Recombinant IKKβ protein

Catalog No: 81066, 81766
Lot No: 25017001
Expressed In: Baculovirus

Quantity: 20,1000 µg
Concentration: 0.15 µg/µl
Source: Human

Buffer Contents: Recombinant IKKβ protein is supplied in 25 mM HEPES-NaOH pH 7.5, 300 mM NaCl, 10% glycerol, 0.04% Triton X-100 and 0.5 mM TCEP.

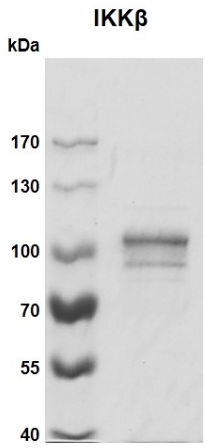
Background: IKKβ (Inhibitor Of Nuclear Factor Kappa B Kinase Subunit Beta), also known as IKBKB, I-Kappa-B-Kinase 2, IKK-Beta and NFKBIKB, is a serine kinase that plays an essential role in the NF-kappa-B signaling pathway which is activated by multiple stimuli such as inflammatory cytokines, bacterial or viral products, DNA damages or other cellular stresses. It acts as part of the canonical IKK complex in the conventional pathway of NF-kappa-B activation and phosphorylates inhibitors of NF-kappa-B on 2 critical serine residues. These modifications allow polyubiquitination of the inhibitors and subsequent degradation by the proteasome. In turn, free NF-kappa-B is translocated into the nucleus and activates the transcription of hundreds of genes involved in immune response, growth control, or protection against apoptosis. In addition to the NF-kappa-B inhibitors, IKKβ can also phosphorylate several other components of the signaling pathway including NEMO/IKBKG, NF-kappa-B subunits RELA and NFKB1, as well as IKK-related kinases TBK1 and IKBKE. IKK-related kinase phosphorylations may prevent the overproduction of inflammatory mediators since they exert a negative regulation on canonical IKKs. IKKβ can phosphorylate FOXO3, mediating the TNF-dependent inactivation of this pro-apoptotic transcription factor.

Protein Details: Recombinant human IKKβ protein was expressed in a baculovirus expression system as the full length protein (accession number NP_001547.1) with an N-terminal FLAG tag. The molecular weight of the protein is 87.8 kDa.

Application Notes: Recombinant IKKβ protein is suitable for use in the study of enzyme kinetics, inhibitor screening, and selectivity profiling.

Kinase Activity Assay Conditions: 1 µM STK S3 substrate was incubated with different concentrations of IKKββ protein in 10 µl reaction system containing 1×Enzymatic Buffer, 2 mM MgCl2, 1 mM DTT and 100 µM ATP for 1 hour. The 10 µl detection reagents containing anti-STK antibody and SA-XL665, each of which was 1:100 diluted with 1× Detection Buffer were added and incubated with the reactions for 1 hr. All the operations and reactions were performed at room temperature, and HTRF KinASE STK assay was used to detect the enzymatic activity.

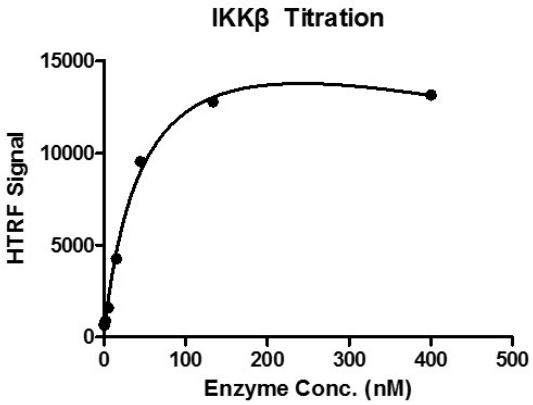
Storage and Guarantee: Recombinant proteins in solution are temperature sensitive and must be stored at -80°C to prevent degradation. Avoid repeated freeze/thaw cycles and keep on ice when not in storage. This product is for research use only and is not for use in diagnostic procedures. This product is guaranteed for 6 months from date of arrival.



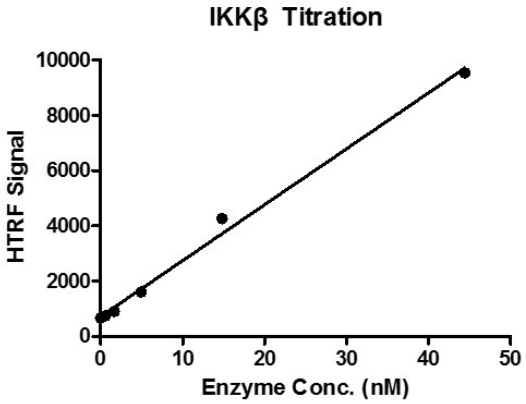
Recombinant IKK&beta protein gel>
7.5% SDS-PAGE Coomassie staining

MW: 87.8 kDa

Purity: ≥ 60%



HTRF assay for IKKβ activity
1 µM STK S3 substrate was incubated with different concentrations of IKKβ protein in 10 µl reaction system containing 1×Enzymatic Buffer, 2 mM MgCl2, 1 mM DTT and 100 µM ATP for 1 hour. The detection reagents were added and incubated with the reactions for 1 hr. All the operations and reactions were performed at room temperature, and HTRF KinASE STK assay was used to detect the enzymatic activity.



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