# abcam

# Product datasheet

# Human MAPK3 (ERK1) knockout HEK-293T cell lysate ab257099

3 图像

#### 概述

产品概述

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line HEK293T
Organism Human

**Mutation description** Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon1.

Passage number <20

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

**Reconstitution notes**To use as WB control, resuspend the lyophilizate in 50 μL of LDS\* Sample Buffer to have a final

concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M

DTT.

\*Usage of SDS sample buffer is not recommended with these lyophilized lysates.

说**明** 

**Lysate preparation:** Our lysates are made using RIPA buffer to which we add a protease inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found **here**. Please refer to our lysis protocol for further details on how our lysates are prepared.

**User storage instructions:** Lyophilizate may be stored at 4°C. After reconstitution, store at -20°C for short-term storage or -80°C for long-term storage.

Access thousands of knockout cell lysates, generated from commonly used cancer cell lines. **See here for more information on knockout cell lysates.** 

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经测试应用 适用于: WB

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### 性能

# 存放说明

Store at -80°C. Please refer to protocols.

组 <b>件</b>	1 kit
ab260942 - Human MAPK3 knockout HEK293T cell lysate	1 x 100µg
ab255553 - Human wild-type HEK293T cell lysate	1 x 100µg

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

#### 靶标

功能 Involved in both the initiation and regulation of meiosis, mitosis, and postmitotic functions in

differentiated cells by phosphorylating a number of transcription factors such as ELK-1. Phosphorylates ElF4EBP1; required for initiation of translation. Phosphorylates microtubule-associated protein 2 (MAP2). Phosphorylates SPZ1 (By similarity). Phosphorylates heat shock

factor protein 4 (HSF4).

序列相似性 Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. MAP kinase

subfamily.

Contains 1 protein kinase domain.

结**构域** The TXY motif contains the threonine and tyrosine residues whose phosphorylation activates the

MAP kinases.

翻译后修饰 Dually phosphorylated on Thr-202 and Tyr-204, which activates the enzyme. Dephosphorylated by

PTPRJ at Tyr-204.

# 应用

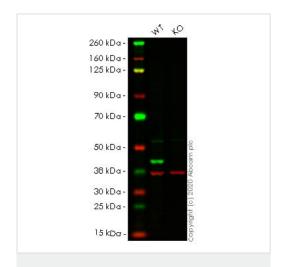
# The Abpromise guarantee

Abpromise™承诺保证使用ab257099于以下的经测试应用

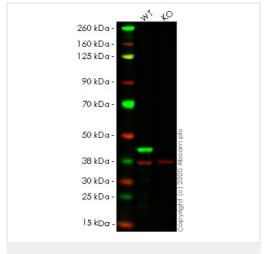
"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration. Predicted molecular weight: 43 kDa.

# 图片



Western blot - Human MAPK3 (ERK1) knockout HEK293T cell lysate (ab257099)



Western blot - Human MAPK3 (ERK1) knockout HEK293T cell lysate (ab257099)

Lane 1: Wild-type HEK-293T cell lysate (20µg)

Lane 2: MAPK3 knockout HEK-293T cell lysate (20µg)

**Lanes 1-2:** Merged signal (red and green). Green - <u>ab109282</u> observed at 43 kDa. Red - loading control <u>ab8245</u> observed at 37 kDa.

ab109282 Anti-ERK1 antibody [EP4967] was shown to specifically react with ERK1 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266519 (knockout cell lysate ab257099) was used. Wild-type and ERK1 knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% nonfat dried milk. ab109282 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 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.

Lane 1: Wild-type HEK-293T cell lysate (20µg)

Lane 2: MAPK3 knockout HEK-293T cell lysate (20µg)

**Lanes 1-2:** Merged signal (red and green). Green - <u>ab32537</u> observed at 43 kDa. Red - loading control <u>ab8245</u> observed at 37 kDa.

<u>ab32537</u> Anti-ERK1 antibody [Y72] was shown to specifically react with ERK1 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line <u>ab266519</u> (knockout cell lysate ab257099) was used. Wild-type and ERK1 knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. <u>ab32537</u> and Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) were incubated overnight at 4 °C at 1 in 1000 and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<u>ab216773</u>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed

(<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Mut G6666CG6GGGGGGGAGCCCCGTAGAAC-GAGGGGTCGGCCCGGGGGTCCCGGGGGAG

WT G6666CGGGGGGGGGGAGCCCCGTAGAACCGAGGGGGTCGGCCCGGGGGTCCCGGGGGAG

Sanger Sequencing - Human MAPK3 knockout

HEK293T cell lysate (ab257099)

Homozygous: 1 bp deletion in exon1

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