abcam

ab105459

Anti-WIPI2 antibody [2A2]

Anti-WIPI2 antibody [2A2] (ab105459) is a mouse monoclonal antibody detecting WIPI2 in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF**. Suitable for **Human, Mouse**.

- KO validated for confirmed specificity
- Over 60 publications

KO Validated

Key facts

Isotype	IgG1
Host species	Mouse
Storage buffer	Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	2A2
Purification technique	Affinity purification
Epitope	EHPPM
Light chain type	kappa
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	0.1 µg for 10^6 Cells
Notes	ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Mouse, Human
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information WIPI2

Function

Component of the autophagy machinery that controls the major intracellular degradation process by which cytoplasmic materials are packaged into autophagosomes and delivered to lysosomes for degradation (PubMed:20505359, PubMed:28561066). Involved in an early step of the formation of preautophagosomal structures (PubMed:20505359, PubMed:28561066). Binds and is activated by phosphatidylinositol 3-phosphate (PtdIns3P) forming on membranes of the endoplasmic reticulum upon activation of the upstream ULK1 and PI3 kinases (PubMed:28561066). Mediates ER-isolation membranes contacts by interacting with the ULK1:RB1CC1 complex and PtdIns3P (PubMed:28890335). Once activated, WIPI2 recruits at phagophore assembly sites the ATG12-ATG5-ATG16L1 complex that directly controls the elongation of the nascent autophagosomal membrane (PubMed:20505359, PubMed:28561066). Isoform 4. Recruits the ATG12-ATG5-ATG16L1 complex to omegasomes and preautophagosomal structures, resulting in ATG8 family proteins lipidation and starvation-induced autophagy. Isoform 4 is also required for autophagic clearance of pathogenic bacteria. Isoform 4 binds the membrane surrounding Salmonella and recruits the ATG12-5-16L1 complex, initiating LC3 conjugation, autophagosomal membrane formation, and engulfment of Salmonella.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C

Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-WIPI2 antibody [2A2] (ab105459) is a mouse monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF) in Human, Mouse samples.

What is the molecular weight of WIPI2?
Anti-WIPI2 [2A2] (ab105459) specifically detects a band for WIPI2 (UniProt: Q9Y4P8) at a molecular weight of 49kDa.

Trusted by the scientific community
Anti-WIPI2 [2A2] (ab105459) was first used in a scientific publication in 2011 and has been cited over 60 times in peer-reviewed journals.

Specificity confirmed
The specificity of Anti-WIPI2 antibody [2A2] (ab105459) has been confirmed by Western blot testing in WIPI2 Knockout HAP1 cells.

Want a custom formulation?
This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact orders@abcam.com

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	WIPI2 also known as WD repeat domain phosphoinositide-interacting protein 2 functions as a critical component in autophagy a cellular degradation process. This protein weighs approximately 46 kDa and plays an important role in the initiation of autophagosome formation by binding to phosphatidylinositol 3-phosphate on lipid bilayers. Its expression is detected in various tissues with significant abundance in skeletal muscle heart and brain. These expression patterns suggest a broad involvement in cellular regulation and maintenance.
Biological function summary	WIPI2 serves as an important scaffold in the formation of the autophagy machinery. It is part of a larger complex that includes key proteins like Atg16L1 and ULK1 which are essential in the autophagy process. WIPI2 assists in recruiting additional autophagic factors to the developing autophagosome contributing to the membrane curvature and expansion necessary for cargo sequestration. Its activity ensures the efficient turnover of damaged organelles and proteins maintaining cellular health and homeostasis.
Pathways	WIPI2 integrates into the autophagy and PI3K/Akt signaling pathways. Each of these pathways plays a significant role in cellular response to nutrient deprivation

	and stress. In the autophagy pathway WIPI2 directly interacts with proteins like LC3 facilitating the biogenesis of autophagosomes. Through the PI3K/Akt pathway WIPI2 supports cell survival and growth regulation linking metabolic status with autophagic responses. This interaction highlights its importance in cellular adaptation to environmental changes.
Associated diseases and disorders	WIPI2 shows a strong connection to neurodegenerative diseases and cancer. Dysfunction in WIPI2's activity has been implicated in the progression of neurodegenerative disorders like Parkinson's Disease where impaired autophagy contributes to the accumulation of misfolded proteins. In cancer altered WIPI2 expression and function are observed linking it with tumorigenesis and cancer cell survival potentially through interactions with proteins such as mTOR and AMPK. Understanding WIPI2’s role in these conditions could lead to novel therapeutic strategies that target autophagy modulation.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

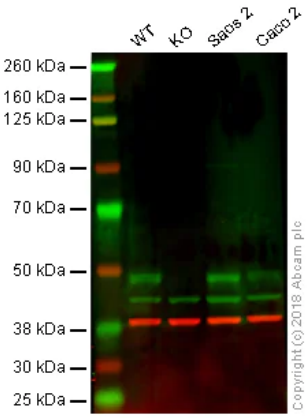
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

9 product images



Western blot - Anti-WIPI2 antibody [2A2] (ab105459)

WIPI2 Western blot staining using mouse Anti-WIPI2 antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab105459 observed at 49 kDa. Red - loading control, ab181602, observed at 37 kDa.
ab105459 was shown to recognize WIPI2 in wild-type HAP1 cells as signal was lost at the expected MW in Wipi2 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and Wipi2 knockout samples were subjected to SDS-PAGE. ab105459 and ab181602 (Rabbit anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed ab216772 and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed ab216777 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-WIPI2 antibody [2A2] (ab105459) at 1/1000 dilution

Lane 1:
Wild-type HAP1 whole cell lysate at 20 µg

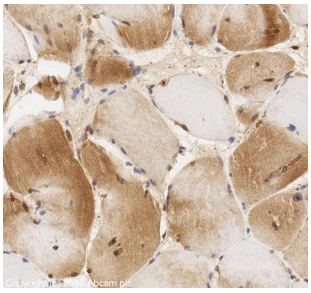
Lane 2:
Wipi2 knockout HAP1 whole cell lysate at 20 µg

Lane 3:
Saos2 whole cell lysate at 20 µg

Lane 4:
CACO2 whole cell lysate at 20 µg

Predicted band size: 49 kDa

Observed band size: 49 kDa

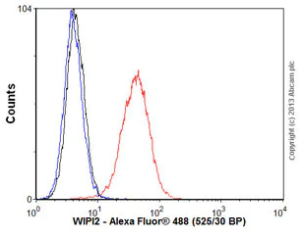


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-WIPI2 antibody [2A2] (ab105459)

WIPI2 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-WIPI2 antibody

IHC image of WIPI2 staining in a section of formalin-fixed paraffin-embedded normal human skeletal muscle* performed on a Leica BOND™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab105459, 1ug/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

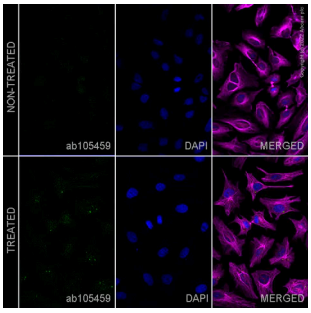
** Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre*



Flow Cytometry (Intracellular) - Anti-WIPI2 antibody [2A2] (ab105459)

WIPI2 Flow Cytometry (Intracellular) staining using mouse Anti-WIPI2 antibody

Overlay histogram showing HeLa cells stained with ab105459 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab105459, 0.1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-mouse IgG (H+L) (ab150113) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

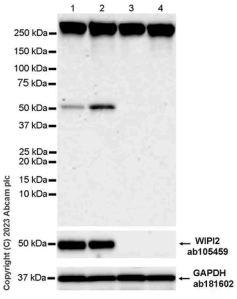


Immunocytochemistry/ Immunofluorescence - Anti-WIPI2 antibody [2A2] (ab105459)

WIPI2 Immunocytochemistry/ Immunofluorescence staining of HeLa Chloroquine treated cells using mouse Anti-WIPI2 antibody

ab105459 staining WIPI2 in HeLa Chloroquine treated cells. The cells were fixed with 100% methanol 5 min), permeabilized with 0.1% PBS-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 overnight at 4°C with ab105459 at 5µg/ml and ab6046, Rabbit polyclonal to beta Tubulin - Loading Control. Cells were then incubated with ab150117, Goat polyclonal Secondary Antibody to Mouse IgG H&L Alexa Fluor® 488) preadsorbed at 1/1000 dilution shown in green) and ab150080, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L Alexa Fluor® 594) at 1/1000 dilution shown in pseudocolour magenta). Nuclear DNA was labelled with DAPI shown in blue).

Image was acquired with a high-content analyser Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Western blot - Anti-WIPI2 antibody [2A2] (ab105459)

Blocking and diluting buffer and concentration: 5% NFDM/TBST The identity of the higher MW band at approximately 250kDa is unknown In Western blot, anti-WIPI2 antibody (ab105459) staining at 1/1000 dilution.Anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution. Exposure time: 180 seconds
All lanes:
Western blot - Anti-WIPI2 (phospho S413) antibody [EPR27037-149] (ab310325) at 1/1000 dilution

Lane 1:
Untreated wild-type HAP1(human chronic myelogenous leukemia near-haploid cell line) whole cell lysate at 20 µg

Lane 2:
HAP1 treated with 200 nM TPA for 30 min, whole cell lysate at 20 µg

Lane 3:
WIPI2 knockout HAP1 whole cell lysate

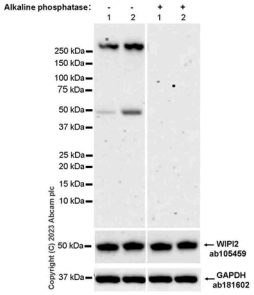
Lane 4:
WIPI2 knockout HAP1 treated with 200 nM TPA for 30 min, whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 50 kDa

Exposure time: 180s



Western blot - Anti-WIPI2 antibody [2A2] (ab105459)

Blocking and diluting buffer and concentration: 5% NFDM/TBST The identity of the higher MW band at approximately 250kDa is unknown. In Western blot, anti-WIPI2 antibody (ab105459) staining at 1/1000 dilution.Anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution. Exposure time: 180 seconds
All lanes:
Western blot - Anti-WIPI2 (phospho S413) antibody [EPR27037-149] (ab310325) at 1/1000 dilution

Lane 1:
Untreated KARPAS-299 (human T cell lymphoma cell) starved overnight whole cell lysate (untreated membrane) at 20 µg

Lane 2:
KARPAS-299 starved overnight, then treated with 200nM TPA for 30min whole cell lysate (untreated membrane) at 20 µg

Lane 3:
Untreated KARPAS-299 starved overnight whole cell lysate (phosphatase treated membrane) at 20 µg

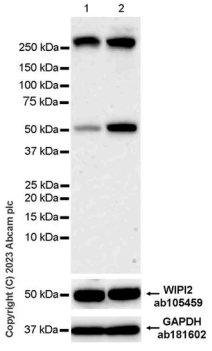
Lane 4:
KARPAS-299 starved overnight, then treated with 200nM TPA for 30min whole cell lysate (phosphatase treated membrane) at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 50 kDa

Exposure time: 180s



Western blot - Anti-WIPI2 antibody [2A2] (ab105459)

Blocking and diluting buffer and concentration: 5% NFDM/TBST The identity of the higher MW band at approximately 250kDa is unknown In Western blot, anti-WIPI2 antibody (ab105459) staining at 1/1000 dilution.Anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution. Exposure time: 180 seconds
All lanes:
Western blot - Anti-WIPI2 (phospho S413) antibody [EPR27037-149] (ab310325) at 1/1000 dilution

Lane 1:
Untreated Raji (human Burkitts lymphoma B lymphocyte) whole cell lysate

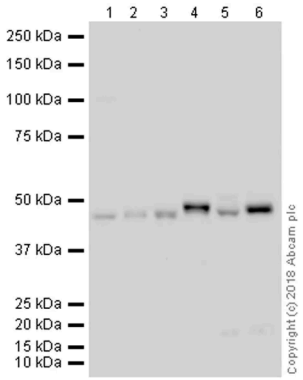
Lane 2:
Raji starved overnight, then treated with 200nM TPA for 30min whole cell lysate

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 50 kDa

Exposure time: 180s



Western blot - Anti-WIPI2 antibody [2A2] (ab105459)

Abcam recommends using milk as the blocking agent.
All lanes:
Western blot - Anti-WIPI2 antibody [2A2] (ab105459) at 1 µg/mL

Lane 1:
Human skeletal muscle tissue lysate at 10 µg

Lane 2:
Human placenta tissue lysate at 10 µg

Lane 3:
HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate at 10 µg

Lane 4:
NIH 3T3 (Mouse) Whole Cell Lysate at 10 µg

Lane 5:
Pancreas (Mouse) Tissue Lysate at 10 µg

Lane 6:
Testis (Mouse) Tissue Lysate at 10 µg

Secondary

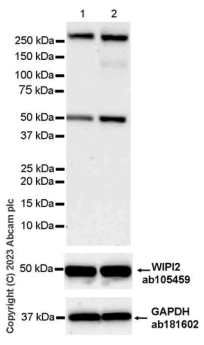
All lanes:
Goat polyclonal to Mouse IgG - H&L - Pre-Adsorbed (HRP) (ab65485) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 49 kDa

Exposure time: 1min



Western blot - Anti-WIPI2 antibody [2A2] (ab105459)

Blocking and diluting buffer and concentration: 5% NFDM/TBST The identity of the higher MW band at approximately 250kDa is unknown. In Western blot, anti-WIPI2 antibody (ab105459) staining at 1/1000 dilution.Anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution. Exposure time: 180 seconds
All lanes:
Western blot - Anti-WIPI2 (phospho S413) antibody [EPR27037-149] (ab310325) at 1/1000 dilution

Lane 1:
Untreated MEF (mouse embryonic fibroblast (immortalized)) whole cell lysate at 20 µg

Lane 2:

MEF treated with 5uM MG-132 for 6h whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 50 kDa

Exposure time: 180s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.