

## Product datasheet

# Human HK2 (Hexokinase II) knockout HEK-293 cell line ab269485

[2 Images](#)

### Overview

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<b>Product name</b>	Human HK2 (Hexokinase II) knockout HEK-293 cell line
<b>Parental Cell Line</b>	HEK-293
<b>Organism</b>	Human
<b>Mutation description</b>	Knockout achieved by CRISPR/Cas9; X = 5 bp deletion; Frameshift: 99%
<b>Passage number</b>	<20
<b>Knockout validation</b>	Next Generation Sequencing (NGS)
<b>Tested applications</b>	<b>Suitable for:</b> WB
<b>Biosafety level</b>	2
<b>General notes</b>	<p>Western blot data indicates that the CRISPR gene edit may have resulted in a truncation of the protein of interest. Please see data images.</p> <p><b>Recommended control:</b> Human wild-type HEK-293 cell line (<a href="#">ab259776</a>). 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><b>Cryopreservation cell medium:</b> Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose.</p> <p><b>Culture medium:</b> DMEM (High Glucose) + 10% FBS</p> <p><b>Initial handling guidelines:</b> 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"><li>1. Thaw the vial in 37°C water bath for approximately 1-2 minutes.</li><li>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.</li><li>3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of <math>2 \times 10^4</math> cells/cm<sup>2</sup>. Seeding density is given as a guide only and should be scaled to align with individual lab schedules.</li><li>4. Incubate the culture at 37°C incubator with 5% CO<sub>2</sub>. Cultures should be monitored daily.</li></ol> <p><b>Subculture guidelines:</b></p>

All seeding densities should be based on cell counts gained by established methods.

A guide seeding density of  $2 \times 10^4$  cells/cm<sup>2</sup> is recommended.

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.

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## Properties

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<b>Number of cells</b>	1 x 10 <sup>6</sup> cells/vial, 1 mL
<b>Viability</b>	~80%
<b>Adherent /Suspension</b>	Adherent
<b>Tissue</b>	Kidney
<b>Cell type</b>	epithelial
<b>Gender</b>	Female
<b>Mycoplasma free</b>	Yes
<b>Storage instructions</b>	Shipped on Dry Ice. Store in liquid nitrogen.
<b>Storage buffer</b>	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

## Target

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<b>Tissue specificity</b>	Predominant hexokinase isozyme expressed in insulin-responsive tissues such as skeletal muscle.
<b>Pathway</b>	Carbohydrate metabolism; hexose metabolism.
<b>Sequence similarities</b>	Belongs to the hexokinase family. Contains 2 hexokinase domains.
<b>Domain</b>	The N- and C-terminal halves of this hexokinase show extensive sequence similarity to each other. The catalytic activity is associated with the C-terminus while regulatory function is associated with the N-terminus. Each domain can bind a single glucose and Gluc-6-P molecule.
<b>Cellular localization</b>	Mitochondrion outer membrane. Its hydrophobic N-terminal sequence may be involved in membrane binding.

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## Applications

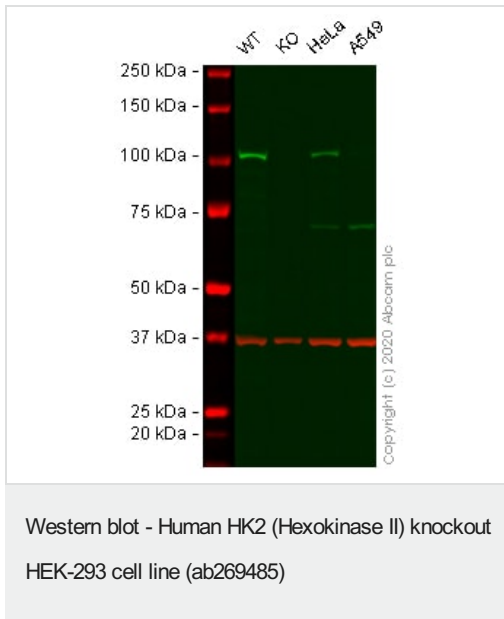
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**The Abpromise guarantee** Our [Abpromise guarantee](#) covers the use of ab269485 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes

Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Western blot data indicates that the CRISPR gene edit may have resulted in a truncation of the protein of interest. Please see data images.

## Images



**All lanes** : Anti-Hexokinase II antibody [EPR20839] ([ab209847](#)) at 1/1000 dilution

**Lane 1** : Wild-type HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

**Lane 2** : HK2 knockout HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

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

**Lane 4** : A549 (Human lung carcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Observed band size:** 110 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - [ab209847](#) observed at 110 kDa. Red - loading control [ab8245](#) (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

[ab209847](#) was shown to bind specifically to HK2. A band was observed at 110 kDa in wild-type cell lysates with no signal at this size in HK2 knockout cell line ab269485 (knockout cell lysate [ab281353](#)). Wild-type and HK2 knockout HEK-293 cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween<sup>®</sup>) before incubation with [ab209847](#) and [ab8245](#) (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed ([ab216776](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

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AGACAGAGAGCTCCACTGGATTACTTCTCTGTTCCCTGACACAGACTAACTAGACAGAGTAGATGGCTCTCAGAC Reference
|||||
AGACAGAGAGCTCCACTGGATTACTTCTCTGTTCCCTGACACAGAGTAGATGGCTCTCAGAC Deletion, 36319 reads, 83.86%
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Knockout achieved by CRISPR/Cas9; X = 5 bp deletion;  
Frameshift: 99%

Next Generation Sequencing - Human HK2  
(Hexokinase II) knockout HEK-293 cell line  
(ab269485)

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