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ProductInformation

NF-κB (p50)
Human, Recombinant
Expressed in bacteria

Product Number **N 9909**
Storage Temperature –70 °C

Product Description

Recombinant human NF-κB (p50) is produced from a cDNA sequence encoding the human homologue of rabbit p50.[1] It is expressed in bacteria. NF-κB (p50) comprises 453 amino acid residues; the amino terminal domain shows strong homology to the proto-oncogene c-rel. Human recombinant NF-κB (p50) is suitable for use in gel shift assays, in footprinting assays, and for transcriptional activation *in vitro*.

Organisms must be able to respond rapidly and effectively to changes in their environment. Most types of signaling molecules induce cellular responses by binding to specific cell-surface receptors. These receptors respond to occupancy by undergoing structural or biochemical changes that can be transmitted to the interior of the cell. One of the most common responses to receptor ligation is the synthesis of new proteins through alteration of the pattern of gene expression. Consequently, the relatively few transcription factors that regulate inducible gene expression can be the targets for many distinct signal transduction pathways, triggered by a wide variety of stimuli.[2] One important transcription factor that plays a pivotal role in many cellular responses to environmental changes is NF-κB, a heterodimeric transcription factor composed of p50 (50 kDa) and p65 (65 kDa) subunits. NF-κB can be activated in many cell types and is thought to regulate a wide variety of genes.[3-5] An extensive set of genes with putative NF-κB-binding sites has been identified, and in many of these, the NF-κB sites appear crucial to the regulation of transcription. A wide range of stimuli lead to translocation of NF-κB from the cytoplasm to the nucleus, where it appears in an active form capable of binding decameric κB sequences motifs.[6] Putative cellular target genes are largely involved in the acute-phase response, inflammation, lymphocyte activation (specific and nonspecific immune responses), and cell growth and differentiation. These genes include cell-surface molecules involved in immune function such as immunoglobulin κ light chain, class I and II major histocompatability complex (MHC), and cytokines such as interleukin-1β (IL-1β), IL-2, IL-6, interferon-β (IFNβ), and tumor necrosis factor α (TNFα).[6] Under normal conditions, NF-κB is bound to an inhibitor protein, I-κB, that sequesters NF-κB in the cytosol. Activation of NF-κB involves its dissociation from I-κB followed by translocation of the p50-p65 heterodimer to the nucleus, where it directly binds to its cognate DNA sequences.[7,8] p50 and p65 are members of a larger NF-κB/Rel family of transcription factors, that in vertebrates includes at least five members: NFKB1 (p50 and its precursor, p105), NFKB2 (p52 and its precursor, p100), p65 (RelA), c-Rel (Rel), and RelB.[9] As dimers, all five proteins can form complexes with κB DNA sequence motifs, and all have been shown to affect transcription of κB reporter genes positively or negatively when assayed following transfection.[6] The high level of interest in Rel-based transcription factors is due to their broad role in inducing and coordinating the expression of genes of significant biomedical importance, such as those encoding inflammatory cytokines, chemokines, interferons, MHC proteins, growth factors, cell adhesion molecules, and viruses.

Reagents

Recombinant human NF-κB (p50) is supplied as a solution in 20 mM HEPES buffer, pH 7.9, containing 50 mM NaCl, 5 mM DTT, 0.5 mM PMSF, 10 μM zinc acetate, 0.1% (w/v) Nonidet P40, and 10% glycerol.

Precautions and Disclaimer

This product is for laboratory research use only. It is not suitable for human therapeutic or diagnostic use. Please consult the Material Safety Data sheet for handling recommendations prior to working with this material.

Storage/Stability
Recombinant human NF-κB (p50) is stable for at least six months when stored at −70 °C. Avoid repeated freeze-thaw cycles. Dilution buffers should include fresh 5 mM DTT which is required for DNA binding activity.

Product Profile
Unit Definition: 1 gsu (gel shift unit) equals the amount of protein required to shift the NF-κB oligonucleotide (5′AGT TGA GGG GAC TTT CCC AGG C3′) under defined binding and electrophoretic conditions. Each vial contains 50 gsu of recombinant human NF-κB (p50).

The ratio of DNA to NF-κB (p50) required for complete gel shift is between 3 and 10, which corresponds to a range of 20% to 66% active protein.

References

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