Revision 6

#5348 Store at -20C

Phospho-c-Fos (Ser32) (D82C12) XP® Rabbit mAb

Cell Signaling TECHNOLOGY®

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For Research Use Only. Not for Use in Diagnostic Procedures.

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
WB, IP, IF-F, IF-IC, FC-FP, ChIP, ChIP-seq	H M R	Endogenous	62	Rabbit IgG	#P01100	2353

Product Usage Information

For optimal ChIP and ChIP-seq, use 10 µl of antibody and 10 µg of chromatin (approximately 4 x 10^6 cells) per IP. This antibody has been validated using SimpleChIP® Enzymatic Chromatin IP Kits.

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:100
Immunofluorescence (Frozen)	1:100 - 1:400
Immunofluorescence (Immunocytochemistry)	1:100 - 1:400
Flow Cytometry (Fixed/Permeabilized)	1:200 - 1:800
Chromatin IP	1:50
Chromatin IP-seq	1:50

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.

For a carrier free (BSA and azide free) version of this product see product #99325.

Specificity / Sensitivity

Phospho-c-Fos (Ser32) (D82C12) XP® Rabbit mAb detects endogenous levels of c-Fos protein only when phosphorylated on Ser32. The antibody does not cross-react with other Fos proteins, including FosB, FRA1 and FRA2.
Non-specific phosphatase-sensitive cytoskeletal staining has been observed by immunofluorescence in P301L Tau Transgenic mouse brain.

Species predicted to react based on 100% sequence homology:

Hamster, Monkey, Bovine, Pig, Horse

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to Ser32 of human c-Fos protein.

Background

The Fos family of nuclear oncogenes includes c-Fos, FosB, Fos-related antigen 1 (FRA1), and Fos-related antigen 2 (FRA2) (1). While most Fos proteins exist as a single isoform, the FosB protein exists as two isoforms: full-length FosB and a shorter form, FosB2 (Delta FosB), which lacks the carboxy-terminal 101 amino acids (1-3). The expression of Fos proteins is rapidly and transiently induced by a variety of extracellular stimuli, including growth factors, cytokines, neurotransmitters, polypeptide hormones, and stress. Fos proteins dimerize with Jun proteins (c-Jun, JunB, and JunD) to form Activator Protein-1 (AP-1), a transcription factor that binds to TRE/AP-1 elements and activates transcription. Fos and Jun proteins contain the leucine-zipper motif that mediates dimerization and an adjacent basic domain that binds to DNA. The various Fos/Jun heterodimers differ in their ability to transactivate AP-1 dependent genes. In addition to increased expression, phosphorylation of Fos proteins by Erk kinases in response to extracellular stimuli may further increase transcriptional activity (4-6). Phosphorylation of c-Fos at Ser32 and Thr232 by Erk5 increases protein stability and nuclear localization (5). Phosphorylation of FRA1 at Ser252 and Ser265 by Erk1/2 increases protein stability and leads to overexpression of FRA1 in cancer cells (6). Following growth factor stimulation, expression of FosB and c-Fos in quiescent fibroblasts is immediate, but very short-lived, with protein levels dissipating after several hours (7). FRA1 and FRA2 expression persists longer, and appreciable levels can be detected in asynchronously growing cells (8). Deregulated expression of c-Fos, FosB, or FRA2 can result in neoplastic cellular transformation; however, Delta FosB lacks the ability to transform cells (2,3).

Background References

1. Tulchinsky, E. (2000) *Histol Histopathol* 15, 921-8.
2. Dobrazanski, P. et al. (1991) *Mol Cell Biol* 11, 5470-8.
3. Nakabeppu, Y. and Nathans, D. (1991) *Cell* 64, 751-9.
4. Rosenberger, S.F. et al. (1999) *J Biol Chem* 274, 1124-30.
5. Sasaki, T. et al. (2006) *Mol Cell* 24, 63-75.
6. Basbous, J. et al. (2007) *Mol Cell Biol* 27, 3936-50.
7. Kovary, K. and Bravo, R. (1991) *Mol Cell Biol* 11, 2451-9.
8. Kovary, K. and Bravo, R. (1992) *Mol Cell Biol* 12, 5015-23.

Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer

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

Applications Key

WB: Western Blotting **IP:** Immunoprecipitation **IF-F:** Immunofluorescence (Frozen)
IF-IC: Immunofluorescence (Immunocytochemistry) **FC-FP:** Flow Cytometry (Fixed/Permeabilized)
ChIP: Chromatin IP **ChIP-seq:** Chromatin IP-seq

Cross-Reactivity Key

H: human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **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
GP: Guinea Pig **Rab:** rabbit **All:** all species expected

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Limited Uses