# abcam

# Product datasheet

# Human LTA4H (Leukotriene A4 hydrolase) knockout HEK-293T cell line ab266467

## 4 Images

#### Overview

Product name Human LTA4H (Leukotriene A4 hydrolase) knockout HEK-293T cell line

Parental Cell Line HEK293T
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, Homozygous: 10 bp deletion in exon 2

Passage number <20

**Knockout validation** Sanger Sequencing, Western Blot (WB)

Tested applications Suitable for: WB

Biosafety level 2

**General notes**Recommended control: Human wild-type HEK293T cell line (<u>ab255449</u>). 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.

Cryopreservation cell medium: Cell Freezing Medium-DMSO Serum free media, contains

8.7% DMSO in MEM supplemented with methyl cellulose.

Culture medium: DMEM (High Glucose) + 10% FBS

**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.

- 1. Thaw the vial in 37°C water bath for 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 or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of 2x10<sup>4</sup> cells/cm<sup>2</sup>. 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% CO<sub>2</sub>. Cultures should be monitored daily.

#### Subculture guidelines:

All seeding densities should be based on cell counts gained by established methods. A guide seeding density of 2x10<sup>4</sup> cells/cm<sup>2</sup> is recommended.

1

A partial media change 24 hours prior to subculture may be helpful to encourage growth, if

Cells should be passaged when they have achieved 80-90% confluence.

This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our **limited use license** and **patent pages**.

We will provide viable cells that proliferate on revival.

#### **Properties**

**Number of cells** 1 x 10<sup>6</sup> cells/vial, 1 mL

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% Dimethylsulfoxide, 2% Cellulose, methyl ether

#### **Target**

**Function** Hydrolyzes an epoxide moiety of leukotriene A4 (LTA-4) to form leukotriene B4 (LTB-4). The

enzyme also has some peptidase activity.

Pathway Lipid metabolism; leukotriene B4 biosynthesis.

**Sequence similarities** Belongs to the peptidase M1 family.

Cellular localization Cytoplasm.

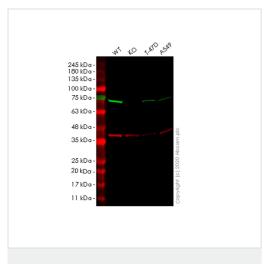
#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab266467 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: 69 kDa.

#### **Images**



Western blot - Human LTA4H knockout HEK293T cell line (ab266467)

**All lanes :** Anti-Leukotriene A4 hydrolase/LTA4H antibody [EPR5712] (ab109434) at 1/1000 dilution

Lane 1: Wild-type HEK293T cell lysate

Lane 2: LTA4H knockout HEK293T cell lysate

Lane 3 : T-47D cell lysate
Lane 4 : A549 cell lysate

Lysates/proteins at 20 µg per lane.

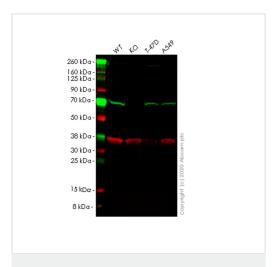
#### Secondary

**All lanes :** Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 69 kDa Observed band size: 69 kDa

**Lanes 1-4:** Merged signal (red and green). Green - <u>ab109434</u> observed at 69 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

ab109434 Anti-Leukotriene A4 hydrolase/LTA4H antibody [EPR5712] was shown to specifically react with Leukotriene A4 hydrolase/LTA4H in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266467 (knockout cell lysate ab258034) was used. Wild-type and Leukotriene A4 hydrolase/LTA4H knockout samples were subjected to SDS-PAGE. ab109434 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 LTA4H knockout HEK293T cell line (ab266467)

**All lanes :** Anti-Leukotriene A4 hydrolase/LTA4H antibody [EPR5713] (ab133512) at 1/1000 dilution

Lane 1: Wild-type HEK293T cell lysate

Lane 2: LTA4H knockout HEK293T cell lysate

Lane 3 : T-47D cell lysate Lane 4 : A549 cell lysate

Lysates/proteins at 20 µg per lane.

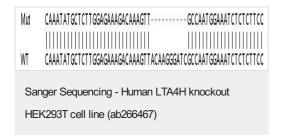
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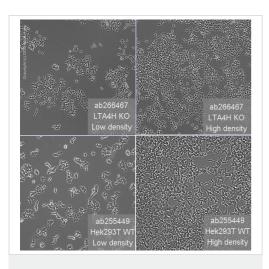
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Homozygous: 10 bp deletion in exon 2



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

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