#### Product datasheet Anti-IkB Alpha/NFKBIA Antibody Catalog Number: PB9291



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Basic Information	
Product Name	Anti-IkB Alpha/NFKBIA Antibody
Gene Name	NFKBIA
Source	Rabbit
lsotype	IgG
Species Reactivity	human, mouse, rat
Tested Application	WB, IHC
Contents	500 ug/ml antibody with PBS $_{ m o}$ 0.02% NaN3 , 1 mg BSA and 50% glycerol.
Immunogen	E.coli-derived human IKB alpha recombinant protein (Position: Q3-Q112). Human IKB alpha shares 87% and 86% amino acid (aa) sequence identity with mouse and rat IKB alpha, respectively.
concentration	500 ug/ml
Purification	Immunogen affinity purified.
Observed MW	39KD
Dilution Ratios	Western blot(WB):1:500-2000Immunohistochemistry in paraffin section (IHC):1:50-400(Boiling the paraffin sections in 10mM citrate buffer,pH6.0,or PH8.0 EDTA repair liquid for 20mins is required for the staining of formalin/paraffin sections.) Optimal working dilutionsmust be determined by end user.

### **Storage**

12 months from date of receipt,  $-20^{\circ}$ C as supplied. 6 months 2 to 8°C after reconstitution. Avoid repeated freezing and thawing.

# **Background Information**

NFKBIA, also called IKBA or MAD-3, is one member of a family of cellular proteins that function to inhibit the NF- $\kappa$ B transcription factor. It is mapped to 14q13.2. NFKBIA inhibits NF- $\kappa$ B by masking the nuclear localization signals(NLS) of NF- $\kappa$ B proteins and keeping them sequestered in an inactive state in the cytoplasm. It moves between the cytoplasm and the nucleus via a nuclear localization signal and CRM1-mediated nuclear export. The effect of the nonpathogenic bacteria is specific to the SCF complex substrates CTNNB1 and NFKBIA. This may help to explain the beneficial effects of treatment of inflammatory bowel disease with nonpathogenic probiotic enteric organisms. In addition, NFKBIA blocks the ability of NF- $\kappa$ B transcription factors to bind to DNA, which is required for NF- $\kappa$ B's proper functioning.

## **Selected Validation Data**

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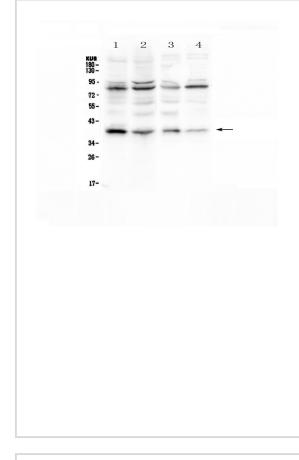


Figure 1. Western blot analysis of IKB Alpha using anti- IKB Alpha antibody (PB9291). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions. Lane 1: human Jurkat whole cell lysates, Lane 2: human MDA-MB-453 whole cell lysates, Lane 3: human SW620 whole cell lysates, Lane 4: human A549 whole cell lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-IKB Alpha antigen affinity purified polyclonal antibody (Catalog # PB9291) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for IKB Alpha at approximately 39KD. The expected band size for IKB Alpha is at 36KD.

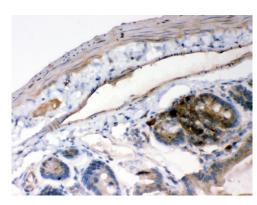


Figure 2. IHC analysis of IKB alpha using anti-IKB alpha antibody (PB9291).IKB alpha was detected in paraffin-embedded section of Mouse Intestine Tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1µg/ml rabbit anti-IKB alpha Antibody (PB9291) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.