## **Technical Data Sheet**

# Purified Mouse Anti-Human IKBα (pS32/pS36)

#### **Product Information**

**Material Number:** 551818 Size: 50 µg 0.5 mg/mlConcentration: 39A1413 Clone:

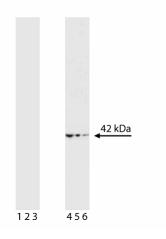
Phosphorylated Human IκBα aa. 32, 36 Immunogen:

Isotype: Mouse IgG1 Reactivity: QC Testing: Human

Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

#### Description

NF-κB is a transcription factor which is a member of the mammalian NF-κB/Rel family of proteins. Members of this family are involved in the regulation of cell proliferation, immune function, as well as development. NF-kB is normally found in the cytoplasm and remains in an inactive state by its association with an inhibitory protein, IκB. Stimulation of NF-κB by a variety of inducers causes the degradation of IκBs and translocation of NF-  $\kappa B$  to the nucleus and activation of the target gene. If  $\kappa B \alpha$  is a member of the I  $\kappa B$  family of proteins including I  $\kappa B \beta$ ,  $I\kappa$ Βγ,  $I\kappa$ Βε, BcI-3, and the precursors of NF- $\kappa$ B1 (p105), and NF- $\kappa$ B2 (p100).  $I\kappa$ Bα is the best characterized member of the family and has been shown to contain three different structural domains: an N-terminal region, an amino acid internal region containing ankyrin repeats, and a C-terminal region containing a PEST domain. In resting cells, IκBα binds to and maintains NF-κB in the cytoplasm by blocking the nuclear localization sequences of NF-κB. In the response to an extracellular signal, IκBα is phosphorylated and subsequently degraded via the ubiquination-proteasome pathway, allowing NF-κB to translocate to the nucleus. Once in the nucleus, NF-κB can induce the transcription of IκB $\alpha$  thereby renewing the cycle so that IκB $\alpha$  can form a complex with NF-κB and maintains it in its cytoplasmic location. IκB $\alpha$  -/- mice have been shown to die soon after birth and show an increased level of NF-kB activity. Furthermore, in Hodgekin's lymphoma (HL) a high constitutive level of NF- $\kappa$ B has been reported in samples in which clonal deleterious mutations were detected in the  $I\kappa$ B $\alpha$  gene. The exact role that  $I\kappa B\alpha$  plays in the pathogenic process which leads to HL remains to be elucidated.  $I\kappa B\alpha$  migrates at ~42 kDa in SDS/PAGE, while the deduced molecular weight based upon its cDNA sequence is ~36 kDa (SWISS PROT Accession number P25963). The antibody specifically recognizes the phosphorylated form of human IκBα; it does not recognize the unphosphorylated form. A synthetic peptide containing phosphorylated serines at amino acid residues of 32 and 36 of human IκBα was used as the immunogen.



Western blot analysis of IκBα. Untreated (lanes 1-3) or TNF-q-treated (20 ng/ml for 10 minutes, Jane 4-6) Jurkat cells were lysed and probed with anti-human IκBα (clone 39A1413. Cat. No. 551818) at concentrations of 2.0 (lanes 1,4), 1.0 (lanes 2.5), and 0.5  $\mu$ g/ml (lanes 3,6). I $\kappa$ B $\alpha$  is identified as a band of ~42 kDa in the TNF-α treated Jurkat cells, but not in the untreated cells,

## **Preparation and Storage**

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4°C.

### **Application Notes**

Application

1	Application				
	Western blot	Routinely Tested			
	Immunoprecipitation	Reported			

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#### **Recommended Assay Procedure:**

Applications include western blot analysis  $(0.5-2.0 \,\mu\text{g/ml})$ . Additional applications not tested at BD Biosciences Pharmingen include immunoprecipitation. Treatment of Jurkat T cells with TNF- $\alpha$  is recommended as a positive control.

## **Suggested Companion Products**

Catalog Number	Name	Size	Clone
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)

#### **Product Notices**

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

#### References

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