abcam

Product datasheet

Anti-Annexin A1/ANXA1 antibody [EPR19342] ab214486

敲除 验证 重组 RabMAb

★★★★★ 11 Abreviews 24 References 17 图像

概述

| 产 品名称 | Anti-Annexin A1/ANXA1 抗体 [EPR19342] |
|----------------------|---|
| 描述 | 兔单 克隆抗体 [EPR19342] to Annexin A1/ANXA1 |
| 宿主 | Rabbit |
| 经测试应 用 | 适用于: WB, IHC-P, ICC/IF, IP, Flow Cyt (Intra) |
| 种属反应性 | 与反应: Mouse, Rat, Human |
| | 预测可用于: Common marmoset 🛛 🕰 |
| 免疫原 | Recombinant fragment. This information is proprietary to Abcam and/or its suppliers. |
| 阳性对照 | WB: K562, BxPC-3, C2C12, C6, PC-12 and NIH/3T3 whole cell lysates; human fetal brain, fetal kidney and placenta lysates; mouse and rat kidney and spleen lysates. IHC-P: Human tonsil, breast, endometrial cancer and bladder cancer tissues; rat lung tissue; mouse spleen tissue. ICC/IF: BxPC-3, NIH/3T3, HeLa cells. ICC/IF KO: Hap1 cells (Hap1-ANXA1 KO used as a negative cell line) Flow Cyt (intra): NIH/3T3 cells. IP: K562 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: PBS, 40% Glycerol, 0.05% BSA |
| 纯 度 | Protein A purified |

| 克隆 | 单 克隆 |
|--------------|-------------|
| 克隆 编号 | EPR19342 |
| 同种型 | lgG |

应用

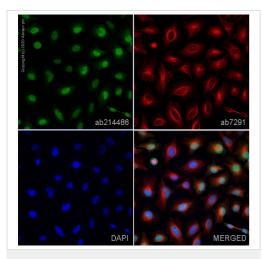
The Abpromise guarantee Abpromise ™承诺保证使用ab214486于以下的经测试应用

"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 |
|------------------|----------------------|--|
| WB | ★ ★ ★ ★ ★ <u>(1)</u> | 1/2000. Detects a band of approximately 37, 33 kDa (predicted molecular weight: 38 kDa). |
| ІНС-Р | ★ ★ ★ ★ ★ <u>(8)</u> | 1/4000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |
| ICC/IF | ★ ★ ★ ★ ★ <u>(2)</u> | Use a concentration of 1 - 5 μ g/ml. |
| IP | | 1/30. |
| Flow Cyt (Intra) | | 1/600. <u>ab172730</u> - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. |

| 靶 标 | |
|------------------------|--|
| 功能 | Calcium/phospholipid-binding protein which promotes membrane fusion and is involved in exocytosis. This protein regulates phospholipase A2 activity. It seems to bind from two to four calcium ions with high affinity. |
| 序列相似性 | Belongs to the annexin family. Contains 4 annexin repeats. |
| 结 构域 | A pair of annexin repeats may form one binding site for calcium and phospholipid. |
| 翻 译 后修 饰 | Phosphorylated by protein kinase C, epidermal growth factor receptor/kinase and TRPM7. Phosphorylation results in loss of the inhibitory activity. |
| 细胞定位 | Nucleus. Cytoplasm. Cell projection > cilium. Basolateral cell membrane. Found in the cilium, nucleus and basolateral cell membrane of ciliated cells in the tracheal endothelium (By similarity). Found in the cytoplasm of type II pneumocytes and alveolar macrophages. |

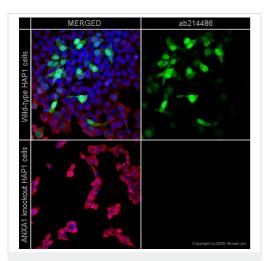
图片



Immunocytochemistry/ Immunofluorescence - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

ab214486 staining ANXA1 in HeLa cells. The cells were fixed with 4% paraformaldehyde (10min) permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab214486 at 5ug/ml concentration and **ab7291** (Tubulin) at 1/1000 dilution overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit lgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse lgG (Alexa Fluor® 594) (**ab150120**) at 2 µg/ml (shown in pseudo color red). Nuclear DNA was labelled in blue with DAPI.

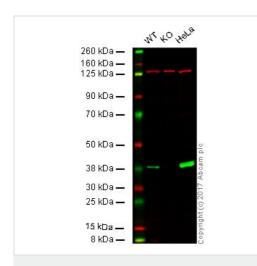
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



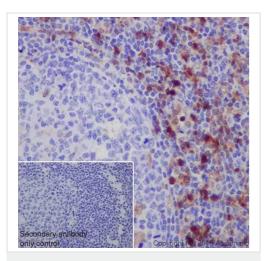
Immunocytochemistry/ Immunofluorescence - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

ab214486 staining ANXA1 in wild-type Hap1 cells (top panel) and ANXA1 knockout Hap1 cells (bottom panel). The cells were fixed with 4% paraformaldehyde (10min) permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab214486 at 5ug/ml concentration and **ab7291** (Tubulin) at 1/1000 dilution overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit lgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green) and a goat secondary antibody to mouse lgG (Alexa Fluor® 594) (**ab150120**) at 2 µg/ml (shown in pseudo color red). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Western blot - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

Lane 1: Wild type HAP1 whole cell lysate (20 μg) Lane 2: Annexin A1 knockout HAP1 whole cell lysate (20 μg) Lane 3: HeLa whole cell lysate (20 μg)

Lanes 1 - 3: Merged signal (red and green). Green - ab214486 observed at 38 kDa. Red - loading control, <u>ab18058</u>, observed at 130 kDa.

ab214486 was shown to specifically react with Annexin A1 when Annexin A1 knockout samples were used. Wild-type and Annexin A1 knockout samples were subjected to SDS-PAGE. Ab214486 and **ab18058** (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/2000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye[®] 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye[®] 680RD) preabsorbed **ab216776** secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

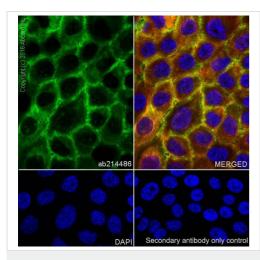
Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling Annexin A1 with ab214486 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Nuclear and cytoplasmic staining on human tonsil tissue is observed [PMID:9720986].

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 paraffinembedded sections) - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized BxPC-3 (Human pancreas adenocarcinoma cell line) cells labeling Annexin A1 with ab214486 at 1/1000 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 membrane and weak cytoplasmic staining on BxPC-3 cell line.

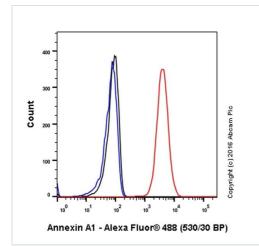
The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] -Microtubule Marker (Alexa Fluor[®] 594) (<u>ab195889</u>) at 1/200 dilution (red).

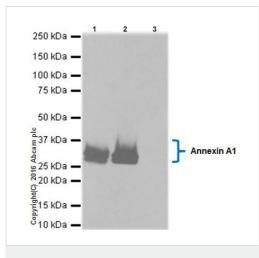
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (<u>ab150077</u>) at 1/1000 dilution.

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

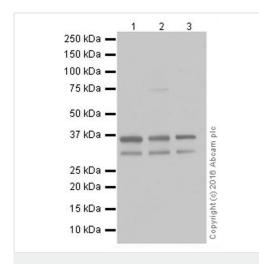
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling Annexin A1 with ab214486 at 1/600 dilution (red) compared withRabbit IgG, monoclonal [EPR25A] - Isotype Control (**ab172730**; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (Alexa Fluor[®] 488) at 1/2000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)



Immunoprecipitation - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)



Western blot - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

Annexin A1 was immunoprecipitated from 0.35 mg of K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate with ab214486 at 1/30 dilution.

Western blot was performed from the immunoprecipitate using ab214486 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>), was used for detection at 1/10,000 dilution

Lane 1: K562 whole cell lysate 10µg (Input).

Lane 2: ab214486 IP in K562 whole cell lysate.

Lane 3: Rabbit IgG, monoclonal [EPR25A] - Isotype Control (**ab172730**) instead of ab214486 in K562 whole cell lysate. Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second.

All lanes : Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) at 1/10000 dilution

Lane 1 : K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate
Lane 2 : BxPC-3 (Human pancreas adenocarcinoma cell line) whole cell lysate
Lane 3 : C2C12 (Mouse myoblast cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

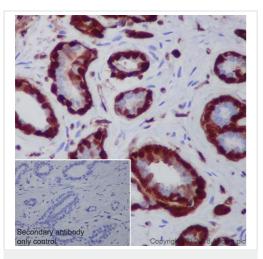
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 38 kDa Observed band size: 33,37 kDa

Exposure time: 1 second

Blocking/Dilution buffer: 5% NFDM/TBST.

The two band 37kDa is the full length band and the 33kDa supposed to be the cleavage form, this expression pattern observed is consistent with what has been described in the literature (PMID: 25510623 and 20679535).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

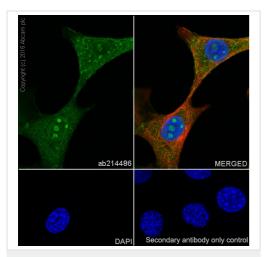
Immunohistochemical analysis of paraffin-embedded human breast tissue labeling Annexin A1 with ab214486 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Nuclear and cytoplasmic staining on human breast tissue is observed [PMID:16949910].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) 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-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling Annexin A1 with ab214486 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (**ab150077**) secondary antibody at 1/1000 dilution (green).

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

The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] -Microtubule Marker (Alexa Fluor[®] 594) (<u>ab195889</u>) at 1/200 dilution (red).

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

| | 1 | | 2 | | 3 | 4 | 5 | | 6 | 7 |
|----------|---|----------------------|---|-----------|---|------------|----------------|-----------|---|---|
| 50 kDa 🗕 | | 250 kDa 🗕 | | 250 kDa 🗕 | | 250 kDa 🗕 | 11. The second | 250 kDa 🗕 | | |
| 0 kDa 🗕 | | 150 kDa 🗕 | | 150 kDa 🗕 | | 150 kDa 🗕 | | 150 kDa 🗕 | | |
| 0 kDa 🗕 | | 100 kDa 🗕 | | 100 kDa 🗕 | | 100 kDa 🗕 | | 100 kDa 🗕 | | |
| 5 kDa 🗕 | | | | 75 kDa 🗕 | | 75 kDa 🗕 | | 75 kDa 🗕 | | |
| 0.400 | | 75 kDa 🗕 | | 50 kDa 🗕 | | 50 kDa 🗕 | | 10 100 - | | |
| 50 kDa 🗕 | | 50 kDa 🗕 | | 0710 | | | | 50 kDa 🗕 | | |
| 37 kDa 🗕 | - | | | 37 kDa 🗕 | 2 | 37 kDa 🗕 🔤 | - | 37 kDa 🗕 | - | |
| | - | 37 kDa 🗕 | - | | 1 | | - | | | ļ |
| 25 kDa 🗕 | | | - | 25 kDa 🗕 | | 25 kDa 🗕 | | 25 kDa 🗕 | | |
| 20 kDa 🗕 | | 25 kDa 🗕 | | 20 kDa 🗕 | | 20 kDa 🗕 | | 20 kDa 🗕 | | |
| 15 kDa 🗕 | | 20 kDa 🗕 | | 15 kDa 🗕 | | 15 kDa 🗕 | | 15 kDa 🗕 | | |
| 10 kDa 🗕 | | 15 kDa 🗕 10 kDa 🗕 | | 10 kDa 🗕 | | 10 kDa 🗕 | | 10 kDa 🗕 | | |

Western blot - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) Lanes 1-2 : Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) at 1/5000 dilution Lanes 3-7 : Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) at 1/2000 dilution

Lane 1: C6 (Rat glial tumor cell line) whole cell lysate

Lane 2 : PC-12 (Rat adrenal gland pheochromocytoma cell line) whole cell lysate

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

Lane 4 : Mouse kidney lysate

Lane 5 : Mouse spleen lysate

Lane 6 : Rat kidney lysate

Lane 7 : Rat spleen lysate

Lysates/proteins at 10 µg per lane.

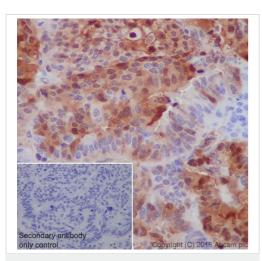
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

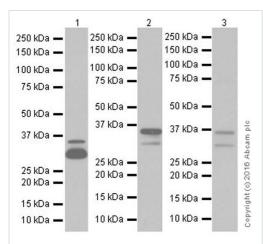
Predicted band size: 38 kDa Observed band size: 33,37 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 3 minutes; Lane 2/6/7: 1 seconds; Lane 3: 15 seconds; Lane 4/5: 30 seconds.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)



Western blot - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) Immunohistochemical analysis of paraffin-embedded human endometrial cancer tissue labeling Annexin A1 with ab214486 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Nuclear and cytoplasmic and weak membrane staining on human endometrial cancer 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) (<u>ab97051</u>) at 1/500 dilution.

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

Lanes 1-2 : Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) at 1/5000 dilution Lane 3 : Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) at 1/20000 dilution

Lane 1 : Human fetal brain lysate Lane 2 : Human fetal kidney lysate Lane 3 : Human placenta lysate

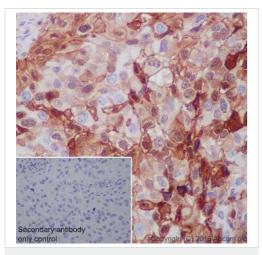
Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 38 kDa Observed band size: 33,37 kDa Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 3 minutes; Lane 2: 15 seconds; Lane 3: 1 second.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

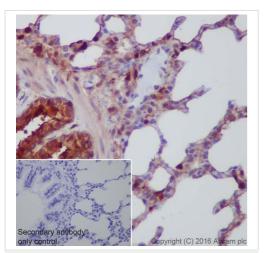
Immunohistochemical analysis of paraffin-embedded human bladder cancer tissue labeling Annexin A1 with ab214486 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Nuclear, cytoplasmic and membrane staining on human bladder cancer 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) (<u>ab97051</u>) 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-Annexin A1/ANXA1 antibody [EPR19342] (ab214486)

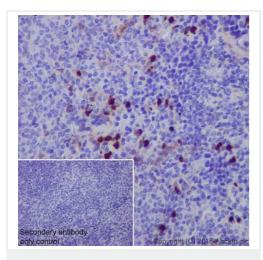
Immunohistochemical analysis of paraffin-embedded rat lung tissue labeling Annexin A1 with ab214486 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Nuclear and cytoplasmic staining on rat lung tissue is observed [PMID:15133855] [PMID:9720986].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) 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-Annexin A1/ANXA1 antibody [EPR19342] (ab214486) Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling Annexin A1 with ab214486 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

Nuclear and cytoplasmic staining on mouse spleen tissue is observed [PMID:9720986].

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.



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

• Replacement or refund for products not performing as stated on the datasheet

- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <u>https://www.abcam.cn/abpromise</u> or contact our technical team.

Terms and conditions

• Guarantee only valid for products bought direct from Abcam or one of our authorized distributors