abcam

ab32041

Anti-IKK alpha antibody [Y463]

Anti-IKK alpha antibody [Y463] (ab32041) is a rabbit monoclonal antibody detecting IKK alpha in **Western Blot, Flow Cytometry (Intra), IP, IHC-P**. Suitable for **Human, Mouse, Rat**.

- KO validated for confirmed specificity
- Biophysical QC for unrivalled batch-batch consistency
- Over 70 publications
- Trusted since 2006

Recombinant | RabMAb | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	Y463
Purification technique	Affinity purification Protein A
Concentration	0.21 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/50
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Species	Mouse
Dilution info	1/50
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Not recommended

Species	Rat
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	1/60
Notes	-

Expected

Species	Mouse
Dilution info	1/60
Notes	-

Species	Rat
Dilution info	1/60
Notes	-

WB

Tested

Species	Rat
Dilution info	1/1000
Notes	**For unpurified use at 1/10000 - 1/50000.**

Species	Human
Dilution info	1/1000
Notes	**For unpurified use at 1/10000 - 1/50000.**

Expected

Species	Mouse
Dilution info	1/1000
Notes	**For unpurified use at 1/10000 - 1/50000.**

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/20
Notes	For unpurified use at 1/50. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	1/20
Notes	For unpurified use at 1/50. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Species	Rat
Dilution info	1/20
Notes	For unpurified use at 1/50. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target data

See full target information CHUK

Function	Serine kinase that plays an essential role in the NF-kappa-B signaling pathway which is activated by multiple stimuli such as inflammatory cytokines, bacterial or viral products, DNA damages or other cellular stresses (PubMed:18626576, PubMed:9244310, PubMed:9252186, PubMed:9346484). Acts as a part of the canonical IKK complex in the conventional pathway of NF-kappa-B activation and phosphorylates inhibitors of NF-kappa-B on serine residues (PubMed:18626576, PubMed:35952808, PubMed:9244310, PubMed:9252186, PubMed:9346484). These modifications allow polyubiquitination of the inhibitors and subsequent degradation by the proteasome (PubMed:18626576, PubMed:9244310, PubMed:9252186, PubMed:9346484). In turn, free NF-kappa-B is translocated into the nucleus and activates the transcription of hundreds of genes involved in immune response, growth control, or protection against apoptosis (PubMed:18626576, PubMed:9244310, PubMed:9252186, PubMed:9346484). Negatively regulates the pathway by phosphorylating the scaffold protein TAXBP1 and thus promoting the assembly of the A20/TNFAIP3 ubiquitin-editing complex (composed of A20/TNFAIP3, TAX1BP1, and the E3 ligases ITCH and RNF11) (PubMed:21765415). Therefore, CHUK plays a key role in the negative feedback of NF-kappa-B canonical signaling to limit inflammatory gene activation. As part of the non-canonical pathway of NF-kappa-B activation, the MAP3K14-activated CHUK/IKKA homodimer phosphorylates NFKB2/p100 associated with RelB, inducing its proteolytic processing to NFKB2/p52 and the formation of NF-kappa-B RelB-p52 complexes (PubMed:20501937). In turn, these complexes regulate genes encoding molecules involved in B-cell survival and lymphoid organogenesis. Also participates in the negative feedback of the non-canonical NF-kappa-B signaling pathway by phosphorylating and destabilizing MAP3K14/NIK. Within the nucleus, phosphorylates CREBBP and consequently increases both its transcriptional and histone acetyltransferase activities (PubMed:17434128). Modulates chromatin accessibility at NF-kappa-B-responsive promoters by phosphorylating histones H3 at 'Ser-10' that are subsequently acetylated at 'Lys-14' by CREBBP (PubMed:12789342). Additionally, phosphorylates the CREBBP-interacting protein NCOA3. Also phosphorylates FOXO3 and may regulate this pro-apoptotic transcription factor (PubMed:15084260). Phosphorylates RIPK1 at 'Ser-25' which represses its kinase activity and consequently prevents TNF-mediated RIPK1-dependent cell death (By similarity). Phosphorylates AMBRA1 following mitophagy induction, promoting AMBRA1 interaction with ATG8 family proteins and its mitophagic activity (PubMed:30217973).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
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-IKK alpha antibody [Y463] (ab32041) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunoprecipitation (IP), Immunohistochemistry (IHC-P) in Human, Mouse, Rat samples.

What is the molecular weight of IKK alpha?
Anti-IKK alpha [Y463] (ab32041) specifically detects a band for IKK alpha (UniProt: O15111) at a molecular weight of 85kDa.

Trusted by the scientific community
Anti-IKK alpha [Y463] (ab32041) was first used in a scientific publication in 2006 and has been cited over 70 times in peer-reviewed journals.

Reviewed by scientists
Anti-IKK alpha [Y463] (ab32041) has over 5 independent reviews from customers.

Specificity confirmed
The specificity of Anti-IKK alpha antibody [Y463] (ab32041) has been confirmed by Western blot testing in CHUK Knockout HAP1 cells.

Other related products
We have a range of other formats of antibody clone [Y463] also available for your convenience: ab32041, Carrier free - ab169743, Alexa Fluor® 488 - ab200412, Alexa Fluor® 647 - ab200414, HRP - ab200415, APC - ab221910

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

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.

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

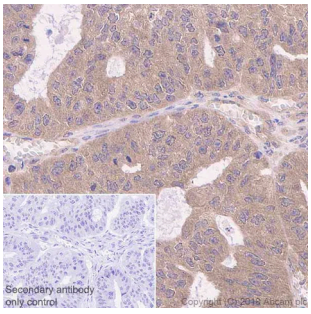
We do not recommend this combination. It is not covered by our product promise.

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.

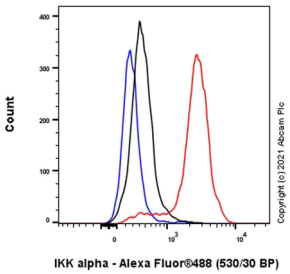
9 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-IKK alpha antibody [Y463] (ab32041)

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

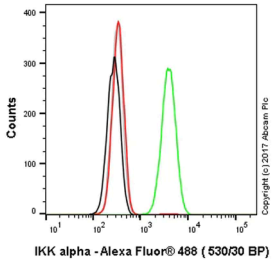
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human ovarian cancer tissue sections labeling IKK alpha with purified ab32041 at 1/50 dilution (3.92 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) secondary antibody was used at 1/0 dilution. PBS instead of the primary antibody was used as the negative control.



Flow Cytometry (Intracellular) - Anti-IKK alpha antibody [Y463] (ab32041)

IKK alpha Flow Cytometry (Intracellular) staining using rabbit Anti-IKK alpha antibody

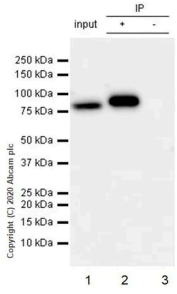
Intracellular Flow Cytometry analysis of Daudi (Human Burkitt's lymphoma lymphoblast) cells labelling IKK alpha with purified ab32041 at 1/20 dilution (10 µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Flow Cytometry (Intracellular) - Anti-IKK alpha antibody [Y463] (ab32041)

IKK alpha Flow Cytometry (Intracellular) staining using rabbit Anti-IKK alpha antibody

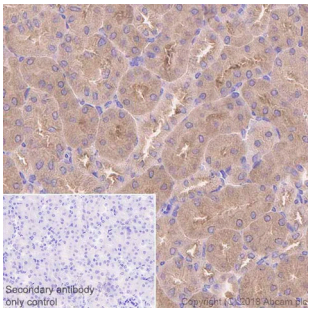
Overlay histogram showing HAP1 wildtype (green line) and HAP1-CHUK knockout cells (red line) stained with ab32041. The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS / 10% normal goat serum to block non-specific protein-protein interactions followed by the antibody (ab32041, 1µg/ml) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) presorbed (ab150081) at 1/2000 dilution for 30 min at 22°C. A rabbit IgG isotype control antibody (ab172730) was used at the same concentration and conditions as the primary antibody (HAP1 wildtype - black line, HAP1-CHUK knockout - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity). Acquisition of >5,000 events were collected using a 50 mW Blue laser (488nm) and 530/30 bandpass filter. This antibody can also be used in HAP1 cells fixed with 80% methanol (5 min), permeabilized with 0.1% PBS-Triton X-100 for 15 min under the same conditions.



Immunoprecipitation - Anti-IKK alpha antibody [Y463] (ab32041)

Purified ab32041 at 1/60 dilution (2µg) immunoprecipitating IKK alpha in HeLa whole cell lysate.
Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10µg
Lane 2 (+): ab32041 + HeLa whole cell lysate.
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab169743 in HeLa whole cell lysate.
VeriBlot for IP Detection Reagent (HRP) (ab131366) (1/1000 dilution) was used for Western blotting.
Blocking Buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM/TBST.
Observed band size: 88 kDa
All lanes:
Immunoprecipitation - Anti-IKK alpha antibody [Y463] (ab32041)

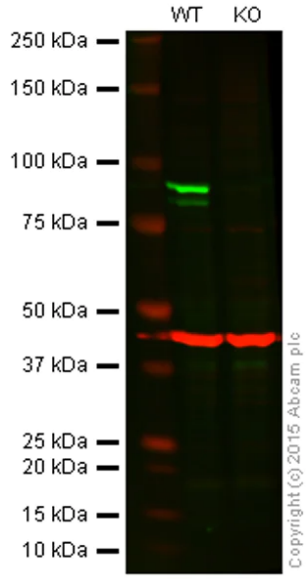
Predicted band size: 84 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-IKK alpha antibody [Y463] (ab32041)

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

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue sections labeling IKK alpha with purified ab32041 at 1/50 dilution (3.92 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) secondary antibody was used at 1/0 dilution. PBS instead of the primary antibody was used as the negative control.



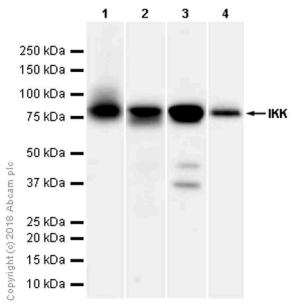
Western blot - Anti-IKK alpha antibody [Y463] (ab32041)

IKK alpha Western blot staining using rabbit Anti-IKK alpha antibody

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: IKK alpha knockout HAP1 cell lysate (20 µg)
Lanes 1 and 2: Merged signal (red and green). Green - ab32041 observed at 84 kDa. Red - loading control, ab8226, observed at 42 kDa.
ab32041 (Unpurified format) was shown to specifically react with IKK alpha when IKK alpha knockout samples were used. Wild-type and IKK alpha knockout samples were subjected to SDS-PAGE. ab32041 and ab8226 (loading control to beta actin) were diluted 1/10000 and 1/1000 respectively and incubated overnight at 4°C. 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 1/10000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-IKK alpha antibody [Y463] (ab32041)

Predicted band size: 84 kDa



Western blot - Anti-IKK alpha antibody [Y463] (ab32041)

All lanes:
Western blot - Anti-IKK alpha antibody [Y463] (ab32041) at 1/1000 dilution

Lane 1:
RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate at 15 µg

Lane 2:
Mouse kidney lysate at 15 µg

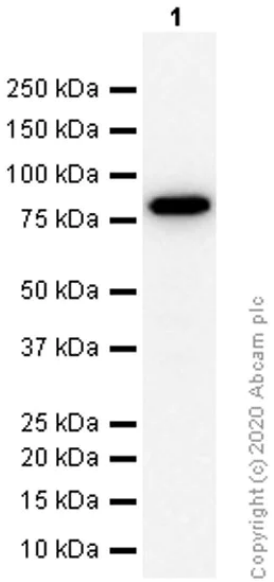
Lane 3:
C6 (Rat glial tumor glial cell) whole cell lysate at 15 µg

Lane 4:
Rat kidney lysate at 15 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 84 kDa



Western blot - Anti-IKK alpha antibody [Y463] (ab32041)

IKK alpha Western blot staining of Daudi (Human Burkitt's lymphoma lymphoblast) whole cell lysate using rabbit Anti-IKK alpha antibody

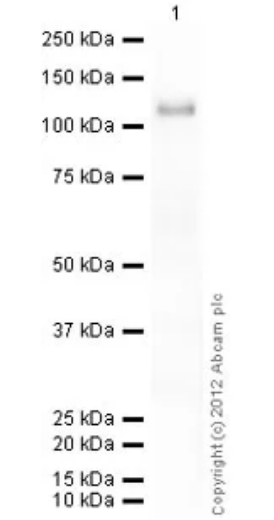
All lanes:
Western blot - Anti-IKK alpha antibody [Y463] (ab32041) at 1/10000 dilution

All lanes:
Daudi (Human Burkitt's lymphoma lymphoblast) whole cell lysate

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/20000 dilution

Predicted band size: 84 kDa



Western blot - Anti-IKK alpha antibody [Y463] (ab32041)

IKK alpha Western blot staining using rabbit Anti-IKK alpha antibody

All lanes:
Western blot - Anti-IKK alpha antibody [Y463] (ab32041) at 1/10000 dilution

All lanes:
Western blot - Recombinant human IKK alpha protein (Active) (GST tag N-Terminus) (ab102103) at 0.01 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 84 kDa

Exposure time: 10s

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