abcam

ab232372

Anti-ULK3 antibody [EPR4888] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR4888
Purification technique	Affinity purification Protein A

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human

Dilution info	-
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Not recommended

Species	Mouse
Dilution info	-
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Species	Rat
Dilution info	-
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target data

See full target information Serine/threonine-protein kinase ULK3

Function	Serine/threonine protein kinase that acts as a regulator of Sonic hedgehog (SHH) signaling and autophagy. Acts as a negative regulator of SHH signaling in the absence of SHH ligand: interacts with SUFU, thereby inactivating the protein kinase activity and preventing phosphorylation of GLI proteins (GLI1, GLI2 and/or GLI3). Positively regulates SHH signaling in the presence of SHH: dissociates from SUFU, autophosphorylates and mediates phosphorylation of GLI2, activating it and promoting its nuclear translocation. Phosphorylates in vitro GLI2, as well as GLI1 and GLI3, although less efficiently. Also acts as a regulator of autophagy: following cellular senescence, able to induce autophagy.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab232372 is the carrier-free version of ab124947.

Patented technology

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?

This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Conjugation ready

Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

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

Activity summary	ULK3 also known as Unc-51 Like Autophagy Activating Kinase 3 is a serine/threonine-protein kinase with a molecular mass of approximately 58 kDa. This protein plays an important role in mediating cellular processes through phosphorylation. ULK3 shows expression in various tissues including the brain and kidney. Its catalytic activity influences multiple cellular events especially those related to autophagy the cell's degradation pathway for removing damaged organelles and recycling cellular components.
Biological function summary	This protein contributes significantly through its involvement in autophagy regulation. ULK3 acts as a kinase within a multiprotein complex which includes proteins such as ATG13 and FIP200. This complex is essential for initiating autophagy by regulating the autophagosome formation. Its activity can promote cellular survival under nutrient-deprived conditions showcasing its role as an important regulatory component in cellular maintenance and stress responses.
Pathways	ULK3 takes part in critical signaling pathways such as the mTOR signaling pathway and the Hedgehog signaling pathway. In the mTOR pathway ULK3 through its kinase activity acts upstream to regulate the activity of the autophagy machinery.

	It shares a functional relationship with proteins like mTOR and AMPK. In the Hedgehog pathway ULK3 regulates pathway components impacting cellular growth and differentiation processes. This positions ULK3 as a modulator connecting different signaling pathways to maintain cellular homeostasis.
Associated diseases and disorders	Researchers have reported associations with cancer and neurodegenerative diseases. Abnormal ULK3 activity can influence tumor growth and progression particularly in its regulatory role in cell proliferation and survival. Studies also suggest an involvement in neurodegenerative diseases where dysregulation in autophagy processes in which ULK3 plays a part contributes to neuronal cell death. In these contexts ULK3's interactions with proteins like mTOR and Beclin-1 may exacerbate disease progression by altering normal cellular processes.

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

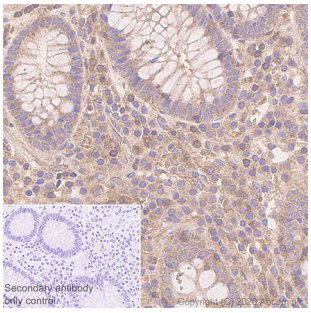
We do not recommend this combination. It is not covered by our product promise.

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.

7 product images

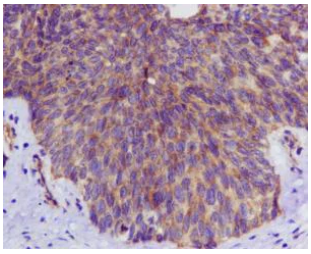


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ULK3 antibody [EPR4888] - BSA and Azide free (ab232372)

ULK3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ULK3 antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue sections labeling ULK3 with purified ab124947 at 1/50 dilution (7.84 µg/mL). Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0). Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

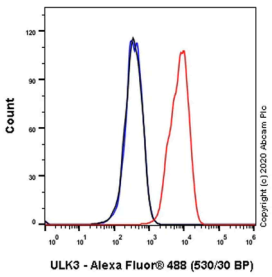
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124947).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ULK3 antibody [EPR4888] - BSA and Azide free (ab232372)

ULK3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ULK3 antibody

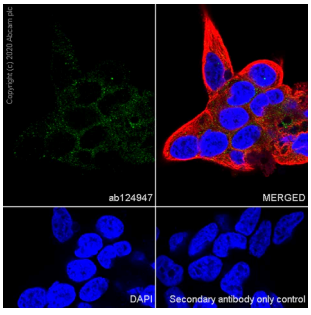
ab124947 (unpurified), at a 1/50 dilution, staining ULK3 in paraffin embedded Human bladder carcinoma tissue by Immunohistochemistry. Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124947).



Flow Cytometry (Intracellular) - Anti-ULK3 antibody [EPR4888] - BSA and Azide free (ab232372)

ULK3 Flow Cytometry (Intracellular) staining using rabbit Anti-ULK3 antibody

Intracellular Flow Cytometry analysis of LNCaP (Human prostate carcinoma epithelial cell) cells labeling ULK3 with Purified ab124947 at 1/40 dilution (10µg/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue). This data was developed using the same antibody clone in a different buffer formulation containing PBS,BSA,glycerol,and sodium azide (ab124947).

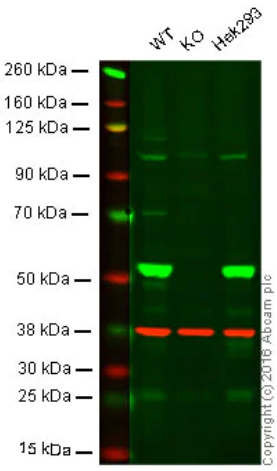


Immunocytochemistry/ Immunofluorescence - Anti-ULK3 antibody [EPR4888] - BSA and Azide free (ab232372)

ULK3 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-ULK3 antibody

Immunocytochemistry/ Immunofluorescence analysis of LNCaP (Human prostate carcinoma epithelial cell) cells labeling ULK3 with Purified ab124947 at 1:50 dilution (7.8 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200

(2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124947).



Western blot - Anti-ULK3 antibody [EPR4888] - BSA and Azide free (ab232372)

ULK3 Western blot staining using rabbit Anti-ULK3 antibody

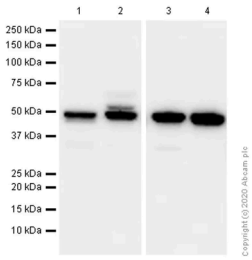
Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: ULK3 knockout HAP1 whole cell lysate (20 µg)
Lane 3: Hek293 whole cell lysate (20 µg)
Lanes 1 - 3: Merged signal (red and green). Green - ab124947 observed at 55 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab124947 (unpurified) was shown to specifically react with ULK3 in wild-type HAP1 cells along with additional cross-reactive bands. No band was observed when ULK3 knockout samples were examined. Wild-type and ULK3 knockout samples were subjected to SDS-PAGE. ab124947 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10,000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) ab216776 secondary antibodies at 1/10,000 dilution for 1 hour at room temperature before imaging.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124947).

All lanes:
Western blot - Anti-ULK3 antibody [EPR4888] (ab124947)

Predicted band size: 102 kDa, 121 kDa, 18 kDa, 53 kDa

Observed band size: 150 kDa



Western blot - Anti-ULK3 antibody [EPR4888] - BSA and Azide free (ab232372)

Blocking/Diluting buffer: 5% NFDM/TBST
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab124947).

All lanes:
Western blot - Anti-ULK3 antibody [EPR4888] (ab124947) at 1/1000 dilution

Lane 1:
Human heart lysate at 20 µg

Lane 2:
Human testis lysate at 20 µg

Lane 3:
LNCaP (Human prostate carcinoma epithelial cell) whole cell lysate at 20 µg

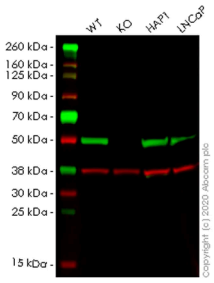
Lane 4:
HEK-293 (Human embryonic kidney epithelial cell) whole cell lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 53 kDa

Observed band size: 53 kDa



Western blot - Anti-ULK3 antibody [EPR4888] - BSA and Azide free (ab232372)

ULK3 Western blot staining using rabbit Anti-ULK3 antibody

This data was developed using ab124947, the same antibody clone in a different buffer formulation.
Lanes 1-4: Merged signal (red and green). Green - ab124947 observed at 55 kDa. Red - loading control ab8245 observed at 36 kDa.

ab124947 Anti-ULK3 antibody [EPR4888] was shown to specifically react with ULK3 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab266152 (knockout cell lysate ab258271) was used. Wild-type and ULK3 knockout samples were subjected to SDS-PAGE. ab124947 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 dilution and 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.

All lanes:
Western blot - Anti-ULK3 antibody [EPR4888] (ab124947) at 1/1000 dilution

Lane 1:
Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate at 20 µg

Lane 2:
ULK3 knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate at 20 µg

Lane 2:
Western blot - Human ULK3 knockout HEK-293T cell line (ab266152)

Lane 3:
HAP1 whole cell lyate at 20 µg

Lane 4:
LNCaP (Human prostate cancer cell line) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 53 kDa

Observed band size: 55 kDa

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