# abcam

## Product datasheet

# Human SDHA knockout HEK-293 cell lysate ab261657

4 图像

## 概述

产品概述 Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell LineHEK-293OrganismHuman

Mutation description Knockout achieved by CRISPR/Cas9; X = 1 bp insertion; Frameshift = 99%

Passage number <20

Knockout validation Next Generation Sequencing (NGS), 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

M- 44:

1

存放说明

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

组 <b>件</b>	1 kit
ab280407 - Human SDHA knockout HEK293 cell lysate	1 x 100µg
ab259780 - Human wild-type HEK-293 cell lysate	1 x 100µg

Cell typeepithelialGenderFemale

靶标

功能

Flavoprotein (FP) subunit of succinate dehydrogenase (SDH) that is involved in complex II of the mitochondrial electron transport chain and is responsible for transferring electrons from succinate to ubiquinone (coenzyme Q).

通路

Carbohydrate metabolism; tricarboxylic acid cycle; fumarate from succinate (eukaryal route): step

1/1

疾病相关

Defects in SDHA are a cause of mitochondrial complex II deficiency (MT-C2D) [MIM:252011]. A disorder of the mitochondrial respiratory chain with heterogeneous clinical manifestations. Clinical features include psychomotor regression in infants, poor growth with lack of speech development, severe spastic quadriplegia, dystonia, progressive leukoencephalopathy, muscle weakness, exercise intolerance, cardiomyopathy. Some patients manifest Leigh syndrome or Kearns-Sayre syndrome.

Defects in SDHA are a cause of Leigh syndrome (LS) [MIM:256000]. LS is a severe disorder characterized by bilaterally symmetrical necrotic lesions in subcortical brain regions.

Defects in SDHA are the cause of cardiomyopathy dilated type 1GG (CMD1GG) [MIM:613642]. CMD1GG is a disorder characterized by ventricular dilation and impaired systolic function, resulting in congestive heart failure and arrhythmia. Patients are at risk of premature death.

序列相似性

Belongs to the FAD-dependent oxidoreductase 2 family. FRD/SDH subfamily.

细胞定位 Mitochondrion inner membrane.

#### 应用

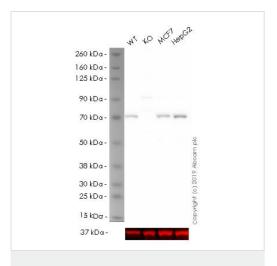
The Abpromise guarantee

Abpromise™承诺保证使用ab261657于以下的经测试应用

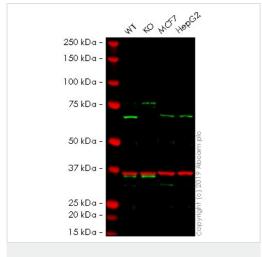
"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration.

#### 图片



Western blot - Human SDHA knockout HEK293 cell lysate (ab261657)



Western blot - Human SDHA knockout HEK293 cell lysate (ab261657)

**Lane 1:** Wild-type HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate 20 ug

Lane 2: SDHA knockout HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate 20 ug

Lane 3: MCF7 (Human breast adenocarcinoma cell line) whole cell lysate 20 ug

**Lane 4:** Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate 20 ug

ab198493 was shown to specifically react with SDHA in wild-type HEK-293 cells as signal was lost in SDHA knockout cell line ab261853 (knockout cell lysate ab261657). Wild-type and SDHA knockout samples were subjected to SDS-PAGE. Ab198493 and ab181602 (Rabbit monoclonal to GAPDH - Loading Control loading) were incubated overnight at 4°C at 1/5000 dilution and 1/20000 dilution respectively. The loading control was imaged using the Licor Odyssey CLx prior to blots being developed with ECL technique.

**Lane 1:** Wild-type HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate 20 ug

Lane 2: SDHA knockout HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate 20 ug

**Lane 3:** MCF7 (Human breast adenocarcinoma cell line) whole cell lysate 20 ug

Lane 4: Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate 20 ug

**Lanes 1 - 4:** Merged signal (red and green). Green - <u>ab139181</u> observed at 72 kDa. Red - loading control, <u>ab8245</u>, observed at 37 kDa.

<u>ab139181</u> was shown to recognize SDHA in wild-type HEK-293 cells as signal was lost at the expected MW in SDHA knockout cell line <u>ab261853</u> (knockout cell lysate ab261657). Additional cross-reactive bands were observed in the wild-type and knockout samples. Wild-type and SDHA knockout samples were subjected to

SDS-PAGE. The membrane was blocked with 3% milk. Ab139181 and <u>ab8245</u> (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed <u>ab216773</u> and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed <u>ab216776</u> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

**Lane 1:** Wild-type HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate 20 ug

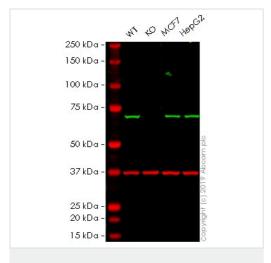
**Lane 2:** SDHA knockout HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate 20 ug

Lane 3: MCF7 (Human breast adenocarcinoma cell line) whole cell lysate 20 ug

**Lane 4:** Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate 20 ug

**Lanes 1 - 4:** Merged signal (red and green). Green - <u>ab137040</u> observed at 72 kDa. Red - loading control, <u>ab8245</u>, observed at 37 kDa.

ab137040 was shown to specifically react with SDHA in wild-type HEK-293 cells as signal was lost in SDHA knockout cell line ab261853 (knockout cell lysate ab261657). Wild-type and SDHA knockout samples were subjected to SDS-PAGE. The membrane was blocked with 3% milk. Ab137040 and ab8245 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human SDHA knockout HEK293 cell lysate (ab261657)

Next Generation Sequencing - Human SDHA knockout HEK293 cell lysate (ab261657)

X = 1 bp insertion

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