## **Product Datasheet**

# IKK alpha Antibody (14A231) - Azide Free NBP2-27409

Unit Size: 0.1 mg

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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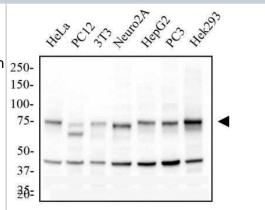


IKK alpha Antibody (14A231) - Azide Free	
Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	14A231
Preservative	No Preservative
Isotype	IgG1 Kappa
Purity	Protein G purified
Buffer	PBS
Product Description	
Host	Mouse
Gene ID	1147
Gene Symbol	CHUK
Species	Human, Mouse, Rat, Primate
Reactivity Notes	New World Monkey
Immunogen	This antibody was raised against a His-tagged full-length human IKK alpha protein.
Product Application Details	
	Western Blot, Simple Western, Flow Cytometry, Flow (Intracellular), Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation, CyTOF-ready
	Western Blot 1ug/ml, Simple Western, Flow Cytometry 1ul/1 million cells, Immunohistochemistry, Immunocytochemistry/ Immunofluorescence, Immunoprecipitation 1-2ug/ml, Immunohistochemistry-Paraffin 5ug/ml, Immunohistochemistry-Frozen reported in scientific literature (PMID 25133425), Flow (Intracellular) reported in scientific literature (PMID 24804954), CyTOF-ready

### **Images**

**Application Notes** 

Western Blot: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] -Total protein from various Human, Mouse and Rat cell lines were separated on a 12% gel by SDS-PAGE, transferred to PVDF membrane and blocked in 5% non-fat milk in TBST. The membrane was probed with  $_{250-}$ 1.0 ug/mL anti-IKK-alpha in 1% non-fat milk in TBST and detected with an anti-mouse HRP secondary antibody using chemiluminescence. Image using the BSA free format of this product.

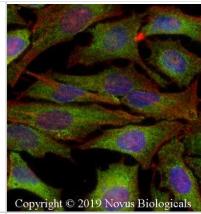




recommended dilution is used per data point.

An 85 kDa band should be observed. In Simple Western only 10-15 uL of the

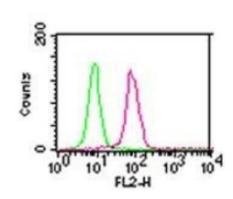
Immunocytochemistry/Immunofluorescence: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] - HeLa cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton X-100. The cells were incubated with anti-IKK alpha Antibody (14A231) at 2 ug/mL overnight at 4C and detected with an anti-mouse DyLight 488 (Green) at a 1:500 dilution. Actin was detected with Phalloidin 568 (Red) at a 1:200 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.



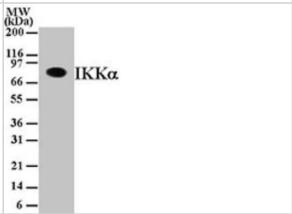
Immunohistochemistry-Paraffin: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] - FFPE human kidney stained with IKKa antibody at 5 ug/mL. Staining of formalin-fixed tissues is enhanced by boiling tissue sections in 10 mM sodium citrate buffer, pH 6.0 for 10-20 min followed by cooling at RT for 20 min.



Flow Cytometry: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] - Intracellular staining of HEK293 cells using 0.5 ug of IKKa antibody (red) and isotype control (green). Intracellular flow kit was used for this test, and an anti-mouse IgG1 PE conjugated secondary antibody.

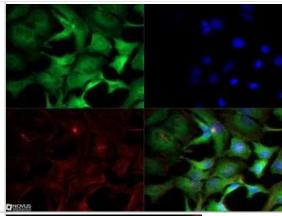


Western Blot: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] - Analysis of IKK in Daudi cell lysate using IKKa monoclonal antibody at 1 ug/mL.

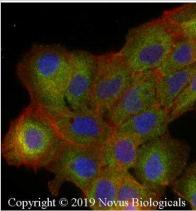




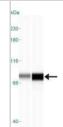
Immunocytochemistry/Immunofluorescence: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] - IKK alpha antibody was tested in HeLa cells with Dylight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and DyLight 550 (red). An antibody dilution of 1:10 was used. Image objective 40X.



Immunocytochemistry/Immunofluorescence: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] - A431 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton X-100. The cells were incubated with anti-IKK alpha Antibody (14A231) at 2 ug/mL overnight at 4C and detected with an anti-mouse DyLight 488 (Green) at a 1:500 dilution. Actin was detected with Phalloidin 568 (Red) at a 1:200 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.



Simple Western: IKK alpha Antibody (14A231) - Azide Free [NBP2-27409] - Analysis using Azide/BSA FREE version of NBP2-27409. Simple Western lane view shows a specific band for IKK alpha in 0.5 mg/mL of Daudi (left) and U937 (right) lysate. This exeriment was performed under reducing conditions using the 12-230 kDa separation system.



#### **Publications**

Fang X, Jeong JH, Long X et al. IKKalpha-mediated biogenesis of miR-196a through interaction with Drosha regulates the sensitivity of cancer cells to radiotherapy. Cell Death Differ. [PMID: 27058318]

#### Details:

Citation using the Azide Free form of this antibody.

Phromnoi K, Reuter S, Sung B et al. A novel pentamethoxyflavone down-regulates tumor cell survival and proliferative and angiogenic gene products through inhibition of IkB kinase activation and sensitizes tumor cells to apoptosis by cytokines and chemotherapeutic agents. Mol Pharmacol. 2011-02-01 [PMID: 20930110]

#### Details:

KBM-5 cells: 1. FLIP (IMG-116-1, -2): WB (Fig 4A) 2. IKKb/IKK2 (IMG-129A): IP Kinase Assay (Fig 2D), IP/WB (Fig 2D). Note, the antibody IP'd an active kinase. The antibody co'IP'd IKKa. 3. IKKa (IMG-136a): WB (Figs 2C, 2D). Note Fig 2D was generated with an IP (IKKb, IMG-129A)/WB (IKKa, IMG-136a) assay. Note: The KBM-5 (human chronic myeloid leukemia) cells were incubated with PMF then treated with TNF-alpha, Figs 2C, 2D, 4A.

Harikumar KB, Sung B, Pandey MK et al. Escin, a pentacyclic triterpene, chemosensitizes human tumor cells through inhibition of nuclear factor-kappaB signaling pathway Mol Pharmacol 2010-05-01 [PMID: 20103608]

#### Details:

This citation used the Alexa Fluor 488 version of this antibody.

Kunnumakkara AB, Ichikawa H, Anand P et al. Coronarin D, a labdane diterpene, inhibits both constitutive and inducible nuclear factor-kappa B pathway activation, leading to potentiation of apoptosis, inhibition of invasion, and suppression of osteoclastogenesis. Mol Cancer Ther. 2008-10-01 [PMID: 18852134] (WB, Human)

#### Details:

IKK-alpha/IKK1 (IMG-136A): WB (human KMB-5 cells), Fig. 2C. 2. IKKb/IKK2 (IMG-129A): WB (human KMB-5 cells), Fig. 2C.

Pandey MK, Sung B, Kunnumakkara AB et al. Berberine modifies cysteine 179 of IkappaBalpha kinase, suppresses nuclear factor-kappaB-regulated antiapoptotic gene products, and potentiates apoptosis. Cancer Res. 2008-07-01 [PMID: 18593939]

#### Details:

IMG-136A (IKKa/IKK1): WB (human multiple myeloma U266 cells), Fig. 3A, C. 2. IMG-129A (IKKb/IKK1): WB (human multiple myeloma U266 cells), Fig. 3A, C.

Saito N, Courtois G, Chiba A et al. Two carboxyl-terminal activation regions of Epstein-Barr virus latent membrane protein 1 activate NF-kappaB through distinct signaling pathways in fibroblast cell lines. J Biol Chem. 2003-11-21 [PMID: 12968033]

Koul D, Yao Y, Abbruzzese JL et al. Tumor suppressor MMAC/PTEN inhibits cytokine-induced NFkappaB activation without interfering with the IkappaB degradation pathway. J Biol Chem. 2001-04-06 [PMID: 11278366]

Chen LW, Egan L, Li ZW et al. The two faces of IKK and NF-kappaB inhibition: prevention of systemic inflammation but increased local injury following intestinal ischemia-reperfusion. Nat Med. 2003-05-01 [PMID: 12692538]

Dejardin E, Droin NM, Delhase M et al. The lymphotoxin-beta receptor induces different patterns of gene expression via two NF-kappaB pathways. Immunity. 2002-10-01 [PMID: 12387745]

Makris C, Roberts JL, Karin M. The carboxyl-terminal region of IkappaB kinase gamma (IKKgamma) is required for full IKK activation. Mol Cell Biol. 2002-09-01 [PMID: 12192055]

Majumdar S, Lamothe B, Aggarwal BB. Thalidomide suppresses NF-kappa B activation induced by TNF and H2O2, but not that activated by ceramide, lipopolysaccharides, or phorbol ester. J Immunol. 2002-03-15 [PMID: 11884428]

Delhase M, Kim SY, Lee H et al. TANK-binding kinase 1 (TBK1) controls cell survival through PAI-2/serpinB2 and transglutaminase 2. Proc Natl Acad Sci U S A. 2012-01-24 [PMID: 22203995]

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#### **Novus Biologicals USA**

10730 E. Briarwood Avenue Centennial, CO 80112 USA

Phone: 303.730.1950 Toll Free: 1.888.506.6887

Fax: 303.730.1966

nb-customerservice@bio-techne.com

#### **Bio-Techne Canada**

21 Canmotor Ave Toronto, ON M8Z 4E6 Canada

Phone: 905.827.6400 Toll Free: 855.668.8722 Fax: 905.827.6402

canada.inquires@bio-techne.com

#### **Bio-Techne Ltd**

19 Barton Lane Abingdon Science Park Abingdon, OX14 3NB, United Kingdom

Phone: (44) (0) 1235 529449 Free Phone: 0800 37 34 15 Fax: (44) (0) 1235 533420 info.EMEA@bio-techne.com

#### **General Contact Information**

www.novusbio.com

Technical Support: nb-technical@bio-

techne.com

Orders: nb-customerservice@bio-techne.com

General: novus@novusbio.com

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NB720-B Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]

NBP1-43319-0.5mg Mouse IgG1 Kappa Isotype Control (P3.6.2.8.1) NBP2-27409APC IKK alpha Antibody (14A231) [Allophycocyanin]

#### Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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