abcam

ab32391

Anti-GSK3 beta antibody [Y174]

Anti-GSK3 beta antibody [Y174] (ab32391) is a rabbit monoclonal antibody detecting GSK3 beta in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF, ELISA**. Suitable for **Human,**.

- KO validated for confirmed specificity
- Biophysical QC for unrivalled batch-batch consistency
- Over 200 publications
- Trusted since 2006

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	Y174
Purification technique	Affinity purification Protein A
Specificity	This antibody is specific for human GSK3 beta. It may also detect the splice isoform 2 based on sequence homology. The immunogen used for this antibody is GSK3 beta phospho S9. This antibody shows partially phospho specificity to phospho S9 under certain conditions, for example, under low peptide concentration in ELISA assay, it has dominant reactivity with phospho S9 peptide.
Concentration	0.542 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	-
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Predicted

Species	Mouse
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide - Human
Dilution info	-
Notes	-

ELISA

Tested

Species	Synthetic peptide - Human
Dilution info	-
Notes	Unit Type: 0 - 1000 ng/ml

Expected

Species	Human
Dilution info	Use at an assay dependent concentration.

Notes	-

Predicted

Species	Mouse
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/5000 - 1/10000
Notes	-

Predicted

Species	Mouse
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide - Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100 - 1/500
Notes	-

Predicted

Species	Mouse
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide - Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/100
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Predicted

Species	Mouse
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide - Human
Dilution info	-
Notes	-

Target data

See full target information GSK3B

Function

Constitutively active protein kinase that acts as a negative regulator in the hormonal control of glucose homeostasis, Wnt signaling and regulation of transcription factors and microtubules, by phosphorylating and inactivating glycogen synthase (GYS1 or GYS2), EIF2B, CTNNB1/beta-catenin, APC, AXIN1, DPYSL2/CRMP2, JUN, NFATC1/NFATC, MAPT/TAU and MACF1 (PubMed:11430833, PubMed:12554650, PubMed:14690523, PubMed:16484495, PubMed:1846781, PubMed:20937854, PubMed:9072970). Requires primed phosphorylation of the majority of its substrates (PubMed:11430833, PubMed:16484495). In skeletal muscle, contributes to insulin regulation of glycogen synthesis by phosphorylating and inhibiting GYS1 activity and hence glycogen synthesis (PubMed:8397507). May also mediate the development of insulin resistance by regulating activation of transcription factors (PubMed:8397507). Regulates protein synthesis by controlling the activity of initiation factor 2B (EIF2BE/EIF2B5) in the same manner as glycogen synthase (PubMed:8397507). In Wnt signaling, GSK3B forms a multimeric complex with APC, AXIN1 and CTNNB1/beta-catenin and phosphorylates the N-terminus of CTNNB1 leading to its degradation mediated by ubiquitin/proteasomes (PubMed:12554650). Phosphorylates JUN at sites proximal to its DNA-binding domain, thereby reducing its affinity for DNA (PubMed:1846781). Phosphorylates NFATC1/NFATC on conserved serine residues promoting NFATC1/NFATC nuclear export, shutting off NFATC1/NFATC gene regulation, and thereby opposing the action of calcineurin (PubMed:9072970). Phosphorylates MAPT/TAU on 'Thr-548', decreasing significantly MAPT/TAU ability to bind and stabilize microtubules (PubMed:14690523). MAPT/TAU is the principal component of neurofibrillary tangles in Alzheimer disease (PubMed:14690523). Plays an important role in ERBB2-dependent stabilization of microtubules at the cell cortex (PubMed:20937854). Phosphorylates MACF1, inhibiting its binding to microtubules which is critical for its role in bulge stem cell migration and skin wound repair (By similarity). Probably regulates NF-kappa-B (NFKB1) at the transcriptional level and is required for the NF-kappa-B-mediated anti-apoptotic response to TNF (TNF/TNFA) (By similarity). Negatively regulates replication in pancreatic beta-cells, resulting in apoptosis, loss of beta-cells and diabetes (By similarity). Through phosphorylation of the anti-apoptotic protein MCL1, may control cell apoptosis in response to growth factors deprivation (By similarity). Phosphorylates MUC1 in breast cancer cells, decreasing the interaction of MUC1 with CTNNB1/beta-catenin (PubMed:9819408). Is necessary for the establishment of neuronal polarity and axon outgrowth (PubMed:20067585). Phosphorylates MARK2, leading to inhibition of its activity (By similarity). Phosphorylates SIK1 at 'Thr-182', leading to sustainment of its activity (PubMed:18348280). Phosphorylates ZC3HAV1 which enhances its antiviral activity (PubMed:22514281). Phosphorylates SNAI1, leading to its ubiquitination and proteasomal degradation (PubMed:15448698, PubMed:15647282, PubMed:25827072, PubMed:29059170). Phosphorylates SFPQ at 'Thr-687' upon T-cell activation (PubMed:20932480). Phosphorylates NR1D1 st 'Ser-55' and 'Ser-59' and stabilizes it by protecting it from proteasomal degradation. Regulates the circadian clock via phosphorylation of the major clock components including BMAL1, CLOCK and PER2 (PubMed:19946213, PubMed:28903391). Phosphorylates FBXL2 at 'Thr-404' and primes it for ubiquitination by the SCF(FBXO3) complex and proteasomal degradation (By similarity). Phosphorylates CLOCK AT 'Ser-427' and targets it for proteasomal degradation (PubMed:19946213). Phosphorylates BMAL1 at 'Ser-17' and 'Ser-21' and primes it for ubiquitination and proteasomal degradation (PubMed:28903391). Phosphorylates OGT at 'Ser-3' or 'Ser-4' which positively regulates its activity. Phosphorylates MYCN in neuroblastoma cells which may promote its degradation (PubMed:24391509). Regulates the circadian rhythmicity of hippocampal long-term potentiation and BMAL1 and PER2 expression (By similarity). Acts as a regulator of

autophagy by mediating phosphorylation of KAT5/TIP60 under starvation conditions, activating KAT5/TIP60 acetyltransferase activity and promoting acetylation of key autophagy regulators, such as ULK1 and RUBCNL/Pacer (PubMed:30704899). Negatively regulates extrinsic apoptotic signaling pathway via death domain receptors. Promotes the formation of an anti-apoptotic complex, made of DDX3X, BRIC2 and GSK3B, at death receptors, including TNFRSF10B. The anti-apoptotic function is most effective with weak apoptotic signals and can be overcome by stronger stimulation (PubMed:18846110). Phosphorylates E2F1, promoting the interaction between E2F1 and USP11, stabilizing E2F1 and promoting its activity (PubMed:17050006, PubMed:28992046). Phosphorylates mTORC2 complex component RICTOR at 'Ser-1235' in response to endoplasmic stress, inhibiting mTORC2 (PubMed:21343617). Phosphorylates mTORC2 complex component RICTOR at 'Thr-1695' which facilitates FBXW7-mediated ubiquitination and subsequent degradation of RICTOR (PubMed:25897075). Phosphorylates FXR1, promoting FXR1 ubiquitination by the SCF(FBXO4) complex and FXR1 degradation by the proteasome (By similarity). Phosphorylates interleukin-22 receptor subunit IL22RA1, preventing its proteasomal degradation (By similarity). Phosphorylates and inhibits the CTP synthase and protein-asparagine deamidase activities of CTPS1 (PubMed:17681942). Phosphorylates DSP at multiple sequential serine residues in the C-terminus tail, promoting its recruitment to developing desmosome cell-cell junctions (PubMed:25733715).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-GSK3 beta antibody [Y174] (ab32391) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF), ELISA in Human, samples.

What is the molecular weight of GSK3 beta?
Anti-GSK3 beta [Y174] (ab32391) specifically detects a band for GSK3 beta (UniProt: P49841) at a molecular weight of 46kDa.

Trusted by the scientific community
Anti-GSK3 beta [Y174] (ab32391) was first used in a scientific publication in 2006 and has been cited over 200 times in

peer-reviewed journals.

Specificity confirmed
The specificity of Anti-GSK3 beta antibody [Y174] (ab32391) has been confirmed by Western blot testing in GSK3B Knockout HAP1 cells.

Other related products
We have a range of other formats of antibody clone [Y174] also available for your convenience: ab32391, Carrier free - ab183177, Alexa Fluor® 647 - ab196910, HRP - ab196911, Alexa Fluor® 488 - ab197236, Alexa Fluor® 594 - ab201737, Alexa Fluor® 555 - ab201738, PE - ab210619

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Glycogen synthase kinase 3 beta or GSK3 beta is a serine/threonine protein kinase that regulates various cellular processes. It is also referred to as GSK3B and has a molecular weight of approximately 47 kilodaltons (kDa). This enzyme is widely expressed in many tissues with notably high expression levels in the brain. Mechanically GSK3 beta phosphorylates target proteins affecting their activity stability and interactions with other proteins.
Biological function summary	As an important regulatory protein GSK3 beta plays a role in multiple cellular functions including metabolism cell cycle and cell signaling. It often interacts with components in signaling complexes and modulates the activity of these complexes involving phosphorylation. It is involved in the regulation of transcription factors such as c-Myc and B-catenin influencing cell fate and survival. This ability to modulate diverse functions highlights GSK3 beta's importance in cellular regulation.
Pathways	GSK3 beta participates in significant signaling pathways such as the Wnt/B-catenin and insulin signaling pathways. In the Wnt pathway GSK3 beta phosphorylates B-catenin marking it for degradation which regulates gene transcription. In insulin

signaling it interacts with proteins like AKT to mediate glycogen synthesis. These interactions highlight its role in maintaining cellular homeostasis and energy balance.

Associated diseases and disorders

GSK3 beta has implications in Alzheimer's disease and type 2 diabetes. It is associated with tau protein hyperphosphorylation and neurofibrillary tangles in Alzheimer's disease. In diabetes its role in insulin signaling impacts glucose metabolism. Further GSK3 beta's dysfunction relates to disrupted signaling related to these conditions positioning it as a potential therapeutic target.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

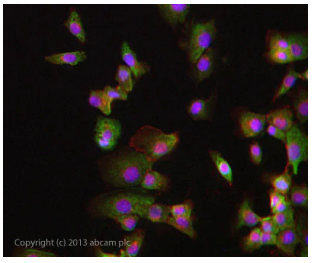
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

11 product images

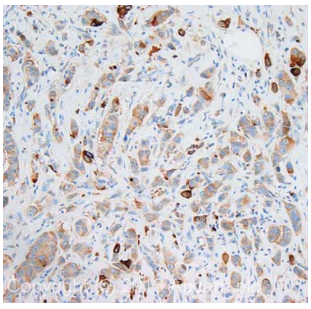


Immunocytochemistry/ Immunofluorescence - Anti-GSK3 beta antibody [Y174] (ab32391)

GSK3 beta Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-GSK3 beta antibody

ICC/IF image of ab32391 stained DU145 cells. The cells were 4% formaldehyde fixed (10 minutes) and then incubated in 1 %BSA / 10 % normal goat serum / 0.3 M glycine in 0.1 % PBS-Tween for 1 hour to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab32391 at 1/200 dilution overnight at +4°C. The secondary

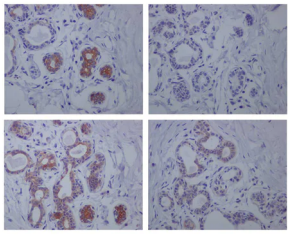
antibody (green) was DyLight® 488 goat anti- rabbit (ab96899) IgG (H+L) used at a 1/250 dilution for 1 hour. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1 hour. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 µM.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GSK3 beta antibody [Y174] (ab32391)

GSK3 beta Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-GSK3 beta antibody

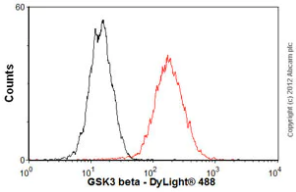
IHC image of GSK3 staining in human breast adenocarcinoma formalin fixed paraffin embedded tissue section, performed on a Leica Bond system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6, epitope retrieval solution 1) for 20 minutes. The section was then incubated with ab32391, 1/200 diution, for 15 minutes at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GSK3 beta antibody [Y174] (ab32391)

GSK3 beta Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-GSK3 beta antibody

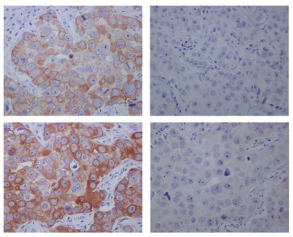
Immunohistochemical analysis of paraffin-embedded human breast tissue labelled with untreated ab75814 (phospho) (top-left) at a dilution of 1/1000, alkaline phosphatase treated ab75814 (phospho) (top-right) at a dilution of 1/1000, untreated ab32391 (bottom-left) at a dilution of 1/1000 and alkaline phophatase treated ab32391 (bottom-right) at a dilution of 1/1000. ab97051 was used as secondary antibody at a dilution of 1/500 and counterstained with hematoxylin.



Flow Cytometry (Intracellular) - Anti-GSK3 beta antibody [Y174] (ab32391)

GSK3 beta Flow Cytometry (Intracellular) staining using rabbit Anti-GSK3 beta antibody

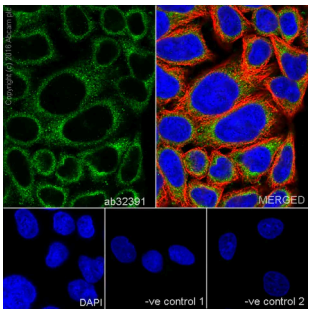
Overlay histogram showing HeLa cells stained with ab32391 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32391, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GSK3 beta antibody [Y174] (ab32391)

GSK3 beta Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-GSK3 beta antibody

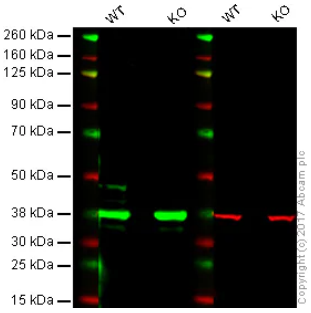
Immunohistochemical analysis of paraffin-embedded human breast cancer tissue labelled with untreated ab75814 (phospho) (top-left) at a dilution of 1/1000, alkaline phosphatase treated ab75814 (phospho) (top-right) at a dilution of 1/1000, untreated ab32391 (bottom-left) at a dilution of 1/1000 and alkaline phophatase treated ab32391 (bottom-right) at a dilution of 1/1000. ab97051 was used as secondary antibody at a dilution of 1/500 and counterstained with hematoxylin.



Immunocytochemistry/ Immunofluorescence - Anti-GSK3 beta antibody [Y174] (ab32391)

GSK3 beta Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-GSK3 beta antibody

Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling GSK3 beta with purified ab32391 at 1/500. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. An Alexa Fluor® 488-conjugated goat anti-rabbit IgG (ab150077) at 1/1000 dilution was used as the secondary antibody. Cells were co-stained with ab7291, a mouse anti-tubulin antibody (1/1000) using ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000) as the secondary antibody. Nuclei were counterstained with DAPI (blue).
For negative control 1, rabbit primary antibody was used followed by anti-mouse secondary antibody (ab150120).
For negative control 2, mouse primary antibody (ab7291) and anti-rabbit secondary antibody (ab150077) were used.



Western blot - Anti-GSK3 beta antibody [Y174] (ab32391)

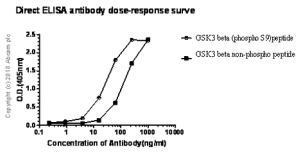
GSK3 beta Western blot staining using rabbit Anti-GSK3 beta antibody

Lane 1 & 3: Wild type HAP1 whole cell lysate (20 µg)
Lane 2 & 4: GSK3B knockout HAP1 whole cell lysate (20 µg)
Lanes 1 - 4: Green - ab32391 observed at 46 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab32391 was shown to recognize GSK3B in wild-type HAP1 cells along with additional cross-reactive bands. No band was observed when GSK3B knockout samples were examined. Wild-type and GSK3B knockout samples were subjected to SDS-PAGE. ab32391 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/2000 dilution and 1/10,000 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 1/10,000 dilution for 1 hour at room temperature before imaging.

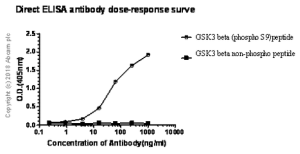
All lanes:
Western blot - Anti-GSK3 beta antibody [Y174] (ab32391)

Predicted band size: 46 kDa



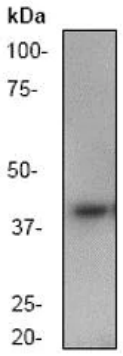
ELISA - Anti-GSK3 beta antibody [Y174] (ab32391)

ELISA of GSK3 beta (phospho S9) peptide and GSK3 beta non-phospho peptide at 1000 ng/ml. Detected with ab32391 at 0~1000 ng/ml. Alkaline Phosphatase-conjugated AffiniPure Goat Anti-Rabbit IgG (H+L) at 1/2500 was used as a secondary antibody.



ELISA - Anti-GSK3 beta antibody [Y174] (ab32391)

ELISA of GSK3 beta (phospho S9) peptide and GSK3 beta non-phospho peptide at 10 ng/ml. Detected with ab32391 at 0~1000 ng/ml. Alkaline Phosphatase-conjugated AffiniPure Goat Anti-Rabbit IgG (H+L) at 1/2500 was used as a secondary antibody.



Western blot - Anti-GSK3 beta antibody [Y174] (ab32391)

All lanes:
Western blot - Anti-GSK3 beta antibody [Y174] (ab32391) at 1/10000 dilution

All lanes:
A431 cell lysate.

Predicted band size: 46 kDa

Observed band size: 46 kDa

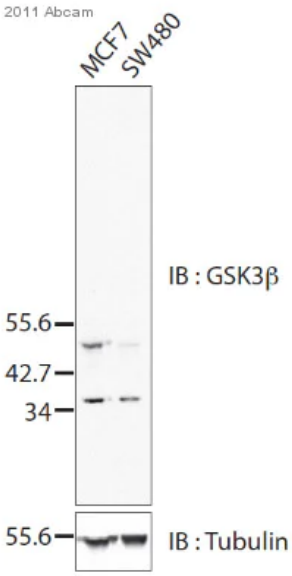


Image courtesy of an anonymous customer review.

Western blot - Anti-GSK3 beta antibody [Y174] (ab32391)

All lanes:
Western blot - Anti-GSK3 beta antibody [Y174] (ab32391) at 1/2500 dilution

Lane 1:
MCF7 (human breast adenocarcinoma cell line) cell lysate at 20 µg

Lane 2:
SW480 (human colorectal adenocarcinoma cell line) cell lysate at 20 µg

Secondary

All lanes:
Donkey polyclonal IRDye 800CW at 1/15000 dilution

Predicted band size: 46 kDa

Observed band size: 37 kDa, 46 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.