# abcam

## Product datasheet

## Anti-GSK3 beta (phospho S9) antibody [EPR2286Y] - BSA and Azide free ab219359

Recombinant RabMAb

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#### Overview

**Product name** Anti-GSK3 beta (phospho S9) antibody [EPR2286Y] - BSA and Azide free

**Description** Rabbit monoclonal [EPR2286Y] to GSK3 beta (phospho S9) - BSA and Azide free

**Host species** Rabbit

Suitable for: IHC-P, ICC/IF, WB, Dot blot **Tested applications** 

Unsuitable for: Flow Cyt or IP

Reacts with: Human Species reactivity

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control 293T cell lysates, untreated or treated with Calyculin A and human ovarian carcinoma tissue.

**General notes** ab219359 is the carrier-free version of ab75814.

> Our carrier-free 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 cellbased 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<sup>®</sup> 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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Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

#### **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 EPR2286Y

**Isotype** IgG

#### **Applications**

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab219359 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
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 47 kDa (predicted molecular weight: 47 kDa).
Dot blot		Use at an assay dependent concentration.

**Application notes** 

Is unsuitable for Flow Cyt or IP.

#### **Target**

### **Function**

Participates in the Wnt signaling pathway. Implicated in the hormonal control of several regulatory proteins including glycogen synthase, MYB and the transcription factor JUN. Phosphorylates JUN at sites proximal to its DNA-binding domain, thereby reducing its affinity for DNA. Phosphorylates MUC1 in breast cancer cells, and decreases the interaction of MUC1 with CTNNB1/beta-catenin. Phosphorylates CTNNB1/beta-catenin. Phosphorylates SNAI1. Plays an important role in ERBB2-dependent stabilization of microtubules at the cell cortex. Prevents the phosphorylation of APC and CLASP2, allowing its association with the cell membrane. In turn, membrane-bound APC allows the localization of MACF1 to the cell membrane, which is required for microtubule capture and stabilization. Phosphorylates MACF1 and this phosphorylation inhibits the binding of MACF1 to microtubules which is critical for its role in bulge stem cell migration and skin wound

repair.

**Tissue specificity** Expressed in testis, thymus, prostate and ovary and weakly expressed in lung, brain and kidney.

**Sequence similarities**Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. GSK-3 subfamily.

Contains 1 protein kinase domain.

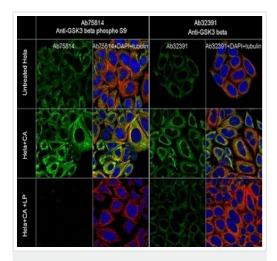
**Post-translational** Phosphorylated by AKT1 and ILK1. Activated by phosphorylation at Tyr-216. modifications

**Cellular localization**Cytoplasm. Nucleus. Cell membrane. The phosphorylated form shows localization to cytoplasm

and cell membrane. The MEMO1-RHOA-DIAPH1 signaling pathway controls localization of the

phosophorylated form to the cell membrane.

#### **Images**



Immunocytochemistry/ Immunofluorescence - Anti-GSK3 beta (phospho S9) antibody [EPR2286Y] -BSA and Azide free (ab219359) Cell line: HeLa (human cervix adenocarcinoma)

Target: Ab75814 anti-GSK3 beta (phospho S9)

Secondary: ab150077 AlexaFluor®488 Goat anti-Rabbit

Fixative: 4% PFA

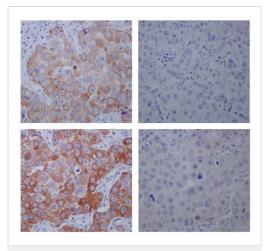
Permeabilization: 0.1% Triton-X

Nuclear counter stain: DAPI

Confocal image showed increased cytoplasmic staining after Calyculin A (CA) (<u>ab141784</u> 100ng/ml, 10min) treatment on Hela cells. The LP treatment decreased the increased cytoplasmic staining caused by CA.

Ab32391 was used as a control for <u>ab75814</u>. The results showed cytoplasmic staining on untreated, CA and CA+LP treated Hela cells.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab75814</u>).

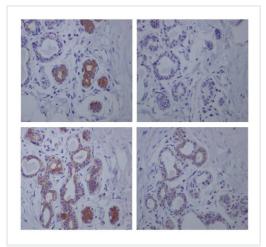


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GSK3 beta (phospho S9) antibody [EPR2286Y] - BSA and Azide free (ab219359)

Immunohistochemical analysis of paraffin-embedded human breast cancer tissue labelled with untreated <u>ab75814</u> (top-left) at a dilution of 1/1000, alkaline phosphatase treated <u>ab75814</u> (top-right) at a dilution of 1/1000, untreated <u>ab32391</u> (bottom-left) at a dilution of 1/1000 and alkaline phophatase treated <u>ab32391</u> (bottom-right) at a dilution of 1/1000. Ab97051 was used as secondary antibody at a dilution of 1/500 and counterstained with hematoxylin.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab75814).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

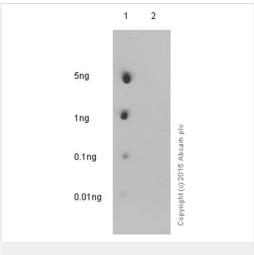


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GSK3 beta (phospho S9) antibody [EPR2286Y] - BSA and Azide free (ab219359)

Immunohistochemical analysis of paraffin-embedded human breast tissue labelled with untreated <a href="mailto:ab75814">ab75814</a> (phospho) (top-left) at a dilution of 1/1000, alkaline phosphatase treated <a href="mailto:ab75814">ab75814</a> (phospho) (top-right) at a dilution of 1/1000, untreated <a href="mailto:ab32391">ab32391</a> (bottom-left) at a dilution of 1/1000 and alkaline phophatase treated <a href="mailto:ab32391">ab32391</a> (bottom-right) at a dilution of 1/1000. Ab97051 was used as secondary antibody at a dilution of 1/500 and counterstained with hematoxylin.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab75814</u>).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



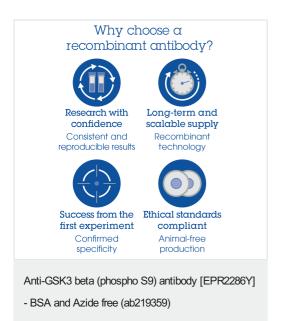
Dot Blot - Anti-GSK3 beta (phospho S9) antibody [EPR2286Y] - BSA and Azide free (ab219359)

Dot blot analysis of GSK3 beta (pS9) peptide (Lane 1) and GSK3 beta non-phospho peptide (Lane 2) labelling GSK3 (pS9) with purified <u>ab75814</u> at a dilution of 1/1000. <u>ab97051</u> (Peroxidase conjugated goat anti-rabbit lgG (H+L)) was used as the secondary antibody at a dilution of 1/100000.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab75814).



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

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