immunoprecipitate using ab221548 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

Lane 1: HeLa whole cell lysate 10 µg (Input).

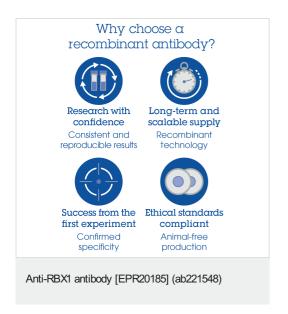
Lane 2: ab221548 IP in HeLa whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab221548 in HeLa whole cell lysate.

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

Exposure time: 30 seconds.

The molecular weight observed is consistent with what has been described in the literature (PMID: 22822056).



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