Store at –20°C

#9246

Phospho-IκBα (Ser32/36) (5A5) Mouse mAb

Cell Signaling TECHNOLOGY®

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For Research Use Only. Not For Use In Diagnostic Procedures.

Entrez-Gene ID #4792
Swiss-Prot Acc. #P25963

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W Endogenous	H, M, R, Mk, (Dg, Pg, B, Guinea pig)	40 kDa	Mouse IgG1**

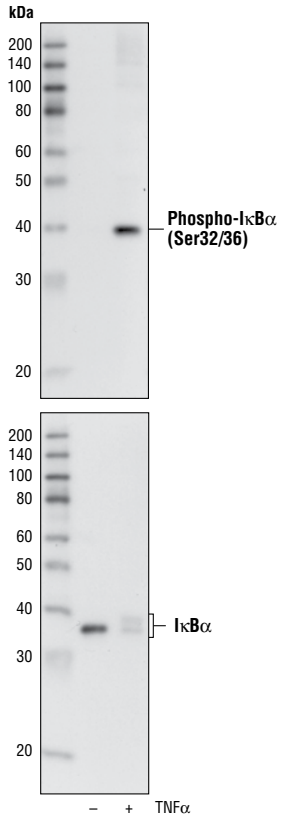
Background: The NF-κB/Rel transcription factors are present in the cytosol in an inactive state complexed with the inhibitory IκB proteins (1-3). Activation occurs via phosphorylation of IκBα at Ser32 and Ser36 followed by proteasome-mediated degradation that results in the release and nuclear translocation of active NF-κB (3-7). IκBα phosphorylation and resulting Rel-dependent transcription are activated by a highly diverse group of extracellular signals including inflammatory cytokines, growth factors and chemokines. Kinases that phosphorylate IκB at these activating sites have been identified (8).

Specificity/Sensitivity: Phospho-IκBα (Ser32/36) (5A5) Mouse mAb detects endogenous levels of IκBα only when phosphorylated at Ser32/36.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser32/36 of human IκBα.

Background References:

(1) Baeuerle, P.A. and Baltimore, D. (1988) *Science* 242, 540–546.
(2) Beg, A.A. et al. (1993) *Genes Dev.* 7, 2064–2070.
(3) Finco, T.S. et al. (1994) *Proc. Natl. Acad. Sci. USA* 91, 11884–11888.
(4) Brown, K. et al. (1995) *Science* 267, 1485–1488.
(5) Brockman, J.A. et al. (1995) *Mol. Cell. Biol.* 15, 2809–2818.
(6) Traenckner, E.B. et al. (1995) *EMBO J.* 14, 2876–2883.
(7) Chen, Z.J. et al. (1996) *Cell* 84, 853–862.
(8) Karin, M. and Ben-Neriah, Y. (2000) *Annu. Rev. Immunol.* 18, 621–663.



Western blot analysis of extracts from NIH/3T3 cells, untreated or TNF-α-treated (#2169, 20 ng/ml) for 5 minutes, using Phospho-IκBα (Ser32/36) (5A5) Mouse mAb (upper) or IκBα (L35A5) Mouse mAb (Amino-terminal Antigen) #4814 (lower).

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. *Do not aliquot the antibody.*

***Species cross-reactivity is determined by western blot.**

****Anti-mouse secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

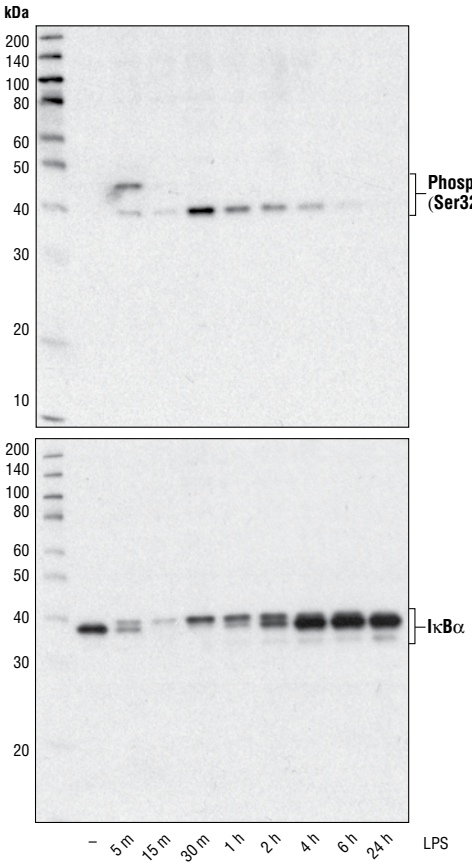
Western blotting	1:1000

For product specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended complementary products.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—Horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Western blot analysis of extracts from THP-1 cells, differentiated with TPA (#9905, 80 nM for 24 h) and treated with 1 µg/ml LPS for the indicated times, using Phospho-IκBα (Ser32/36) (5A5) Mouse mAb (upper) and IκBα (L35A5) Mouse mAb (Amino-terminal Antigen) #4814 (lower).