# abcam

## Product datasheet

## Anti-PBK/SPK antibody [EPR21982] ab236871





重组 RabMAb

## 10 图像

#### 概述

产品名称 Anti-PBK/SPK抗体[EPR21982]

描述 兔单克隆抗体[EPR21982] to PBK/SPK

宿主 Rabbit

特异性 IHC is recommended for human only. 适用于: Flow Cyt (Intra), WB, IHC-P, IP 经测试应用

种属反应性 与反应: Rat, Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: Wild-type HAP1 cell lysate; HEK293T, HeLa, HepG2, A431, U-87 MG, C6 and PC-12 whole

cell lysates; Rat testis lysate. IHC-P: Human testis, gastric cancer and esophagus tissues. Flow

Cyt (intra): HeLa and HepG2 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: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

纯度 Protein A purified

克隆 单克隆

**克隆编号** EPR21982

**同种型** IgG

#### 应用

## The Abpromise guarantee Abpromise™承诺保证使用ab236871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)		1/50.
WB		1/1000. Detects a band of approximately 38 kDa (predicted molecular weight: 36 kDa).
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. IHC is recommended for human only.
IP		1/30.

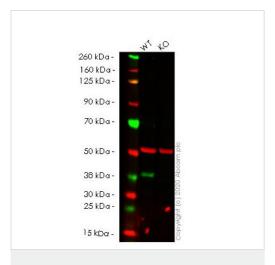
功能	Phosphorylates MAP kinase p38. Seems to be active only in mitosis. May also play a role in the activation of lymphoid cells. When phosphorylated, forms a complex with TP53, leading to TP53 destabilization and attenuation of G2/M checkpoint during doxorubicin-induced DNA damage.
组织 <b>特异性</b>	Expressed in the testis and placenta. In the testis, restrictedly expressed in outer cell layer of seminiferous tubules.
序列相似性	Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. MAP kinase kinase subfamily.

Contains 1 protein kinase domain.

翻译后修饰 Phosphorylated; in a cell-cycle dependent manner at mitosis.

图片

靶标



Western blot - Anti-PBK/SPK antibody [EPR21982] (ab236871)

**All lanes :** Anti-PBK/SPK antibody [EPR21982] (ab236871) at 1/1000 dilution

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

Lane 2: PBK knockout HEK-293T cell lysate

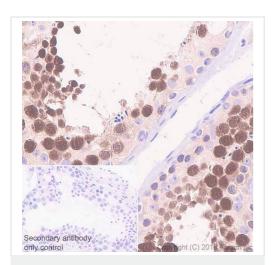
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 36 kDa **Observed band size:** 40 kDa

**Lanes 1-2:** Merged signal (red and green). Green - ab236871 observed at 40 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) observed at 50 kDa.

ab236871 was shown to react with PBK/SPK in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line <a href="mailto:ab266827">ab266827</a> (knockout cell lysate <a href="mailto:ab257575">ab257575</a>) was used. Wild-type HEK-293T and PBK knockout HEK-293T cell lysates were subjected to SDS-PAGE. ab236871 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (<a href="mailto:ab7291">ab7291</a>) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye®800CW) preadsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye®680RD) preadsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

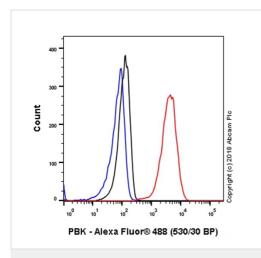


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PBK/SPK antibody
[EPR21982] (ab236871)

Immunohistochemical analysis of paraffin-embedded human testis tissue labeling PBK/SPK with ab236871 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining in germ cells of human testis (PMID:16982762; PMID:25909225) is observed. Counterstained with hematoxylin.

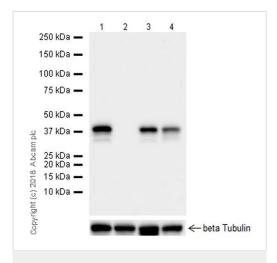
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

Heat mediated antigen retrieval using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0).



Flow Cytometry (Intracellular) - Anti-PBK/SPK antibody [EPR21982] (ab236871)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cell line labeling PBK/SPK with ab236871 at 1/50 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) (ab150077) at 1/2000 dilution was used as the secondary antibody.



Western blot - Anti-PBK/SPK antibody [EPR21982] (ab236871)

**All lanes :** Anti-PBK/SPK antibody [EPR21982] (ab236871) at 1/1000 dilution

Lane 1: Wild-type HAP1 cell lysate

Lane 2: PBK/SPK knockout HAP1 cell lysate

Lane 3 : HeLa (human epithelial cell line from cervix

adenocarcinoma) whole cell lysate

Lane 4 : HepG2 (human liver hepatocellular carcinoma cell line)

whole cell lysate

Lysates/proteins at 20 µg per lane.

#### **Secondary**

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

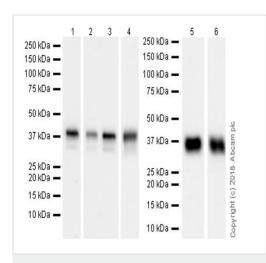
Predicted band size: 36 kDa Observed band size: 38 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been described in the literature (PMID:23547718; PMID:25909225).

ab236871 was shown to specifically react with PBK/SPK in wild-type HAP1 cells as signal was lost in PBK/SPK knockout cells. Wild-type and PBK/SPK knockout samples were subjected to SDS-PAGE. ab236871 and <a href="mailto:ab181602">ab181602</a> (Rabbit anti-GAPDH loading control) were incubated 1 hour at room temperature at 1/1000 dilution and 1/200,000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (<a href="mailto:ab97051">ab97051</a>) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-PBK/SPK antibody [EPR21982] (ab236871)

**All lanes :** Anti-PBK/SPK antibody [EPR21982] (ab236871) at 1/1000 dilution

Lane 1 : SW480 (human colorectal adenocarcinoma cell line) whole cell lysate at 20 µg

**Lane 2 :** A431 (human epidermoid carcinoma cell line) whole cell lysate at 20  $\mu g$ 

 $\textbf{Lane 3: } \text{U-87 MG (human glioblastoma-astrocytoma epithelial cell line) whole cell lysate at 20 ~\mu g}$ 

Lane 4: Rat testis lysate at 20 µg

**Lane 5**: C6 (rat glial tumor cell line) whole cell lysate at 10 μg **Lane 6**: PC-12 (rat adrenal gland pheochromocytoma cell line)
whole cell lysate at 10 μg

#### **Secondary**

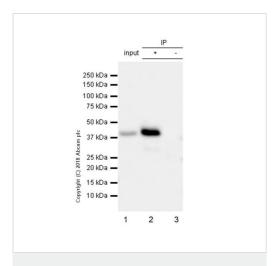
**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/100000 dilution

**Predicted band size:** 36 kDa **Observed band size:** 38 kDa

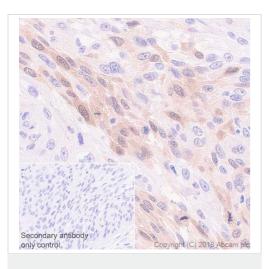
**Exposure time :** Lanes 1-3: 26 seconds; Lane 4: 8 seconds; Lanes 5-6: 70 seconds.

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been described in the literature (PMID:23547718; PMID:25909225).



Immunoprecipitation - Anti-PBK/SPK antibody [EPR21982] (ab236871)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PBK/SPK antibody
[EPR21982] (ab236871)

PBK/SPK was immunoprecipitated from 0.35 mg of HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab236871 at 1/30 dilution. Western blot was performed from the immunoprecipitate using <a href="mailto:ab184276">ab184276</a> at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (<a href="mailto:ab131366">ab131366</a>), was used for detection at 1/5000 dilution.

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

Lane 2: ab231871 IP in HeLa whole cell lysate.

**Lane 3:** Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab231871</u> in HeLa whole cell lysate.

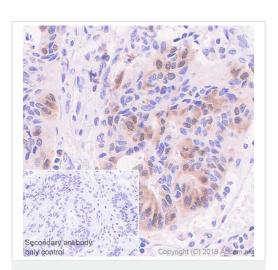
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds.

Immunohistochemical analysis of paraffin-embedded human esophagus cancer tissue labeling PBK/SPK with ab236871 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining in human esophagus cancer (PMID: 27919968) is observed. Counterstained 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.

Heat mediated antigen retrieval using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0).



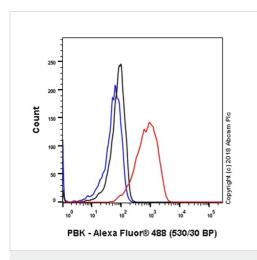
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PBK/SPK antibody
[EPR21982] (ab236871)

Immunohistochemical analysis of paraffin-embedded human gastric cancer tissue labeling PBK/SPK with ab236871 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining in human gastric cancer (PMID:26894977;

PMID:27898655) is observed. Counterstained with hematoxylin.

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

Heat mediated antigen retrieval using <u>ab93684</u> (Tris/EDTA buffer, pH 9.0).



Flow Cytometry (Intracellular) - Anti-PBK/SPK antibody [EPR21982] (ab236871)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized HepG2 (human liver hepatocellular carcinoma cell line) cell line labeling PBK/SPK with ab236871 at 1/50 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) (ab150077) at 1/2000 dilution was used as the secondary antibody.



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