

## Anti-IKKε pT501 (RABBIT) Antibody

IKK E phospho T501 Antibody Catalog # ASR5178

## **Specification**

## Anti-IKK pT501 (RABBIT) Antibody - Product Information

Host Conjugate Target Species Reactivity Clonality Application Application Note	Rabbit Unconjugated Human Human Polyclonal WB, E, I, LCI IKKE pT501 antibody is tested in ELISA, western blotting, and although not tested, this antibody is likely functional in immunohistochemistry and immunoprecipitation. An 85 kDa band corresponding to human IKKe is detected. HeLa cells or TNF inducible KBM-5 cells can be used as a positive control. Researchers should determine optimal titers for other applications.
Physical State Buffer	Liquid (sterile filtered) 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Immunogen	IKKe phospho peptide corresponding to a region of the human protein surrounding pT501 conjugated to KLH.
Preservative	0.1% (w/v) Sodium Azide

# Anti-IKKε pT501 (RABBIT) Antibody - Additional Information

Gene ID 9641

Other Names 9641

#### Purity

Anti-IKKɛ pT501 antibody was affinity purified from monospecific antiserum by immunoaffinity purification against the phosphopeptide and cross adsorption against the non-phosphorylated form of the peptide followed by non-adsorption against a non-specific peptide backbone to further reduce non-specific reactivity. This phospho specific polyclonal antibody is specific for phosphorylated pT501 human IKKe. Reactivity with non-phosphorylated IKKe is minimal. Cross reactivity with pT501 phosphorylated IKKe from mouse, rat or other species has not been determined.

### Storage Condition

Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted



liquid. Dilute only prior to immediate use.

#### Precautions Note

This product is for research use only and is not intended for therapeutic or diagnostic applications.

## Anti-IKKε pT501 (RABBIT) Antibody - Protein Information

## Name IKBKE

Synonyms IKKE, IKKI, KIAA0151

#### Function

Serine/threonine kinase that plays an essential role in regulating inflammatory responses to viral infection, through the activation of the type I IFN, NF-kappa-B and STAT signaling. Also involved in TNFA and inflammatory cytokines, like Interleukin-1, signaling. Following activation of viral RNA sensors, such as RIG-I- like receptors, associates with DDX3X and phosphorylates interferon regulatory factors (IRFs), IRF3 and IRF7, as well as DDX3X. This activity allows subsequent homodimerization and nuclear translocation of the IRF3 leading to transcriptional activation of pro-inflammatory and antiviral genes including IFNB. In order to establish such an antiviral state, IKBKE forms several different complexes whose composition depends on the type of cell and cellular stimuli. Thus, several scaffolding molecules including IPS1/MAVS, TANK, AZI2/NAP1 or TBKBP1/SINTBAD can be recruited to the IKBKE-containing-complexes. Activated by polyubiquitination in response to TNFA and interleukin-1, regulates the NF-kappa-B signaling pathway through, at least, the phosphorylation of CYLD. 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. In addition, is also required for the induction of a subset of ISGs which displays antiviral activity, may be through the phosphorylation of STAT1 at 'Ser-708'. Phosphorylation of STAT1 at 'Ser-708' seems also to promote the assembly and DNA binding of ISGF3 (STAT1:STAT2:IRF9) complexes compared to GAF (STAT1:STAT1) complexes, in this way regulating the balance between type I and type II IFN responses. Protects cells against DNA damage-induced cell death. Also plays an important role in energy balance regulation by sustaining a state of chronic, low-grade inflammation in obesity, wich leads to a negative impact on insulin sensitivity. Phosphorylates AKT1.

#### **Cellular Location**

Cytoplasm. Nucleus. Nucleus, PML body. Note=Targeting to PML nuclear bodies upon DNA damage is TOPORS-dependent (PubMed:20188669) Located diffusely throughout the cytoplasm but locates to punctate cytoplasmic bodies when coexpressed with TRIM6 (PubMed:24882218)

#### **Tissue Location**

Highly expressed in spleen followed by thymus, peripheral blood leukocytes, pancreas, placenta. Weakly expressed in lung, kidney, prostate, ovary and colon

### Anti-IKKε pT501 (RABBIT) Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

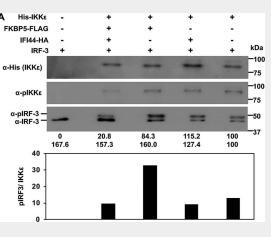
- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation



Flow Cytomety

<u>Cell Culture</u>

## Anti-IKK pT501 (RABBIT) Antibody - Images



IFI44 decreases the kinase activity of IKKβ and IKKε. Human 293T cells were silenced for IFI44, or for FKBP5, and were transfected with plasmids expressing His-IKK $\epsilon$  (A) or MYC-IKK $\beta$  (B), together with IFI44-HA, and FKBP5-FLAG expression plasmids. At 24 hpt, IKK $\epsilon$  (A) and IKK $\beta$  (B) complexes were purified with anti-His and anti-MYC antibodies, respectively, and these complexes were assayed in kinase assays using IRF-3 (for the IKK  $\epsilon$  complexes shown in panel A) and IkB $\alpha$  (for the IKKβ complexes shown in panel B) as substrates. The levels of phosphorylated and unphosphorylated forms of IRF-3 (panel A, bottom blot) and IkB $\alpha$  (panel B, third and fourth blots) were analyzed by Western blotting using specific antibodies. Levels of IKKE were analyzed using an anti-His-specific antibody (A, first blot) and anti-pIKK $\epsilon$  (A, second blot), and levels of IKK $\beta$  were analyzed using an anti-MYC-specific antibody (B, first blot) and anti-pIKK $\beta$  (B, second blot). Western blots were quantified by densitometry using ImageJ software (v1.46). Protein expression levels in cells expressing IKKE (A) and IKKB (B) alone were assigned a value of 100% for comparisons with the levels of expression in cells expressing the different combinations of IKK $\epsilon$ /IFI44/FKBP5 (A) or IKK $\beta$ /IFI44/FKBP5 (B) (numbers are indicated below each plot). pIRF-3 and IRF-3 levels (observed in the same bottom blot in panel A) and plkB $\alpha$  and lkB $\alpha$  (third and bottom blot in panel B) are represented with numbers below each blot. Levels of pIRF-3 and pIkB $\alpha$ normalized to the levels of IKK $\varepsilon$  and IKK $\beta$  are represented in the bottom graphs in panels A and B, respectively. Molecular weight markers are indicated (in kilodaltons) on the right. Figure provided by CiteAb. Source: MBio, PMID: 31455651.

### Anti-IKK pT501 (RABBIT) Antibody - Background

Nuclear Factor kappa B (NF-kB) is a ubiquitous transcription factor and an essential mediator of gene expression during the activation of immune and inflammatory responses. NF-kB mediates the expression of a great variety of genes in response to extracellular stimuli. NF-kB is associated with IkB proteins in the cytoplasm of the cell, which inhibit NF-kB activity. IkB proteins are phosphorylated by an IkB kinase complex consisting of at least three proteins, IKKa, IKKb, and IKKg. Isolated from a cDNA library of LPS-stimulated mouse macrophage cells, a novel molecule in the IKK complex has been recently identified and designated IKKi and/or IKKe. IKKepsilon is required for the activation of NF-kB by mitogens and T cell receptors but not by TNF $\alpha$  or IL-1. LPS increases the IKKe mRNA level in mouse macrophage cell lines. This protein has significant sequence homology with kinase domains of IKKa and IKKb. Overexpression of wild type IKKe in cells phosphorylates Ser32 and Ser36 of IkBa. Anti-IKK $\alpha$  pT501 antibody is ideal for investigators involved in NFkappaB and apoptosis research.