# abcam

## Product datasheet

## Anti-Phosphotyrosine antibody [EPR16871] ab179530



重组 RabMAb

15 References 11 图像

概述

产品名称 Anti-Phosphotyrosine抗体[EPR16871]

描述 兔单克隆抗体[EPR16871] to Phosphotyrosine

宿主 Rabbit

经测试应用 适用于: Flow Cyt (Intra), WB, ICC/IF, IP, ELISA, Dot blot

种属反应性 与反应: Species independent

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

阳性对照 WB: Whole cell lysate from A431, L6 and NIH/3T3 cells treated with pervanadate. ICC/IF: C2C12

cells treated with Hydrogen peroxide. Flow Cyt (intra): A431 cells treated with pervanadate. IP:

Whole cell extract from A431 cells treated with pervanadate.

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 Preservative: 0.01% Sodium azide

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

纯度 Protein A purified

克隆 单克降

克隆编号 EPR16871

同种型 ΙgG

#### The Abpromise guarantee

#### Abpromise™承诺保证使用ab179530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。

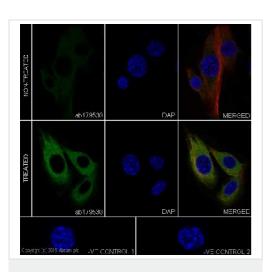
应用	Ab评论	说明
Flow Cyt (Intra)		1/160.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/1000.
ICC/IF		1/100.
IP		1/100.
ELISA		1/6400.
Dot blot		1/1000.

#### 靶标

#### 相关性

The phosphorylation of specific tyrosine residues has been shown to be a primary mechanism of signal transduction during normal mitogenesis, cell cycle progression and oncogenic transformation, its role in other areas such as differentiation and gap junction communication, is a matter of active and ongoing research. Antibodies that specifically recognize phosphorylated tyrosine residues have proved to be invaluable to the study of tyrosine phosphorylated proteins and the biochemical pathways in which they function.

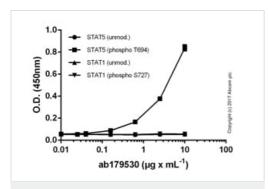
#### 图片



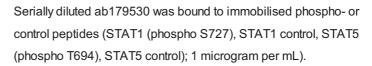
Immunocytochemistry/ Immunofluorescence - Anti-Phosphotyrosine antibody [EPR16871] (ab179530) Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized C2C12 (Mouse myoblast cell line) cells labeling Phosphotyrosine with ab179530 at 1/100 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/400 dilution (green). Cytoplasm staining on C2C12 cells is observed. The expression increased after treatment with H2O2 (2mM) for 10 minutes. The nuclear counterstain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/500 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:-

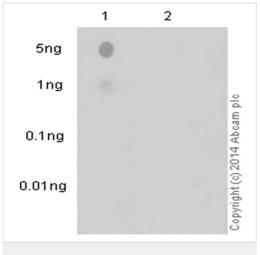
-ve control 1: - ab179530 at 1/100 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: - <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/500 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit IgG



ELISA - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)



The antibody was detected by Goat anti-Rabbit HRPO and signal was developed by TMB substrate.

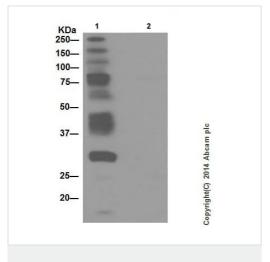


Dot Blot - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)

Dot blot analysis of INSR/IGF-1R (pY1009) phospho peptide (lane 1) and INSR/IGF-1R non-phospho peptide (lane 2) labelling Phosphotyrosine with ab179530 at a dilution of 1/1000. A peroxidase-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/2500).

Blocking and dilution buffer: 5% NFDM/TBST.

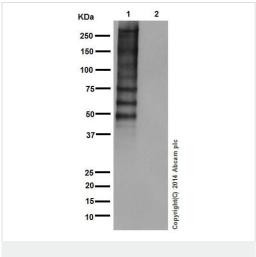
Exposure time: 3 minutes.



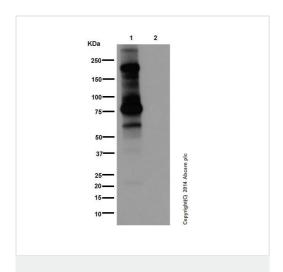
Immunoprecipitation - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)

Phosphotyrosine was immunoprecipitated from 1mg of A431 (Human epidermoid carcinoma) whole cell extract treated with 1mM pervanadate for 30 minutes with ab179530 at 1/100 dilution. Western blot was performed from the immunoprecipitate using ab179530 at 1/1000 dilution. Anti-Rabbit lgG (HRP), specific to the non-reduced form of lgG, was used as secondary antibody at 1/1500 dilution. Lane 1: A431 treated with 1mM pervanadate for 30 minutes whole cell extract. Lane 2: PBS instead of A431 whole cell extract.

Blocking and dilution buffer and concentration: 5% NFDM/TBST. Multiple bands represent phosph-tyrosine containing proteins precipitated and detected by ab179530.



Western blot - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)



Western blot - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)

**All lanes :** Anti-Phosphotyrosine antibody [EPR16871] (ab179530) at 1/1000 dilution

Lane 1 : Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysates treated with 1mM pervanadate for 20minutes

Lane 2 : Untreated Jurkat whole cell lysates

Lysates/proteins at 10 µg per lane.

#### **Secondary**

**All lanes :** Goat Anti-Rabbit lgG (H+L) Peroxidase conjugated at 1/1000 dilution

Multiple bands represent phosph-tyrosine containing proteins detected by ab179530

**All lanes :** Anti-Phosphotyrosine antibody [EPR16871] (ab179530) at 1/10000 dilution

**Lane 1 :** A431 (Human epidermoid carcinoma) whole cell lysates treated with 50mM pervanadate for 30 minutes

Lane 2: Untreated A431 whole cell lysates

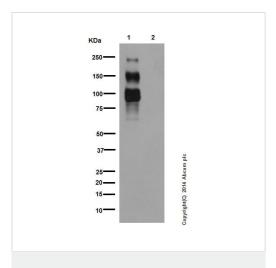
Lysates/proteins at 10 µg per lane.

#### **Secondary**

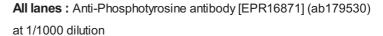
**All lanes :** Goat Anti-Rabbit lgG, (H+L),Peroxidase conjugated at 1/1000 dilution

Multiple bands represent phosph-tyrosine containing proteins detected by ab179530.

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)



Lane 1: NIH/3T3 (Mouse embyro fibroblast cells) whole cell lysates treated with 1mM pervanadate for 10 minutes

Lane 2: Untreated NIH/3T3 whole cell lysates

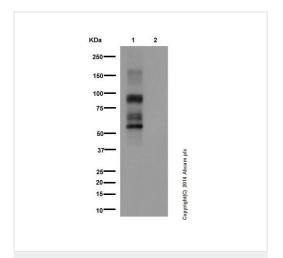
Lysates/proteins at 10 µg per lane.

#### **Secondary**

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

Multiple bands represent phosph-tyrosine containing proteins detected by ab179530.

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)

**All lanes :** Anti-Phosphotyrosine antibody [EPR16871] (ab179530) at 1/1000 dilution

**Lane 1**: L6 (Rat skeletal muscle cell line) whole cell lysates treated with 1mM pervanadate for 20 minutes

Lane 2: Untreated L6 whole cell lysates

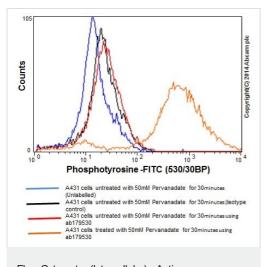
Lysates/proteins at 10 µg per lane.

## **Secondary**

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

Multiple bands represent phosph-tyrosine containing proteins detected by ab179530.

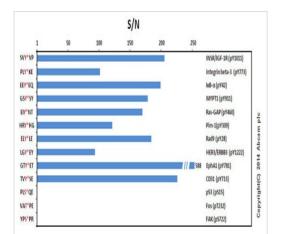
Blocking/Dilution buffer: 5% NFDM/TBST.



Intracellular flow cytometric analysis of 2% paraformaldehyde-fixed Pervanadate (50mM, 30min.) treated (orange)/untreated (red)A431 (Human epidermoid carcinoma) cells labeling Phosphotyrosine with ab179530 at 1/160 dilutioncompared with a rabbit monoclonal lgG control (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit lgG (FITC) at 1/150 dilution was used as the secondary antibody.

Flow Cytometry (Intracellular) - Anti-

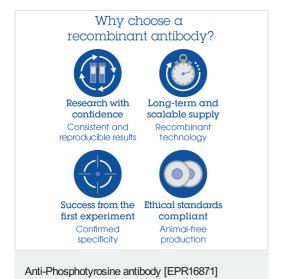
Phosphotyrosine antibody [EPR16871] (ab179530)



ELISA - Anti-Phosphotyrosine antibody [EPR16871] (ab179530)

ELISA analysis of various antigens (1  $\mu$ g/ml) using ab179530 at 1/6400 dilution followed by Alkaline Phosphatase-conjugated Goat Anti-Rabbit lgG (H+L) at 1/2500 dilution.

S/N = signal-to-noise ratio of phospho-versus nonphosphopeptides.



(ab179530)

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