abcam

Product datasheet

Anti-SQSTM1 / p62 antibody ab155686

★★★★★ 3 Abreviews 57 References 10 Images

Overview

Product name Anti-SQSTM1 / p62 antibody

Description Rabbit polyclonal to SQSTM1 / p62

Host species Rabbit

Tested applications Suitable for: IHC-P, Flow Cyt, IP, WB, ICC/IF

Species reactivity Reacts with: Rat, Human

Immunogen Synthetic peptide corresponding to Human SQSTM1/ p62 aa 157-251 (internal sequence).

Carrier-protein conjugated synthetic peptide.

Database link: Q13501

Positive control ICC/IF: HeLa cells treated with 50µM Chloroguine for 24 hr, HepG2 cells treated with 3µM

thapsigargin 12 hrs. WB: HepG2 whole cell extracts, A549 whole cell lysate, HepG2 treated with 3µM Thapsigargin for 24 hours whole cell extracts, PC-12 whole cell lysate/extract, Rat2 whole cell lysate/extract. IP: HeLa whole cell extracts. IHC-P: human ovarian carcinoma tissue ICC/IF:

treatment with bafilomycin A1 (ab120497) at 10nM-1uM.

General notes

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.00

Preservative: 0.025% Proclin 300

Constituents: 79% PBS, 20% Glycerol (glycerin, glycerine)

Purity Immunogen affinity purified

Clonality Polyclonal

Isotype IgG

1

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab155686 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
IHC-P		1/100 - 1/1000.
Flow Cyt		1/50 - 1/200.
IP	★★★★ <u>(1)</u>	1/100 - 1/500.
WB	*** <u>*</u>	1/500 - 1/3000. Predicted molecular weight: 47 kDa.
ICC/IF		1/100 - 1/1000.

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.

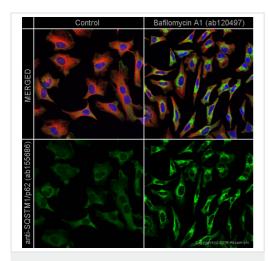
Post-translational modifications

Phosphorylated. May be phosphorylated by PRKCZ (By similarity). Phosphorylated in vitro by TTN.

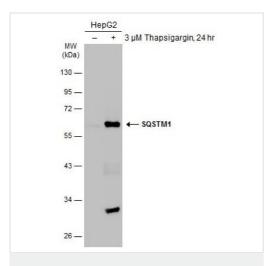
Cellular localization

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

Images



Immunocytochemistry/ Immunofluorescence - Anti-SQSTM1 / p62 antibody (ab155686)



Western blot - Anti-SQSTM1 / p62 antibody (ab155686)

ab155686 staining SQSTM1/p62 (autophagosome) in control HeLa cells (left panel) and SQSTM1/p62 in HeLa cells treated with 1uM bafilomycin A1 (ab120497) for 18hrs (right panel). The cells were fixed with methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab155686 at 1/500 dilution and ab7291 (Tubulin) at 1/1000 dilution overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to rabbit lgG (Alexa Fluor® 488) (ab150081) at 2 μ g/ml (shown in green) and a goat secondary antibody to mouse lgG (Alexa Fluor® 594) (ab150120) at 2 μ g/ml (shown in pseudo color red). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

All lanes : Anti-SQSTM1 / p62 antibody (ab155686) at 1/1000 dilution

Lane 1: Untreated (-) HepG2 whole cell extracts

Lane 2 : HepG2 treated with $3\mu M$ Thapsigargin for 24 hours whole cell extracts (+)

Lysates/proteins at 30 µg per lane.

Secondary

All lanes: Rabbit IgG antibody (HRP) at 1/10000 dilution

Predicted band size: 47 kDa

10% gel.

Running condition: 80V, 15min; 140V, 40min.

Transfer condition: Semi-dry, 18 V, 60min (Nitrocellulose

membrane).

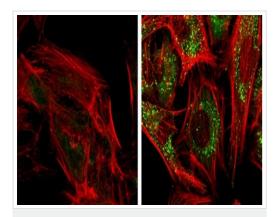
Blocking condition: 5% non-fat milk in TBST, RT, 60min.

Primary antibody incubation: 4°C overnight.

Secondary antibody incubation: Room temperature for 1 hour.

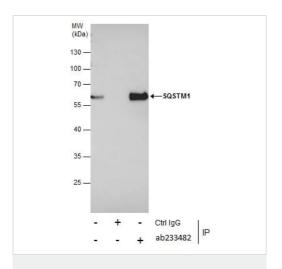
Washing condition: 5 ml TBST, 4 x 5min.

ECL exposure.



Immunocytochemistry/ Immunofluorescence - Anti-SQSTM1 / p62 antibody (ab155686)

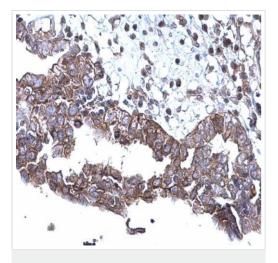
Immunocytochemistry/Immunofluorescent analysis of HeLa cells mock (left) and treated with $50\mu M$ Chloroquine for 24 hr (right) labeling SQSTM1/p62 (green) with ab155686 at 1/1000 dilution. Cells were fixed with 4% paraformaldehyde at RT for 15 min. Red: Phalloidin, a F-actin marker.



Immunoprecipitation - Anti-SQSTM1 / p62 antibody (ab155686)

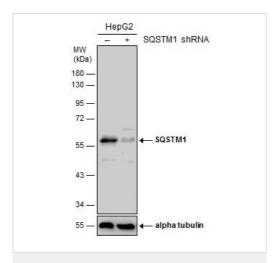
Immunoprecipitation of SQSTM1/p62 protein from HeLa whole cell extracts using 5 μ g of ab155686.

Western blot analysis was performed using ab155686. Anti-Rabbit lgG was used as a secondary reagent.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-SQSTM1 / p62 antibody (ab155686)

Paraffin-embedded human ovarian carcinoma tissue stained for SQSTM1/p62 with ab155686 (1/500) in Immunohistochemical analysis. Antigen retrieval: EDTA based buffer (pH 8) for 15 minutes.



Western blot - Anti-SQSTM1 / p62 antibody (ab155686)

All lanes : Anti-SQSTM1 / p62 antibody (ab155686) at 1/500 dilution

Lane 1: HepG2 whole cell extracts

Lane 2: SQSTM1/p62 shRNA transfected HepG2 whole cell extracts

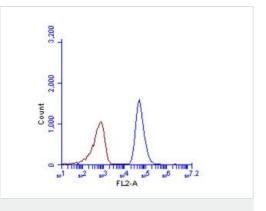
Lysates/proteins at 30 µg per lane.

Secondary

All lanes: HRP-conjugated anti-rabbit lgG antibody

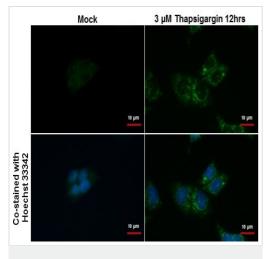
Predicted band size: 47 kDa

10% SDS-PAGE

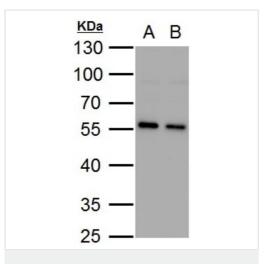


Flow Cytometry - Anti-SQSTM1 / p62 antibody (ab155686)

4% paraformaldehyde-fixed HeLa cells stained for SQSTM1/p62 (blue) using ab155686 (1/100) in Flow Cytometry. The cells were fixed at 4°C for 5 minutes. The brown line is unlabelled sample. Acquisition of >20,000 events were collected using Argon ion laser (488nm) and 525/30 bandpass filter.



Immunocytochemistry/ Immunofluorescence - Anti-SQSTM1 / p62 antibody (ab155686) Immunocytochemistry/ Immunofluorescence analysis of HepG2 cells treated with 3μ M thapsigargin for 12 hours (right) and mock (left) labeling SQSTM1/p62 with ab155686 at 1:500 (green). Cells were fixed in ice-cold MeOH for 10 minutes, permeabilized with cooled acetone for 1 minute. Cells were co-stained with Hoechst (blue).



Western blot - Anti-SQSTM1 / p62 antibody (ab155686)

All lanes : Anti-SQSTM1 / p62 antibody (ab155686) at 1/1000 dilution

Lane 1 : PC-12 whole cell lysate/extract
Lane 2 : Rat2 whole cell lysate/extract

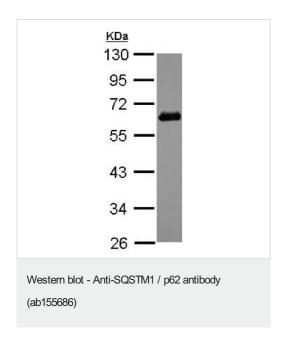
Lysates/proteins at 30 µg per lane.

Secondary

All lanes: HRP-conjugated anti-rabbit lgG antibody

Predicted band size: 47 kDa

10% SDS-PAGE



Anti-SQSTM1 / p62 antibody (ab155686) at 1/1000 dilution + A549 whole cell lysate at 30 μg

Secondary

HRP-conjugated anti-rabbit IgG antibody

Predicted band size: 47 kDa

10% SDS PAGE

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