

Human SCD (SCD1) knockout HeLa cell lysate ab257658

7 图像

概述

产品名称	人SCD (SCD1) knockout HeLa cell裂解物
产品概述	Knockout cell lysate achieved by CRISPR/Cas9.
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 4 bp deletion in exon3 and Insertion of the selection cassette in exon3.
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. <i>*Usage of SDS sample buffer is not recommended with these lyophilized lysates.</i>

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

性能

存放说明 Store at -80°C. Please refer to protocols.

组件	1 kit
ab262177 - Human SCD knockout HeLa cell lysate	1 x 100µg
ab255929 - Human wild-type HeLa cell lysate	1 x 100µg

Cell type	epithelial
Disease	Adenocarcinoma
Gender	Female
STR Analysis	Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8,12 CSF1PO: 9, 10

靶标

功能	Terminal component of the liver microsomal stearyl-CoA desaturase system, that utilizes O(2) and electrons from reduced cytochrome b5 to catalyze the insertion of a double bond into a spectrum of fatty acyl-CoA substrates including palmitoyl-CoA and stearyl-CoA.
序列相似性	Belongs to the fatty acid desaturase family.
结构域	The histidine box domains may contain the active site and/or be involved in metal ion binding.
细胞定位	Endoplasmic reticulum membrane.

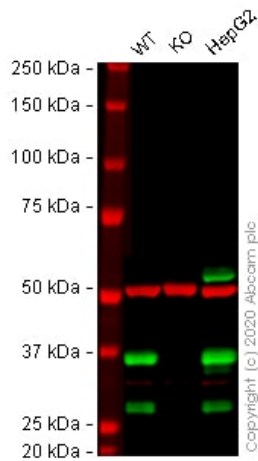
应用

The Abpromise guarantee **Abpromise™**承诺保证使用ab257658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: 42 kDa.

图片



Western blot - Human SCD (SCD1) knockout HeLa cell lysate (ab257658)

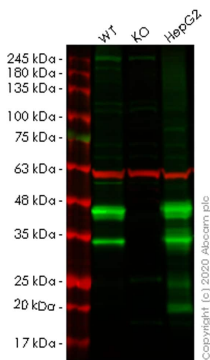
Lane 1: Wild-type HeLa cell lysate 20 ug

Lane 2: SCD knockout HeLa cell lysate 20 ug

Lane 3: HepG2 cell lysate 20 ug

Lanes 1 - 3: Merged signal (red and green). Green - **ab19862** observed at 36 kDa. Red - loading control, **ab2866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55kDa.

ab19862 was shown to react with SCD1 in wild-type HeLa cells in western blot with loss of signal observed in SCD knockout cell line **ab262177** (SCD knockout cell lysate ab257658). HeLa wild-type and SCD knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab19862** and **ab2866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4 °C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (**ab216772**) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (**ab216777**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human SCD knockout HeLa cell lysate (ab257658)

Lane 1: Wild-type HeLa cell lysate (20 ug)

Lane 2: SCD knockout HeLa cell lysate (20 ug)

Lane 3: HepG2 cell lysate (20 ug)

Lanes 1-3: Merged signal (red and green). Green - **ab236868**. Red - loading control **ab8245** observed at 50 kDa.

ab236868 Anti-SCD1 antibody [EPR21963] was shown to specifically react with SCD1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265220** (knockout cell lysate ab257658) was used. Wild-type and SCD1 knockout samples were subjected to SDS-PAGE. **ab236868** and Anti-tubulin antibody [DM1A] - Loading Control (**ab7291**) 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 10000 dilution for 1 hour at room temperature before imaging.

```

Mut  CCCTACGGCTCTTTCTGATCATTGCCAACACA---CATTCCAGTAAGAAGTTGTCTCT
      |||
WT   CCCTACGGCTCTTTCTGATCATTGCCAACACAATGGCATTCCAGTAAGAAGTTGTCTCT

```

Allele-1: 4 bp deletion in exon3

Sanger Sequencing - Human SCD knockout HeLa cell lysate (ab257658)

```

Mut  CTTTCTGATCATTGCCAACAAAAInsertionAAAAA CAATGGCATTCCAGTAAGA
      |||
WT   CTTTCTGATCATTGCCAACAA CAATGGCATTCCAGTAAGA

```

Allele-2: 4 bp deletion in exon3

Sanger Sequencing - Human SCD knockout HeLa cell lysate (ab257658)

```

Mut  CCCTACGGCTCTTTCTGATCATTGCCAACACA---CATTCCAGTAAGAAGTTGTCTCT
      |||
WT   CCCTACGGCTCTTTCTGATCATTGCCAACACAATGGCATTCCAGTAAGAAGTTGTCTCT

```

Allele-3: 4 bp deletion in exon3

Sanger Sequencing - Human SCD knockout HeLa cell lysate (ab257658)

```

Mut  CTTTCTGATCATTGCCAACAAAAInsertionAAAAA CAATGGCATTCCAGTAAGA
      |||
WT   CTTTCTGATCATTGCCAACAA CAATGGCATTCCAGTAAGA

```

Allele-4: Insertion of the selection cassette in exon3

Sanger Sequencing - Human SCD knockout HeLa cell lysate (ab257658)

```

Mut  CTTTCTGATCATTGCCAACAAAAInsertionAAAAA CAATGGCATTCCAGTAAGA
      |||
WT   CTTTCTGATCATTGCCAACAA CAATGGCATTCCAGTAAGA

```

Allele-5: Insertion of the selection cassette in exon3

Sanger Sequencing - Human SCD knockout HeLa cell lysate (ab257658)

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