

Product datasheet

Human GSK3A (GSK3 alpha) knockout HEK293T cell line ab266742

[5 Images](#)

Overview

Product name	Human GSK3A (GSK3 alpha) knockout HEK293T cell line
Parental Cell Line	HEK293T
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon 1 and Insertion of the selection cassette in exon 1
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Tested applications	Suitable for: WB
Biosafety level	2
General notes	<p>Recommended control: Human wild-type HEK293T cell line (ab255449). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1.</p> <p>Cryopreservation cell medium: Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p>Culture medium: DMEM (High Glucose) + 10% FBS</p> <p>Initial handling guidelines: Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability.</p> <ol style="list-style-type: none"> 1. Thaw the vial in 37°C water bath approximately 1-2 minutes. 2. Transfer the cell suspension (0.8 ml) to a 15 ml/50 ml conical sterile polypropylene centrifuge tube containing 8.4 ml pre-warmed culture medium, wash vial with an additional 0.8 ml culture medium (total volume 10 ml) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 ml represents minimum recommended dilution. 20 ml represents maximum recommended dilution. 3. Resuspend the cell pellet in 5 ml pre-warmed culture medium and count using a haemocytometer (Click here to view haemocytometer protocol) or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of 2×10^4 cells/cm². This should allow for confluency within 48 hours. Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37°C incubator with 5% CO₂. Cultures should be monitored daily. <p>Subculture guidelines:</p>

All seeding densities should be based on cell counts gained by established methods. A guide seeding density of 2×10^4 cells/cm² is recommended for confluency (80-90% confluence) within 48 hours. A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required. Cells should be passaged when they have achieved 80-90% confluence. [Click here to view the Mammalian cell tissue culture protocol](#)

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Properties

Number of cells	1 x 10 ⁶ cells/vial, 1 mL
Viability	~90%
Adherent /Suspension	Adherent
Tissue	Kidney
Cell type	epithelial
STR Analysis	Amelogenin X D5S818: 8, 9 D13S317: 12, 14 D7S820: 11 D16S539: 9, 13 vWA: 16, 19 TH01: 7, 9.3 TPOX: 11 CSF1PO: 11, 12
Mycoplasma free	Yes
Storage instructions	Shipped on Dry Ice. Store in liquid nitrogen.
Storage buffer	Constituents: 8.7% DMSO, 2% Cellulose, methyl ether
Purity	Immunogen affinity purified

Target

Function	Implicated in the hormonal control of several regulatory proteins including glycogen synthase, MYB and the transcription factor JUN.
Sequence similarities	Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. GSK-3 subfamily. Contains 1 protein kinase domain.

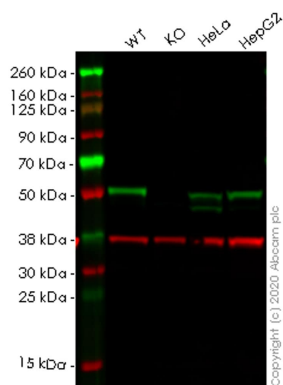
Applications

Our [Abpromise guarantee](#) covers the use of **ab266742** 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
WB		Use at an assay dependent concentration. Predicted molecular weight: 51 kDa.

Images



Western blot - Human GSK3A (GSK3 alpha)
knockout HEK293T cell line (ab266742)

All lanes : Anti-GSK3 alpha antibody [EP793Y] ([ab40870](#)) at 1/1000 dilution

Lane 1 : Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

Lane 2 : GSK3A knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole 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

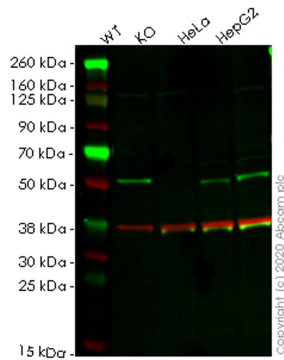
All lanes : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#)) at 1/10000 dilution

Predicted band size: 51 kDa

Observed band size: 51 kDa

Lanes 1-4: Merged signal (red and green). Green - [ab40870](#) observed at 51 kDa. Red - loading control [ab8245](#) observed at 36 kDa.

[ab40870](#) Anti-GSK3 alpha antibody [EP793Y] was shown to specifically react with GSK3 alpha in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab266742 (knockout cell lysate [ab257979](#)) was used. Wild-type and GSK3 alpha knockout samples were subjected to SDS-PAGE. [ab40870](#) and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed ([ab216776](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human GSK3A (GSK3 alpha)
knockout HEK293T cell line (ab266742)

All lanes : Anti-GSK3 alpha antibody ([ab78664](#)) at 1/1000 dilution

Lane 1 : Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

Lane 2 : GSK3A knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

Lane 3 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

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[ab78664](#) Anti-GSK3 alpha antibody was shown to specifically react with GSK3 alpha in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab266742 (knockout cell lysate [ab257979](#)) was used. Wild-type and GSK3 alpha knockout samples were subjected to SDS-PAGE. [ab78664](#) and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed ([ab216776](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

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Mut CAGGGCGCGGACTAGCTCGTTCGCGGAGCC- GGC GCGGAGGC GGAGGAGCGGC GGC GG
WT  ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| |||
CAGGGCGCGGACTAGCTCGTTCGCGGAGCCCGCGGCGGAGGC GGAGGAGCGGC GGC GG

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Sanger Sequencing - Human GSK3A knockout
HEK293T cell line (ab266742)

Allele-1: 1 bp deletion in exon1

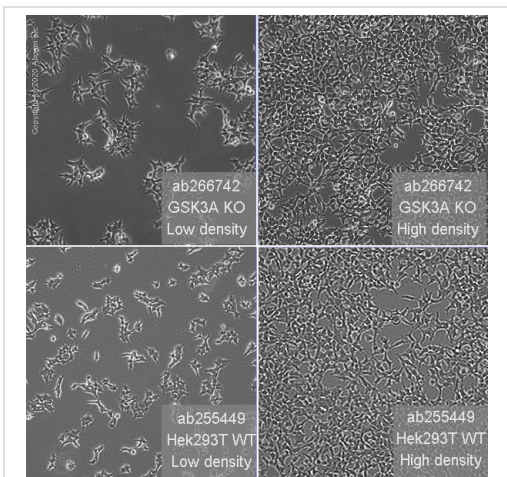
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Mut ACTAGCTCGTTCGCGGAGCC*****Insertion*****C GGC GCGGAGGC GGAGGAG
WT  ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| ||| |||
ACTAGCTCGTTCGCGGAGCC C GGC GCGGAGGC GGAGGAG

```

Sanger Sequencing - Human GSK3A knockout
HEK293T cell line (ab266742)

Allele-2: Insertion of the selection cassette in exon 1.



Human GSK3A (GSK3 alpha) knockout HEK293T cell line (ab266742)

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