# abcam

# Product datasheet

# Human ADAR (ADAR1) knockout HEK-293T cell line ab266846

4 图像

常规说明

#### 概述

Parental Cell Line HEK293T
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, 17 bp deletion in exon 2 and 1 bp insertion in exon 2

Passage number <20

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

经测试应用 适用于: WB

Biosafety level

•

**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  $2x10^4$  cells/cm<sup>2</sup> is recommended.

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

1

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

This product is subject to limited use licenses from The Broad Institute, ERS Genomics Limited and Sigma-Aldrich Co. LLC, and is developed with patented technology. For full details of the licenses and patents please refer to our <u>limited use license</u> and <u>patent pages</u>.

We will provide viable cells that proliferate on revival.

#### 性能

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

Antibiotic resistance Puromycin 1.00µg/ml

Mycoplasma free Yes

**存放说明** Shipped on Dry Ice. Store in liquid nitrogen.

存储溶液 Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

#### 靶标

功能 Converts multiple adenosines to inosines and creates I/U mismatched base pairs in double-

helical RNA substrates without apparent sequence specificity. Has been found to modify more frequently adenosines in AU-rich regions, probably due to the relative ease of melting A/U base pairs as compared to G/C pairs. Functions to modify viral RNA genomes and may be responsible for hypermutation of certain negative-stranded viruses. Edits the messenger RNAs for glutamate receptor (GLUR) subunits by site-selective adenosine deamination. Produces low-level editing at the GLUR-B Q/R site, but edits efficiently at the R/G site and HOTSPOT1. Binds to short

interfering RNAs (siRNA) without editing them and suppresses siRNA-mediated RNA interference. Binds to ILF3/NF90 and up-regulates ILF3-mediated gene expression.

组织特异性 Ubiquitously expressed, highest levels were found in brain and lung.

疾病相关 Defects in ADAR are a cause of dyschromatosis symmetrical hereditaria (DSH) [MIM:127400];

also known as reticulate acropigmentation of Dohi. DSH is a pigmentary genodermatosis of autosomal dominant inheritance characterized by a mixture of hyperpigmented and

hypopigmented macules distributed on the dorsal parts of the hands and feet.

序列相似性 Contains 1 A to I editase domain.

Contains 2 DRADA repeats.

Contains 3 DRBM (double-stranded RNA-binding) domains.

翻译后修饰 Sumoylation reduces RNA-editing activity.

细胞定位 Cytoplasm. Nucleus > nucleolus. Isoform 1 is found predominantly in cytoplasm but appears to

shuttle between the cytoplasm and nucleus. Isoform 5 is found exclusively in the nucleolus.

#### 应用

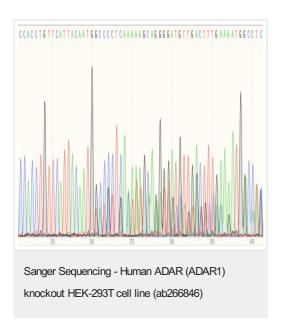
## The Abpromise guarantee

#### Abpromise™承诺保证使用ab266846于以下的经测试应用

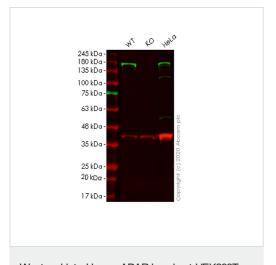
"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration. Predicted molecular weight: 136 kDa.

## 图片



Sequencing chromatogram displaying sequence edit in exon 2



Western blot - Human ADAR knockout HEK293T cell line (ab266846)

**All lanes :** Anti-ADAR1 antibody [EPR7033] (ab126745) at 1/1000 dilution

Lane 1 : Wild-type HEK293T cell lysate

Lane 2: ADAR knockout HEK293T cell lysate

Lane 3: HeLa 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:** 136 kDa **Observed band size:** 130 kDa

Lanes 1-3: Merged signal (red and green). Green - <u>ab126745</u> observed at 130 kDa. Red - loading control <u>ab8245</u> observed at 36

kDa.

ab126745 Anti-ADAR1 antibody [EPR7033] was shown to specifically react with ADAR1 in wild-type HEK293T cells. Loss of signal was observed when knockout cell line ab266846 (knockout cell lysate ab257131) was used. Wild-type and ADAR1 knockout samples were subjected to SDS-PAGE. ab126745 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 lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Allele-1: 17 bp deletion in exon2

MT CONCLOTION THOUSAND CONCOUNTS CON	CCA
THE CONCENTRATIONAL ACADEMIC CONCENTRATION C	
WT CCACCTGTTCATTACAATGGCCCCTCAAAA GCAGGGTATGTTGACTTTGAAAATGC	CCA

Allele-2: 1 bp insertion in exon 2.

Sanger Sequencing - Human ADAR knockout HEK293T cell line (ab266846)

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

#### Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- · Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <a href="https://www.abcam.cn/abpromise">https://www.abcam.cn/abpromise</a> or contact our technical team.

# Terms and conditions

• Guarantee only valid for products bought direct from Abcam or one of our authorized distributors