abcam

Product datasheet

Anti-PHAPI2 / APRIL antibody [EPR14588] ab200836

敲除验证 重组 RabMAb

5 References 14 图像

概述

产品名称	Anti-PHAPI2 / APRIL抗体[EPR14588]
描述	兔单克隆抗体[EPR14588] to PHAPI2 / APRIL
宿主	Rabbit
经测试应用	**适用于:** IHC-P, WB, Flow Cyt (Intra), ICC/IF
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: HEK293T, PC-3, LNCap, Jurkat, C6, RAW 264.7, PC-12, and NIH/3T3 whole cell lysates; Human fetal heart and fetal kidney lysates. IHC-P: Human prostatic hyperplasia, Human spleen, mouse spleen and rat cardiac muscle tissues. ICC/IF: Raw264.7 and Jurkat cells. Flow Cyt (intra): Jurkat cells.
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: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR14588

同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用**ab200836**于以下的**经测试应用

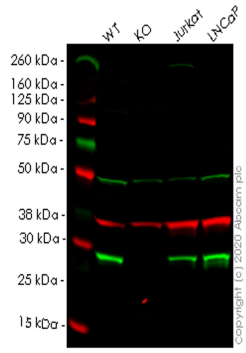
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
IHC-P		1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/1000. Detects a band of approximately 29 kDa (predicted molecular weight: 29 kDa).
Flow Cyt (Intra)		1/180.
ICC/IF		1/500.

靶标

功能	Multifunctional protein working as a cell cycle progression factor as well as a cell survival factor. Required for the progression from the G1 to the S phase. Anti-apoptotic protein which functions as a caspase-3 inhibitor. Has no phosphatase 2A (PP2A) inhibitor activity (By similarity). Exhibits histone chaperone properties, stimulating core histones to assemble into a nucleosome.
组织特异性	Expressed in heart, lung, pancreas, prostate and in spleen, thymus and placenta.
序列相似性	Belongs to the ANP32 family. Contains 4 LRR (leucine-rich) repeats. Contains 1 LRRCT domain.
结构域	Histone binding is mediated by the concave surface of the LRR region.
翻译后修饰	Some glutamate residues are glycylated by TTLL8. This modification occurs exclusively on glutamate residues and results in a glycine chain on the gamma-carboxyl group.
细胞定位	Nucleus. Accumulates in the nuclei at the S phase and Cytoplasm. Lacks a nuclear localization signal.

图片



Western blot - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

All lanes : Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate
Lane 2 : ANP32B knockout HEK293T cell lysate
Lane 3 : Jurkat cell lysate
Lane 4 : LNCaP cell lysate

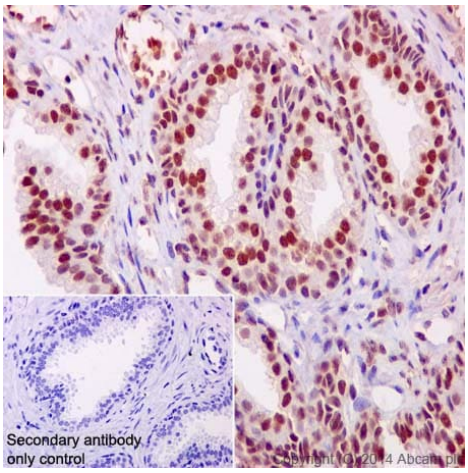
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

Predicted band size: 29 kDa
Observed band size: 29 kDa

Lanes 1-4: Merged signal (red and green). Green - ab200836 observed at 29 kDa. Red - loading control **ab8245** observed at 36 kDa.

ab200836 Anti-PHAPI2 / APRIL antibody [EPR14588] was shown to specifically react with PHAPI2 / APRIL in wild-type HEK293T cells. Loss of signal was observed when knockout cell line **ab266818** (knockout cell lysate **ab257831**) was used. Wild-type and PHAPI2 / APRIL knockout samples were subjected to SDS-PAGE. ab200836 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

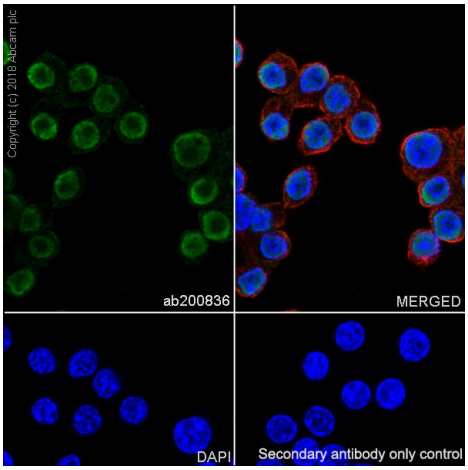


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Immunohistochemical analysis of paraffin-embedded Human prostatic hyperplasia tissue labeling PHAPI2 / APRIL with ab200836 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nuclear staining on Human prostatic hyperplasia tissue is observed. Counter stained with Hematoxylin.

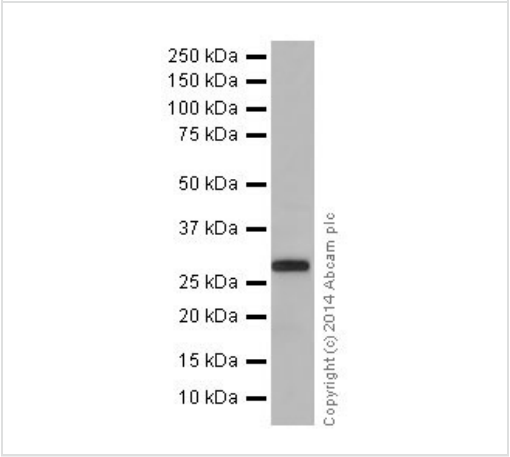
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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



Immunocytochemistry/ Immunofluorescence - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Raw264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) cells labeling PHAPI2 / APRIL with ab200836 at 1/250 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). The nuclear counter stain is DAPI (blue). Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and Ab195889 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/200 dilution (red). Confocal image showing mostly nuclear staining in Raw264.7 cell line.



Western blot - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836) at 1/1000 dilution + PC-3 (Human prostate cancer cell line) whole cell lysate at 10 µg

Secondary
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 29 kDa
Observed band size: 29 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

Western blot - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

All lanes : Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836) at 1/10000 dilution

Lane 1 : LNCaP (Human prostate cancer cell line) whole cell lysate
Lane 2 : Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysate

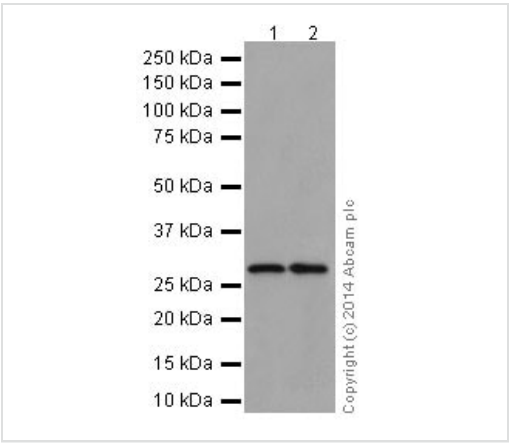
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 29 kDa
Observed band size: 29 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

All lanes : Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836) at 1/1000 dilution

Lane 1 : Human fetal heart lysate
Lane 2 : Human fetal kidney lysate

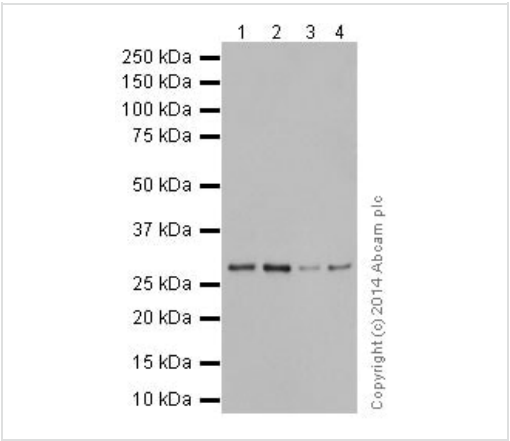
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 29 kDa
Observed band size: 29 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

All lanes : Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836) at 1/1000 dilution

Lane 1 : C6 (Rat glial tumor cells) whole cell lysate
Lane 2 : RAW 264.7 (Mouse macrophage cells transformed with Abelson murine leukemia virus) whole cell lysate
Lane 3 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysate
Lane 4 : NIH/3T3 (Mouse embyro fibroblast cells) whole cell lysate

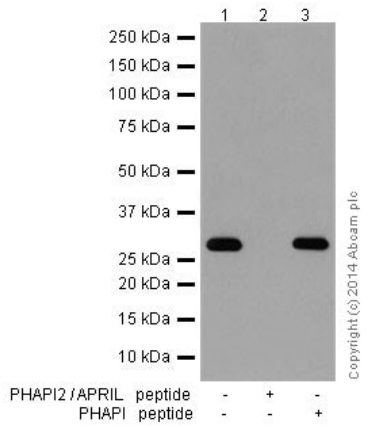
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 29 kDa
Observed band size: 29 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

All lanes : Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836) at 1/1000 dilution

Lane 1 : PC-3 (Human prostate cancer cell line) whole cell lysate
Lane 2 : PC-3 (Human prostate cancer cell line) whole cell lysate with PHAPI2 / APRIL peptide
Lane 3 : PC-3 (Human prostate cancer cell line) whole cell lysate with PHAPI peptide

Lysates/proteins at 10 µg per lane.

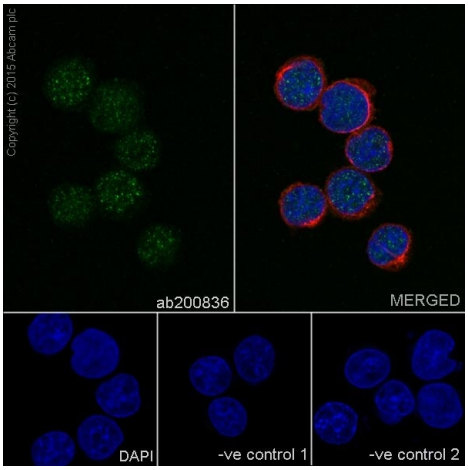
Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 29 kDa
Observed band size: 29 kDa

Exposure time: 1 minute

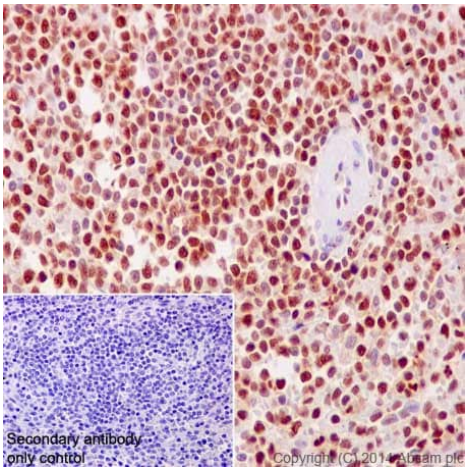
Blocking/Dilution buffer: 5% NFDM/TBST.

Based on sequence analysis ab200836 has 78% homology with PHAPI protein. The levels of XR were tested in the accompanying blocking experiment.



Immunocytochemistry/ Immunofluorescence - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling PHAPI2 / APRIL with ab200836 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/500 dilution (green). Nuclear and cytoplasmic staining on Jurkat cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows;
-ve control 1: ab200836 at 1/500 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.

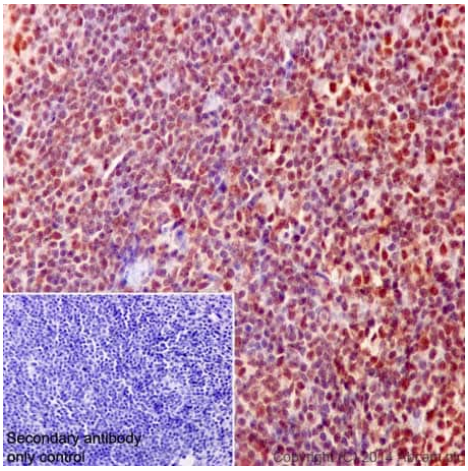


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Immunohistochemical analysis of paraffin-embedded Human spleen tissue labeling PHAPI2 / APRIL with ab200836 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nuclear staining on Human spleen tissue is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

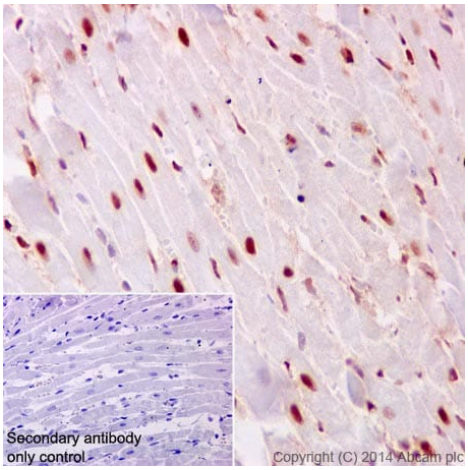


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labeling PHAPI2 / APRIL with ab200836 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nuclear staining on Mouse spleen tissue is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

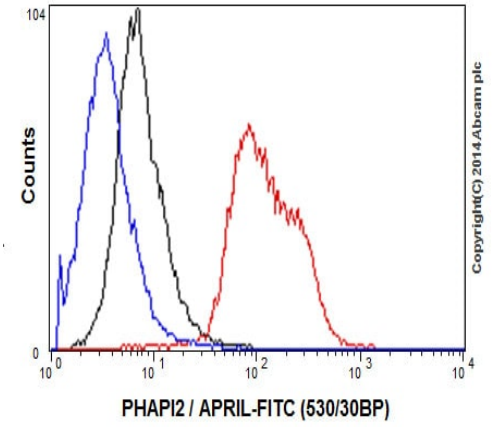


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Immunohistochemical analysis of paraffin-embedded Rat cardiac muscle tissue labeling PHAPI2 / APRIL with ab200836 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nuclear staining on Rat cardiac muscle tissue is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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



Flow Cytometry (Intracellular) - Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

Intracellular flow cytometric analysis of 2% paraformaldehyde-fixed Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling PHAPI2 / APRIL with ab200836 at 1/180 dilution (red) compared with a rabbit monoclonal IgG isotype control (**ab172730**;black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (FITC) at 1/150 dilution was used as the secondary antibody.

Anti-PHAPI2 / APRIL antibody [EPR14588] (ab200836)

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