abcam

Product datasheet

Anti-PBK/SPK antibody [EPR21983] - BSA and Azide free ab239760



Recombinant

RabMAb

9 Images

Overview

Product name Anti-PBK/SPK antibody [EPR21983] - BSA and Azide free

Description Rabbit monoclonal [EPR21983] to PBK/SPK - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IHC-P, IP

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control WB: HeLa, HepG2 and wild-type HAP1 cell lysate.

General notes ab239760 is the carrier-free version of ab236872.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar[®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.

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**.

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Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR21983

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab239760 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 38 kDa (predicted molecular weight: 36 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		Use at an assay dependent concentration.

Target

FunctionPhosphorylates 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.

Tissue specificity Expressed in the testis and placenta. In the testis, restrictedly expressed in outer cell layer of

seminiferous tubules.

Sequence similarities Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. MAP kinase kinase

subfamily.

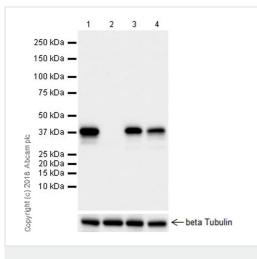
Contains 1 protein kinase domain.

Post-translational

modifications

Phosphorylated; in a cell-cycle dependent manner at mitosis.

Images



Western blot - Anti-PBK/SPK antibody [EPR21983] - BSA and Azide free (ab239760)

All lanes : Anti-PBK/SPK antibody [EPR21983] (<u>ab236872</u>) at 1/1000 dilution

Lane 1: Wild-type HAP1 cell lysate

Lane 2: PBK/SPK knockout HAP1 cell lysate

Lane 3: HeLa (human cervix adenocarcinoma epithelial cell),

whole cell lysate

Lane 4: HepG2 (human hepatocellular carcinoma epithelial cell),

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

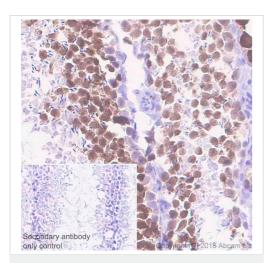
Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

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

ab236872 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. ab236872 and ab181602 (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 (ab97051) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide (ab236872).

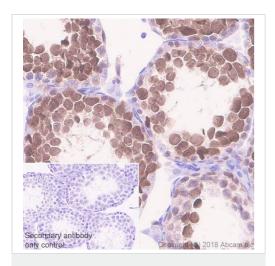


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PBK/SPK antibody
[EPR21983] - BSA and Azide free (ab239760)

Immunohistochemical analysis of paraffin-embedded rat testis tissue labeling PBK/SPK with <u>ab236872</u> at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining in germ cells of rat testis (PMID:16982762; PMID:25909225) is observed. Counterstained with hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab236872</u>).

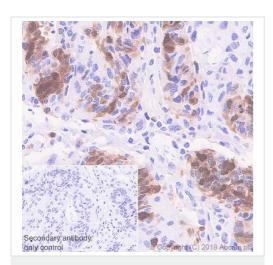


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PBK/SPK antibody
[EPR21983] - BSA and Azide free (ab239760)

Immunohistochemical analysis of paraffin-embedded mouse testis tissue labeling PBK/SPK with <u>ab236872</u> at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining in germ cells of mouse testis (PMID:16982762; PMID:25909225) is observed. Counterstained with hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab236872).

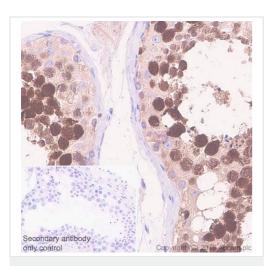


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PBK/SPK antibody
[EPR21983] - BSA and Azide free (ab239760)

Immunohistochemical analysis of paraffin-embedded human gastric cancer tissue labeling PBK/SPK with <u>ab236872</u> at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). 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 a ready to use Goat Anti-Rabbit IgG H&L (HRP).

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab236872).



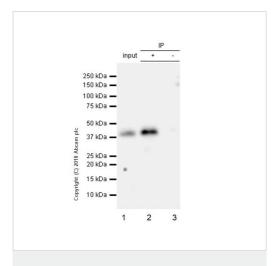
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PBK/SPK antibody

[EPR21983] - BSA and Azide free (ab239760)

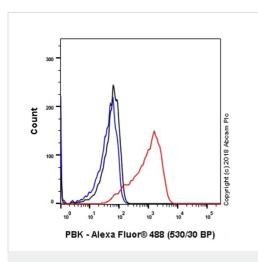
Immunohistochemical analysis of paraffin-embedded human testis tissue labeling PBK/SPK with <u>ab236872</u> at 1/4000 dilution, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Positive staining in germ cells of human testis (PMID:16982762; PMID:25909225) is observed. Counterstained with hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).

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

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab236872).



Immunoprecipitation - Anti-PBK/SPK antibody [EPR21983] - BSA and Azide free (ab239760)



Flow Cytometry (Intracellular) - Anti-PBK/SPK antibody [EPR21983] - BSA and Azide free (ab239760)

PBK/SPK was immunoprecipitated from 0.35 mg HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate with **ab236872** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab236872** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/5000 dilution.

Lane 1: HepG2 whole cell lysate 10 µg (Input).

Lane 2: ab236872 IP in HepG2 whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab236872</u> in HepG2 whole cell lysate.

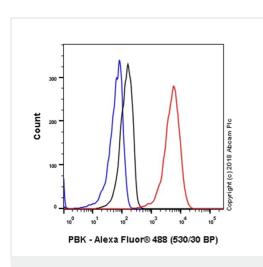
Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 10 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab236872).

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized HepG2 (human hepatocellular carcinoma epithelial cell) cell line labeling PBK/SPK with <u>ab236872</u> at 1/500 (red) compared with a Rabbit monoclonal lgG (<u>ab172730</u>) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit lgG (Alexa Fluor[®] 488, <u>ab150077</u>), at 1/2000 dilution was used as the secondary antibody.

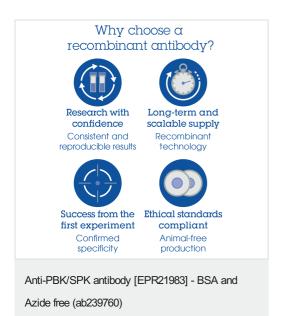
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab236872).



Flow Cytometry (Intracellular) - Anti-PBK/SPK antibody [EPR21983] - BSA and Azide free (ab239760)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized HeLa (human cervix adenocarcinoma epithelial cell) cell line labeling PBK/SPK with <u>ab236872</u> at 1/500 (red) compared with a Rabbit monoclonal IgG (<u>ab172730</u>) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor[®] 488, <u>ab150077</u>), at 1/2000 dilution was used as the secondary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab236872).



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