# abcam

### **Product datasheet**

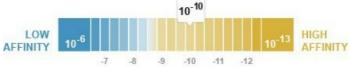
## Anti-IKK beta antibody [EPR6043] ab124957

敲除 验证 重组 RabMAb

#### 49 References 8 图像

概述	
产品名称	Anti-IKK beta <b>抗体</b> [EPR6043]
描述	兔单克隆抗体[EPR6043] to IKK beta
宿主	Rabbit
经测试应 <b>用</b>	适用于: WB, IHC-P
	不适用于: Flow Cyt (Intra),ICC/IF or IP
<b>种属反</b> 应性	与反应: Mouse, Human
	预测可用于: Rat 🔺
免疫原	Synthetic peptide within Human IKK beta aa 1-100 (N terminal). The exact sequence is
	proprietary.
	Database link: <u>014920</u>
	(Peptide available as <u>ab154148</u> )
<b>阳性</b> 对照	HeLa and 293T cell lysates; Human cervix carcinoma and Mouse kidney tissues; HeLa cells.
<b>常</b> 规说 <b>明</b>	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 production
	For more information see here.
	Our RabMAb $^{ extsf{B}}$ technology is a patented hybridoma-based technology for making rabbit
	monoclonal antibodies. For details on our patents, please refer to <b><u>RabMAb<sup>®</sup> patents</u></b> .

性能	
形式	Liquid
存 <b>放</b> 说明	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
解离常数(K <sub>D</sub> )	$K_{D} = 1.28 \times 10^{-10} M$
	10 <sup>-10</sup>



#### Learn more about K<sub>D</sub>

存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture supernatant
纯 <b>度</b>	Protein A purified
克隆	单 <b>克隆</b>
克 <b>隆</b> 编号	EPR6043
同种型	lgG

应用

#### The Abpromise guarantee Abpromise ™承诺保证使用ab124957于以下的经测试应用

#### "应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

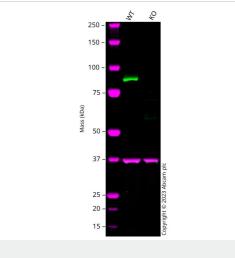
应用	Ab评论	说明
WB		1/1000 - 1/10000. Detects a band of approximately 85 kDa (predicted molecular weight: 87 kDa).
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. (Heat to 98°C, allow to cool for 10-20 minutes)

应**用**说明

Is unsuitable for Flow Cyt (Intra), ICC/IF or IP.

Acts as part of the IKK complex in the conventional pathway of NF-kappa-B activation and phosphorylates inhibitors of NF-kappa-B thus leading to the dissociation of the inhibitor/NF-kappa-B complex and ultimately the degradation of the inhibitor. Also phosphorylates NCOA3.
Highly expressed in heart, placenta, skeletal muscle, kidney, pancreas, spleen, thymus, prostate, testis and peripheral blood.
Belongs to the protein kinase superfamily. Ser/Thr protein kinase family. I-kappa-B kinase subfamily. Contains 1 protein kinase domain.
Upon cytokine stimulation, phosphorylated on Ser-177 and Ser-181 by MEKK1 and/or MAP3K14/NIK; which enhances activity. Once activated, autophosphorylates on the C-terminal serine cluster; which decreases activity and prevents prolonged activation of the inflammatory response. Acetylation of Thr-180 by Yersinia yopJ prevents phosphorylation and activation, thus blocking the Hkappa-B pathway. Ubiquitinated. Monoubiquitination involves TRIM21 that leads to inhibition of Tax-induced NF- kappa-B signaling. According to PubMed:19675099, 'Ser-163' does not serve as a monoubiquitination site. According to PubMed:16267042, ubiquitination on 'Ser-163' modulates phosphorylation on C-terminal serine residues. Monoubiquitination by TRIM21 is dirupted by Yersinia yopJ.

#### 图片



Western blot - Anti-IKK beta antibody [EPR6043] (ab124957)

**All lanes :** Anti-IKK beta antibody [EPR6043] (ab124957) at 1/1000 dilution

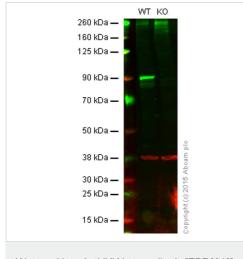
Lane 1 : Wild-type HeLa cell lysate Lane 2 : IKKa/b knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

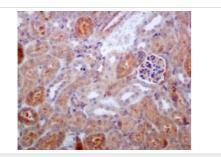
Performed under reducing conditions.

Predicted band size: 87 kDa Observed band size: 85 kDa

Western blot: Anti-IKKa/b antibody [EPR6043] (ab124957) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab124957 was shown to bind specifically to IKKa/b. A band was observed at 85 kDa in wild-type HeLa cell lysates with no signal observed at this size in IKKa/b knockout cell line. To generate this image, wild-type and IKKa/b knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween<sup>®</sup> 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



Western blot - Anti-IKK beta antibody [EPR6043] (ab124957)



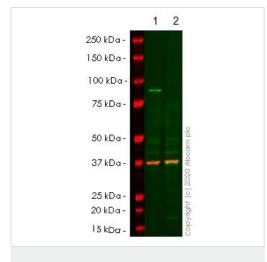
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IKK beta antibody [EPR6043] (ab124957)

Lane 1: Wild-type HAP1 cell lysate (20 μg) Lane 2: IKK beta knockout HAP1 cell lysate (20 μg) Lanes 1 and 2: Merged signal (red and green). Green - ab124957 observed at 90 kDa. Red - loading control, <u>ab8245</u>, observed at 37 kDa.

ab124957 was shown to specifically react with IKK beta when IKK beta knockout samples were used. Wild-type and IKK beta knockout samples were subjected to SDS-PAGE. ab124957 and **ab8245** (loading control to GAPDH) were diluted 1/500 and 1/2000 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 1/10,000 dilution for 1 h at room temperature before imaging.

ab124957, at 1/100 dilution staining IKK beta in paraffin-embedded Mouse kidney tissue, by Immunohistochemistry.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Western blot - Anti-IKK beta antibody [EPR6043] (ab124957) **All lanes :** Anti-IKK beta antibody [EPR6043] (ab124957) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate Lane 2 : IKBKB CRISPR/Cas9 edited HeLa cell lysate

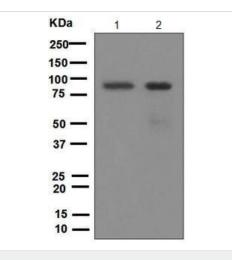
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 87 kDa Observed band size: 90 kDa

Lanes 1-2: Merged signal (red and green). Green - ab124957 observed at 90 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (<u>ab8245</u>) observed at 37 kDa.

ab124957 was shown to react with IKK beta in wild-type HeLa cells in western blot. The band observed in CRISPR/Cas9 edited cell line **ab264847** (CRISPR/Cas9 edited cell lysate **ab257228**) lane below 90kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type HeLa and IKBKB CRISPR/Cas9 edited HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab124957 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at a 1 in 500 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup>800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup>680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-IKK beta antibody [EPR6043] (ab124957) **All lanes :** Anti-IKK beta antibody [EPR6043] (ab124957) at 1/1000 dilution

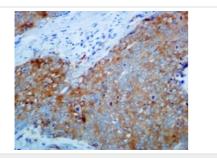
Lane 1 : HeLa cell lysates Lane 2 : 293T cell lysates

Lysates/proteins at 10 µg per lane.

#### Secondary

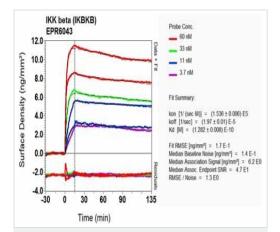
All lanes : HRP labelled Goat anti-Rabbit IgG at 1/2000 dilution

Predicted band size: 87 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IKK beta antibody [EPR6043] (ab124957) ab124957, at 1/100 dilution staining IKK beta in paraffin-embedded Human cervix carcinoma tissue, by Immunohistochemistry.

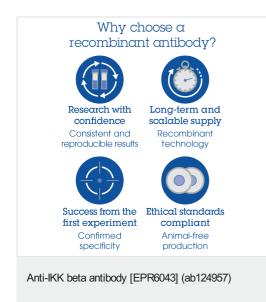
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



OI-RD Scanning - Anti-IKK beta antibody [EPR6043] (ab124957) Equilibrium disassociation constant (K<sub>D</sub>)

Learn more about K<sub>D</sub>

#### Click here to learn more about KD



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