

## Product datasheet

# Human SQSTM1 (p62) knockout HCT116 cell lysate ab257052

3 Images

### Overview

---

<b>Product name</b>	Human SQSTM1 (p62) knockout HCT116 cell lysate
<b>Product overview</b>	Western blot data indicates that the CRISPR gene edit may have resulted in a truncation of the protein of interest. Please see data images.
<b>Parental Cell Line</b>	HCT116
<b>Organism</b>	Human
<b>Mutation description</b>	Knockout achieved by using CRISPR/Cas9, Homozygous: 49 bp deletion in exon4.
<b>Passage number</b>	<20
<b>Knockout validation</b>	Sanger Sequencing
<b>Reconstitution notes</b>	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 REACH Authorisation, and any other relevant authorisations, for their intended uses.

This product is subject to limited use licenses from The Broad Institute and ERS Genomics 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

## Properties

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

Components	1 kit
ab263488 - Human SQSTM1 knockout HCT116 cell lysate	1 x 100µg
ab255555 - Human wild-type HCT116 cell lysate	1 x 100µg

**Cell type** epithelial

**Disease** Carcinoma

**STR Analysis** Amelogenin X D5S818: 10, 11 D13S317: 10, 12 D7S820: 11, 12 D16S539: 11, 13 vWA: 17, 22 TH01: 8,9 TPOX: 8, 9 CSF1PO: 7, 10

## Target

**Function** Adapter protein which binds ubiquitin and may regulate the activation of NFkB1 by TNF-alpha, nerve growth factor (NGF) and interleukin-1. May play a role in titin/TTN downstream signaling in muscle cells. May regulate signaling cascades through ubiquitination. Adapter that mediates the interaction between TRAF6 and CYLD (By similarity). May be involved in cell differentiation, apoptosis, immune response and regulation of K(+) channels.

**Tissue specificity** Ubiquitously expressed.

**Involvement in disease** Defects in SQSTM1 are a cause of Paget disease of bone (PDB) [MIM:602080]. PDB is a metabolic bone disease affecting the axial skeleton and characterized by focal areas of increased and disorganized bone turn-over due to activated osteoclasts. Manifestations of the disease include bone pain, deformity, pathological fractures, deafness, neurological complications and increased risk of osteosarcoma. PDB is a chronic disease affecting 2 to 3% of the population above the age of 40 years.

**Sequence similarities** Contains 1 OPR domain.  
Contains 1 UBA domain.  
Contains 1 ZZ-type zinc finger.

**Domain** The UBA domain binds specifically 'Lys-63'-linked polyubiquitin chains of polyubiquitinated substrates. Mediates the interaction with TRIM55.  
The OPR domain mediates homooligomerization and interactions with PRKCZ, PRKCI, MAP2K5 and NBR1.  
The ZZ-type zinc finger mediates the interaction with RIPK1.

**Post-translational modifications** Phosphorylated. May be phosphorylated by PRKCZ (By similarity). Phosphorylated in vitro by TTN.

**Cellular localization** Cytoplasm. Late endosome. Nucleus. Sarcomere (By similarity). In cardiac muscles localizes to the sarcomeric band (By similarity). Localizes to late endosomes. May also localize to the nucleus. Accumulates in neurofibrillary tangles and in Lewy bodies of neurons from individuals with Alzheimer and Parkinson disease respectively. Enriched in Rosenthal fibers of pilocytic astrocytoma. In liver cells, accumulates in Mallory bodies associated with alcoholic hepatitis, Wilson disease, indian childhood cirrhosis and in hyaline bodies associated with hepatocellular carcinoma.

## Applications

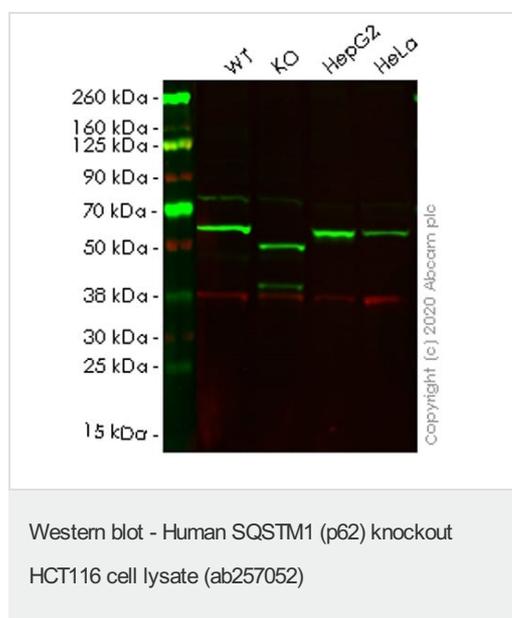
### The Abpromise guarantee

Our [Abpromise guarantee](#) covers the use of ab257052 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: 47 kDa. Western blot data indicates that the CRISPR gene edit may have resulted in a truncation of the protein of interest. Please see data images.

## Images



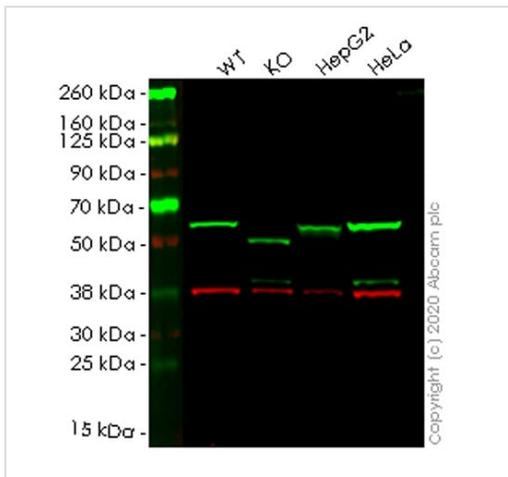
**Lane 1:** Wild-type HCT116 cell lysate (20 ug)

**Lane 2:** SQSTM1 knockout HCT116 cell lysate (20 ug)

**Lane 3:** HepG2 cell lysate (20 ug)

**Lane 4:** HeLa cell lysate (20 ug)

[ab109012](#) was shown to specifically react with SQSTM1 / p62 in wild-type HCT116 cells. The band observed in knockout cell line [ab266871](#) (knockout cell lysate ab257052) lane below 55 kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type and SQSTM1 / p62 knockout samples were subjected to SDS-PAGE. [ab109012](#) and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) were used at 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preadsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preadsorbed ([ab216776](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human SQSTM1 (p62) knockdown  
HCT116 cell lysate (ab257052)

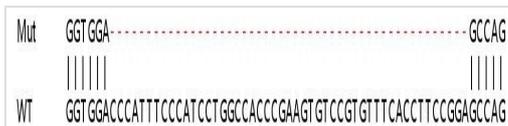
**Lane 1:** Wild-type HCT116 cell lysate (20 ug)

**Lane 2:** SQSTM1 knockout HCT116 cell lysate (20 ug)

**Lane 3:** HepG2 cell lysate (20 ug)

**Lane 4:** HeLa cell lysate (20 ug)

ab207305 was shown to specifically react with SQSTM1 / p62 in wild-type HCT116 cells. The band observed in knockout cell line ab266871 (knockout cell lysate ab257052) lane below 55 kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type and SQSTM1 / p62 knockout samples were subjected to SDS-PAGE. ab207305 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were used at 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 20000 dilution for 1 hour at room temperature before imaging.



Sanger Sequencing - Human SQSTM1 knockout  
HCT116 cell lysate (ab257052)

Homozygous: 49 bp deletion in exon4

**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 <https://www.abcam.com/abpromise> or contact our technical team.

### Terms and conditions

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