Revision 1

Store at -20C

#4767

NF-κB p65 Antibody Sampler Kit

1 Kit (5 x 20 microliters)

Cell Signaling TECHNOLOGY®

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Product Includes	Product #	Quantity	Mol. Wt	Isotype/Source
Phospho-NF-κB p65 (Ser536) (93H1) Rabbit mAb	3033	20 µl	65 kDa	Rabbit IgG
Acetyl-NF-κB p65 (Lys310) (D2S3J) Rabbit mAb	12629	20 µl	65 kDa	Rabbit IgG
NF-κB p65 (L8F6) Mouse mAb	6956	20 µl	65 kDa	Mouse IgG2b
NF-κB p65 (D14E12) XP® Rabbit mAb	8242	20 µl	65 kDa	Rabbit IgG
Phospho-NF-κB p65 (Ser468) Antibody	3039	20 µl	65 kDa	Rabbit
Anti-rabbit IgG, HRP-linked Antibody	7074	100 µl		Goat
Anti-mouse IgG, HRP-linked Antibody	7076	100 µl		Horse

Please visit cellsignal.com for individual component applications, species cross-reactivity, dilutions, protocols, and additional product information.

Description

The NF-κB p65 Antibody Sampler Kit contains reagents to examine NF-κB p65/RelA phosphorylation at Ser468 and Ser536; acetylation at Lys310; and total p65 levels.

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.

Background

Transcription factors of the nuclear factor κB (NF-κB)/Rel family play a pivotal role in inflammatory and immune responses (1,2). There are five family members in mammals: RelA, c-Rel, RelB, NF-κB1 (p105/p50), and NF-κB2 (p100/p52). Both p105 and p100 are proteolytically processed by the proteasome to produce p50 and p52, respectively. Rel proteins bind p50 and p52 to form dimeric complexes that bind DNA and regulate transcription. In unstimulated cells, NF-κB is sequestered in the cytoplasm by IκB inhibitory proteins (3-5). NF-κB-activating agents can induce the phosphorylation of IκB proteins, targeting them for rapid degradation through the ubiquitin-proteasome pathway and releasing NF-κB to enter the nucleus where it regulates gene expression (6-8). NIK and IKKα (IKK1) regulate the phosphorylation and processing of NF-κB2 (p100) to produce p52, which translocates to the nucleus (9-11).

RelA/p65 is a subunit of the NF-κB transcription complex, which plays a crucial role in inflammatory and immune responses. The complex is composed of various homodimeric and heterodimeric Rel family member combinations, the activity of which is modulated by post-translational modifications including phosphorylation and acetylation. p65 phosphorylation by PKA and/or MSK1 at Ser276 allows for increased interaction with the transcriptional coactivator p300/CBP to enhance transcriptional activity. NF-κB dimer assembly with IκB, as well as its DNA binding and transcriptional activities, are regulated by p300/CBP acetyltransferases that principally target Lys218, Lys221 and Lys310 (12-14). This process is reciprocally regulated by histone deacetylases (HDACs); several HDAC inhibitors have been shown to activate NF-κB (12-14). T-cell co-stimulation and Calyculin A have both been shown to increase Ser468 phosphorylation (15,16). IKKβ (but not IKKα) and GSK-3β both target this site (16,17), which appears to have a negative regulatory role not involving inhibition of nuclear translocation after TNF-α or IL-1β stimulation (17). p65 phosphorylation at Ser536 regulates activation, nuclear localization, protein-protein interactions, transcriptional activity, and apoptosis (18-22).

Background References

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