# abcam

#### Product datasheet

## Human E2F3 knockout HeLa cell lysate ab257415

#### 3 Images

Overview

Product name Human E2F3 knockout HeLa cell lysate

**Product overview** 

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line HeLa

**Organism** Human

Mutation description Knockout achieved by using CRISPR/Cas9, 19 bp deletion in exon2 and 38 bp deletion in exon2.

Passage number <20

Knockout validation Sanger Sequencing

**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.

Notes

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.

Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of

products that contain European Authorisation list (Annex XIV) substances.

It is the responsibility of our customers to check the necessity of application of  $\ensuremath{\mathsf{REACH}}$ 

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Limited, and is developed with patented technology. For full details of the limited use licenses and

relevant patents please refer to our limited use license and patent pages.

Tested applications Suitable for: WB, Sanger Sequencing

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#### **Properties**

**Storage instructions** Store at -80°C. Please refer to protocols.

Components	1 kit
ab262948 - Human E2F3 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

#### **Target**

Function Transcription activator that binds DNA cooperatively with DP proteins through the E2 recognition

site, 5'-TTTC[CG]CGC-3' found in the promoter region of a number of genes whose products are involved in cell cycle regulation or in DNA replication. The DRTF1/E2F complex functions in the control of cell-cycle progression from G1 to S phase. E2F-3 binds specifically to RB1 protein, in a

cell-cycle dependent manner.

Sequence similarities Belongs to the E2F/DP family.

Cellular localization Nucleus.

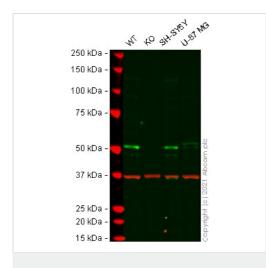
#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab257415 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: 49 kDa.
Sanger Sequencing		Use at an assay dependent concentration.

### Images



Western blot - Human E2F3 knockout HeLa cell lysate (ab257415)

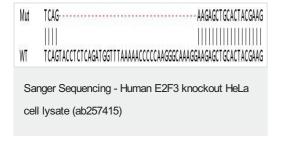
Lane 1: Wild-type HeLa cell lysate 20 µg

Lane 2: E2F3 knockout HeLa cell lysate 20 µg

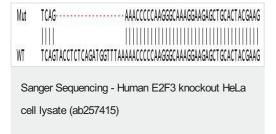
Lane 3: SH-SY5Y cell lysate 20 µg

Lane 4: U-87 MG cell lysate 20 µg

False colour image of Western blot: Anti-E2F3 antibody staining at 1/500 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab152126 was shown to bind specifically to E2F3. A band was observed at 49 kDa in wild-type HeLa cell lysates with no signal observed at this size in E2F3 knockout cell line ab265362 (knockout cell lysate ab257415). To generate this image, wild-type and E2F3 knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (ab216776) at 1/20000 dilution.



Allele-1: 38 bp deletion in exon2



Allele-2: 19 bp deletion in exon2

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

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