

Anti-p62/SQSTM1 Rabbit Monoclonal Antibody

Catalog # ABO14032

Specification

Anti-p62/SQSTM1 Rabbit Monoclonal Antibody - Product Information

Application WB, IHC, IF, ICC, IP, FC

Primary Accession

Host
Isotype

Q13501
Rabbit
Rabbit IgG

Reactivity Rat, Human, Mouse

Clonality Monoclonal Format Liquid

Description

Anti-p62/SQSTM1 Rabbit Monoclonal Antibody . Tested in WB, IHC, ICC/IF, Flow Cytometry, IP applications. This antibody reacts with Human, Mouse, Rat.

Anti-p62/SQSTM1 Rabbit Monoclonal Antibody - Additional Information

Gene ID 8878

Other Names

Sequestosome-1, EBI3-associated protein of 60 kDa, EBIAP, p60, Phosphotyrosine-independent ligand for the Lck SH2 domain of 62 kDa, Ubiquitin-binding protein p62, SQSTM1 {ECO:0000303|PubMed:16286508, ECO:0000312|HGNC:HGNC:11280}

Calculated MW 47687 MW KDa

Application Details

WB 1:5000-1:10000
IHC 1:50-1:200
ICC/IF 1:50-1:200
IP 1:50
FC 1:50

Subcellular Localization

Cytoplasm. Late endosome. Lysosome. Cytoplasmic vesicle, autophagosome. Nucleus. Endoplasmic reticulum. Cytoplasm, P-body. Sarcomere (By similarity). In cardiac muscles localizes to the sarcomeric band (By similarity). Commonly found in inclusion bodies containing polyubiquitinated protein aggregates. In neurodegenerative diseases, detected in Lewy bodies in Parkinson disease, neurofibrillary tangles in Alzheimer disease, and HTT aggregates in Huntington disease. In protein aggregate diseases of the liver, found in large amounts in Mallory bodies of alcoholic and nonalcoholic steatohepatitis, hyaline bodies in hepatocellular carcinoma, and in SERPINA1 aggregates. Enriched in Rosenthal fibers of pilocytic astrocytoma. In the cytoplasm, observed in both membrane-free ubiquitin- containing protein aggregates (sequestosomes) and membrane- surrounded autophagosomes. Colocalizes with TRIM13 in the perinuclear endoplasmic reticulum. Co-localizes with TRIM5 in the cytoplasmic bodies..

Tissue Specificity

Ubiquitously expressed..

Contents

Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50%



glycerol, 0.4-0.5mg/ml BSA.

Immunogen

A synthesized peptide derived from human p62/SQSTM1

Purification

Affinity-chromatography

Storage

Store at -20°C for one year. For short term storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.

Anti-p62/SQSTM1 Rabbit Monoclonal Antibody - Protein Information

Name SQSTM1 {ECO:0000303|PubMed:16286508, ECO:0000312|HGNC:HGNC:11280}

Function

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Molecular adapter required for selective macroautophagy (aggrephagy) by acting as a bridge
between polyubiquitinated proteins and autophagosomes (PubMed: <a
href="http://www.uniprot.org/citations/15340068" target="_blank">15340068</a>, PubMed:<a href="http://www.uniprot.org/citations/15953362" target="_blank">15953362</a>, PubMed:<a
href="http://www.uniprot.org/citations/16286508" target="blank">16286508</a>, PubMed:<a
href="http://www.uniprot.org/citations/17580304" target="blank">17580304</a>, PubMed:<a
href="http://www.uniprot.org/citations/20168092" target="_blank">20168092</a>, PubMed:<a
href="http://www.uniprot.org/citations/22017874" target="_blank">22017874</a>, PubMed:<a
href="http://www.uniprot.org/citations/22622177" target="_blank">22622177</a>, PubMed:<a
href="http://www.uniprot.org/citations/24128730" target="blank">24128730</a>, PubMed:<a
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href="http://www.uniprot.org/citations/29343546" target="blank">29343546</a>, PubMed:<a
href="http://www.uniprot.org/citations/29507397" target="blank">29507397</a>, PubMed:<a
href="http://www.uniprot.org/citations/31857589" target="_blank">31857589</a>, PubMed:<a
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href="http://www.uniprot.org/citations/34471133" target="blank">34471133</a>, PubMed:<a
href="http://www.uniprot.org/citations/34893540" target="blank">34893540</a>, PubMed:<a
href="http://www.uniprot.org/citations/35831301" target="_blank">35831301</a>, PubMed:<a href="http://www.uniprot.org/citations/37306101" target="_blank">37306101</a>, PubMed:<a
href="http://www.uniprot.org/citations/37802024" target="_blank">37802024</a>). Promotes the
recruitment of ubiquitinated cargo proteins to autophagosomes via multiple domains that bridge
proteins and organelles in different steps (PubMed:<a
href="http://www.uniprot.org/citations/16286508" target=" blank">16286508</a>, PubMed:<a
href="http://www.uniprot.org/citations/20168092" target="blank">20168092</a>, PubMed:<a
href="http://www.uniprot.org/citations/22622177" target="_blank">22622177</a>, PubMed:<a
href="http://www.uniprot.org/citations/24128730" target="blank">24128730</a>, PubMed:<a
href="http://www.uniprot.org/citations/28404643" target="blank">28404643</a>, PubMed:<a
href="http://www.uniprot.org/citations/29343546" target=" blank">29343546</a>, PubMed:<a
href="http://www.uniprot.org/citations/29507397" target="blank">29507397</a>, PubMed:<a
href="http://www.uniprot.org/citations/34893540" target="blank">34893540</a>, PubMed:<a
href="http://www.uniprot.org/citations/37802024" target="blank">37802024</a>). SQSTM1 first
mediates the assembly and removal of ubiquitinated proteins by undergoing liquid-liquid phase
separation upon binding to ubiquitinated proteins via its UBA domain, leading to the formation of
insoluble cytoplasmic inclusions, known as p62 bodies (PubMed: <a
href="http://www.uniprot.org/citations/15911346" target=" blank">15911346</a>, PubMed:<a
href="http://www.uniprot.org/citations/20168092" target=" blank">20168092</a>, PubMed:<a
href="http://www.uniprot.org/citations/22017874" target="_blank">22017874</a>, PubMed:<a
href="http://www.uniprot.org/citations/24128730" target="blank">24128730</a>, PubMed:<a
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href="http://www.uniprot.org/citations/29343546" target=" blank">29343546, PubMed:29507397, PubMed:31857589, PubMed:37802024). SQSTM1 then interacts with ATG8 family proteins on autophagosomes via its LIR motif, leading to p62 body recruitment to autophagosomes, followed by autophagic clearance of ubiquitinated proteins (PubMed:16286508, PubMed: 17580304, PubMed: 20168092, PubMed: 22622177, PubMed:24128730, PubMed:28404643, PubMed: 37802024). SQSTM1 is itself degraded along with its ubiquitinated cargos (PubMed: 16286508, PubMed:17580304, PubMed:37802024). Also required to recruit ubiquitinated proteins to PML bodies in the nucleus (PubMed: 20168092). Also involved in autophagy of peroxisomes (pexophagy) in response to reactive oxygen species (ROS) by acting as a bridge between ubiquitinated PEX5 receptor and autophagosomes (PubMed:26344566). Acts as an activator of the NFE2L2/NRF2 pathway via interaction with KEAP1: interaction inactivates the BCR(KEAP1) complex by sequestering the complex in inclusion bodies, promoting nuclear accumulation of NFE2L2/NRF2 and subsequent expression of cytoprotective genes (PubMed: 20452972, PubMed:28380357, PubMed:33393215, PubMed:37306101). Promotes relocalization of 'Lys-63'-linked ubiquitinated STING1 to autophagosomes (PubMed: 29496741). Involved in endosome organization by retaining vesicles in the perinuclear cloud: following ubiquitination by RNF26, attracts specific vesicle-associated adapters, forming a molecular bridge that restrains cognate vesicles in the perinuclear region and organizes the endosomal pathway for efficient cargo transport (PubMed: 27368102, PubMed:33472082). Sequesters tensin TNS2 into cytoplasmic puncta, promoting TNS2 ubiquitination and proteasomal degradation (PubMed:25101860). May regulate the activation of NFKB1 by TNF-alpha, nerve growth factor (NGF) and interleukin-1 (PubMed: 10356400, PubMed:10747026, PubMed:11244088, PubMed:12471037, PubMed:16079148, PubMed:19931284). May play a role in titin/TTN downstream signaling in muscle cells (PubMed: 15802564). Adapter that mediates the interaction between TRAF6 and CYLD (By similarity).

Cellular Location

Cytoplasmic vesicle, autophagosome. Preautophagosomal structure. Cytoplasm, cytosol. Nucleus, PML body. Late endosome. Lysosome. Nucleus Endoplasmic reticulum. Cytoplasm, myofibril, sarcomere {ECO:0000250|UniProtKB:008623}. Note=In cardiac muscle, localizes to the sarcomeric band (By similarity). Localizes to cytoplasmic membraneless inclusion bodies, known as p62 bodies, containing polyubiquitinated protein aggregates (PubMed:11786419, PubMed:20357094, PubMed:22017874, PubMed:29343546, PubMed:29507397,



PubMed:37857589, PubMed:37306101, PubMed:37802024). In neurodegenerative diseases, detected in Lewy bodies in Parkinson disease, neurofibrillary tangles in Alzheimer disease, and HTT aggregates in Huntington disease (PubMed:15158159). In protein aggregate diseases of the liver, found in large amounts in Mallory bodies of alcoholic and nonalcoholic steatohepatitis, hyaline bodies in hepatocellular carcinoma, and in SERPINA1 aggregates (PubMed:11981755) Enriched in Rosenthal fibers of pilocytic astrocytoma (PubMed:11786419). In the cytoplasm, observed in both membrane-free ubiquitin-containing protein aggregates (sequestosomes) and membrane- surrounded autophagosomes (PubMed:15953362, PubMed:17580304) Colocalizes with TRIM13 in the perinuclear endoplasmic reticulum (PubMed:22178386). Co-localizes with TRIM5 in cytoplasmic bodies (PubMed:20357094). When nuclear export is blocked by treatment with leptomycin B, accumulates in PML bodies (PubMed:20168092) {ECO:0000250|UniProtKB:008623, ECO:0000269|PubMed:11786419, ECO:0000269|PubMed:11981755, ECO:0000269|PubMed:15158159, ECO:0000269|PubMed:15953362,

ECO:0000269|PubMed:11786419, ECO:0000269|PubMed:11981753, ECO:0000269|PubMed:15158159, ECO:0000269|PubMed:15953362, ECO:0000269|PubMed:17580304, ECO:0000269|PubMed:20168092, ECO:0000269|PubMed:20357094, ECO:0000269|PubMed:22017874, ECO:0000269|PubMed:22178386, ECO:0000269|PubMed:29343546, ECO:0000269|PubMed:29507397, ECO:0000269|PubMed:31857589, ECO:0000269|PubMed:37306101, ECO:0000269|PubMed:37802024}

Tissue LocationUbiquitously expressed.

Anti-p62/SQSTM1 Rabbit Monoclonal Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- Blocking Peptides
- Dot Blot
- <u>Immunohistochemistry</u>
- Immunofluorescence
- <u>Immunoprecipitation</u>
- Flow Cytomety
- Cell Culture

Anti-p62/SQSTM1 Rabbit Monoclonal Antibody - Images

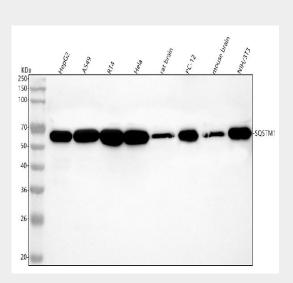


Figure 1. Western blot analysis of p62/SQSTM1 using anti-p62/SQSTM1 antibody (M00300).





Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human HepG2 whole cell lysates,

Lane 2: human A549 whole cell lysates,

Lane 3: human RT4 whole cell lysates,

Lane 4: human Hela whole cell lysates,

Lane 5: rat brain tissue lysates,

Lane 6: rat PC-12 whole cell lysates,

Lane 7: mouse brain tissue lysates,

Lane 8: mouse NIH/3T3 whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-p62/SQSTM1 antigen affinity purified monoclonal antibody (Catalog # M00300) at 1:5000 overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:1000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for p62/SQSTM1 at approximately 62 kDa. The expected band size for p62/SQSTM1 is at 48 kDa.

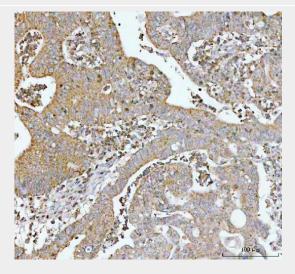


Figure 2. IHC analysis of p62/SQSTM1 using anti-p62/SQSTM1 antibody (M00300). p62/SQSTM1 was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-p62/SQSTM1 Antibody (M00300) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



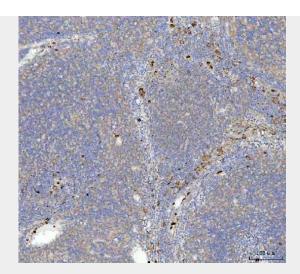


Figure 3. IHC analysis of p62/SQSTM1 using anti-p62/SQSTM1 antibody (M00300). p62/SQSTM1 was detected in a paraffin-embedded section of human spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-p62/SQSTM1 Antibody (M00300) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

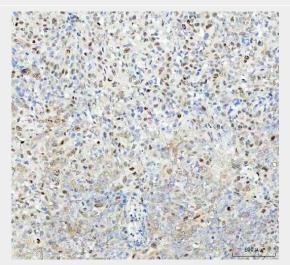
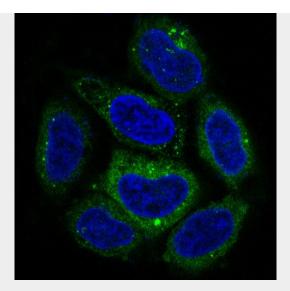
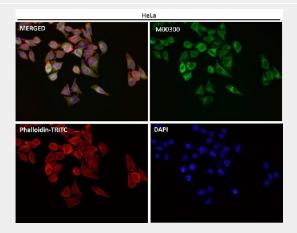


Figure 4. IHC analysis of p62/SQSTM1 using anti-p62/SQSTM1 antibody (M00300). p62/SQSTM1 was detected in a paraffin-embedded section of human cervix squamous cell carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-p62/SQSTM1 Antibody (M00300) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.





Immunofluorescent analysis of Hela cells, using p62/SQSTM1 Antibody.



Immunofluorescent analysis using the Antibody at 1:50 dilution.