# abcam

# Product datasheet

# Human NDUFA13 (GRIM19) knockout HeLa cell line ab265863

# 5 Images

#### Overview

Product name Human NDUFA13 (GRIM19) knockout HeLa cell line

Parental Cell Line HeLa
Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, 2 bp deletion in exon 2 and 73 bp insertion in exon 2

and 8 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 HeLa cell line (<u>ab255928</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.

1

A guide seeding density of 2x10<sup>4</sup> cells/cm<sup>2</sup> is recommended.

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

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.

#### **Properties**

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

Adherent /Suspension Adherent

Tissue Cervix

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

Antibiotic resistance Puromycin 1.00µg/ml

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** Accessory subunit of the mitochondrial membrane respiratory chain NADH dehydrogenase

(Complex I), that is believed not to be involved in catalysis. Complex I functions in the transfer of electrons from NADH to the respiratory chain. The immediate electron acceptor for the enzyme is believed to be ubiquinone. Involved in the interferon/all-trans-retinoic acid (IFN/RA) induced cell death. This apoptotic activity is inhibited by interaction with viral IRF1. Prevents the transactivation of STAT3 target genes. May play a role in CARD15-mediated innate mucosal responses and

serve to regulate intestinal epithelial cell responses to microbes.

**Tissue specificity** Widely expressed, with highest expression in heart, skeletal muscle, liver, kidney and placenta. In

intestinal mucosa, down-regulated in areas involved in Crohn disease and ulcerative colitis.

**Involvement in disease** Defects in NDUFA13 may be a cause of susceptibility to Hurthle cell thyroid carcinoma (HCTC)

[MIM:607464]. Hurthle cell thyroid carcinoma accounts for approximately 3% of all thyroid cancers. Although they are classified as variants of follicular neoplasms, they are more often multifocal and

somewhat more aggressive and are less likely to take up iodine than are other follicular

neoplasms.

**Sequence similarities**Belongs to the complex I NDUFA13 subunit family.

**Developmental stage** Expressed in numerous fetal tissues.

Cellular localization Mitochondrion inner membrane. Nucleus. May be translocated into the nucleus upon IFN/RA

treatment.

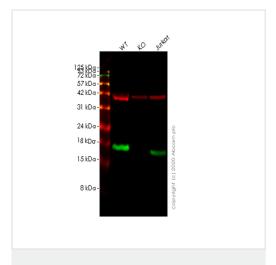
### **Applications**

### The Abpromise guarantee Our Abpromise guarantee covers the use of ab265863 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: 17 kDa.

#### **Images**



Western blot - Human NDUFA13 knockout HeLa cell line (ab265863)

**All lanes :** Anti-GRIM19 antibody [EPR4471(2)] (**ab109017**) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: NDUFA13 knockout HeLa cell lysate

Lane 3: Jurkat 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:** 17 kDa **Observed band size:** 17 kDa

**Lanes 1-3:** Merged signal (red and green). Green - <u>ab109017</u> observed at 17 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

ab109017 Anti-GRIM19 antibody [EPR4471(2)] was shown to specifically react with GRIM19 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265863 (knockout cell lysate ab257136) was used. Wild-type and GRIM19 knockout samples were subjected to SDS-PAGE. ab109017 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

3

 $lgG~H\&L~(IRDye^{®}~680RD)$  preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

125 kDa - 12 kDa - 24 kDa - 13 kDa - 13 kDa - 15 kDa - 18 kDa - 18

Western blot - Human NDUFA13 knockout HeLa cell

line (ab265863)

**All lanes :** Anti-GRIM19 antibody [6E1BH7] (**ab110240**) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: NDUFA13 knockout HeLa cell lysate

Lane 3: Jurkat cell lysate

Lysates/proteins at 20 µg per lane.

# Secondary

**All lanes :** Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (<u>ab216777</u>) at 1/10000 dilution

**Predicted band size:** 17 kDa **Observed band size:** 17 kDa

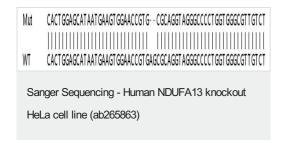
**Lanes 1-3:** Merged signal (red and green). Green - <u>ab110240</u> observed at 17 kDa. Red - loading control <u>ab181602</u> observed at 36 kDa.

ab110240 GRIM19 antibody [6E1BH7] was shown to specifically react with GRIM19 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265863 (knockout cell lysate ab257136) was used. Wild-type and GRIM19 knockout samples were subjected to SDS-PAGE. ab110240 and Anti-GAPDH antibody[EPR16891] - Loading Control (ab181602) 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® 680RD) preadsorbed (ab216777) and Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Mut CACTGGAGCATAATGAAGTGGAACCGTG-----TAGGGCCCCTGGTGGGCGTTGTCT

WT CACTGGAGCATAATGAAGTGGAACCGTGAGCGCAGGTAGGGCCCCTGGTGGGCGTTGTCT

Sanger Sequencing - Human NDUFA13 knockout HeLa cell line (ab265863) Allele-1: 8 bp deletion in exon 2.



Allele-2: 2 bp deletion in exon 2.

Mut	TGGAACCGTGAGGCGATGGTTTATGCCAATGTGATTGGCTGCGATTTGGGACCGAAAATT
WT	TGGAACCGTGAG
Sa	inger Sequencing - Human NDUFA13 knockout

HeLa cell line (ab265863)

Allele-3: 73 bp insertion in exon 2.

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.com/abpromise">https://www.abcam.com/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