Certificate of Analysis
IKBKB (IKK beta), 10 µg

Recombinant Human Inhibitor of Kappa Light Chain Gene Enhancer in B Cells, Kinase Beta, GST-tagged

Part Number: PV3836
Lot Number: 1885150-2J
Immediate Storage: -80°C
Shipping Conditions: dry ice

5781 Van Allen Way
Carlsbad, CA 92008
Phone: 760.603.7200
www.thermofisher.com

Description:

Recombinant Human Full-Length protein, GST-tagged, expressed in insect cells. No special measures were taken to activate this kinase.

Specific Activity:

96 nmoles of phosphate transferred to IKKtide peptide substrate (KKKKERLLDDRHDSGLDSMKDEE) per minute per mg of total protein at 30°C. Activity determined at a final protein concentration of 8.33 µg/mL.

Concentration:

0.42 mg/mL total protein as measured using the Bradford protein assay with BSA as a standard.

Calculated **3,680 nM**.

Aliases:

IKK2, NFKBIKB, IKK Beta

Storage and Handling:

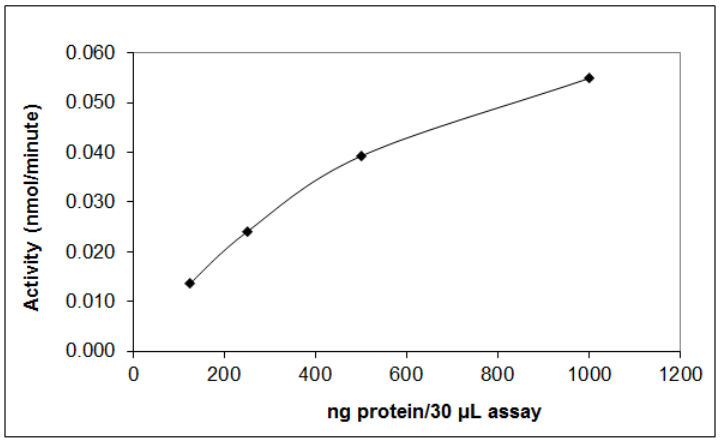
For maximum recovery please spin prior to use. Unless noted below, aliquots of the 5 ug, 10ug and 20ug sizes of kinase are not recommended as materials can be used in original packaging until exhausted. For larger sizes, the number of freeze/thaws may be reduced by preparing aliquots, aliquots below 20 µL are not recommended. **Please never store a kinase diluted.** If properly stored at −80°C, this product is guaranteed for 6 months from date of purchase.

Storage Buffer:

50 mM Tris (pH 7.5), 150 mM NaCl, 0.5 mM EDTA, 0.02% Triton® X–100, 2 mM DTT and 50% Glycerol.

QUALITY ASSURANCE

IKBKB (IKK beta) Activity Graph



Dilution Buffer:

20 mM Tris (pH 7.5), 0.02% Triton® X−100, 0.1 mg/mL BSA, 2 mM DTT, 0.5 mM Na_3VO_4 and 10% Glycerol.

Assay Conditions:

IKBKB (IKK beta) was pre-diluted in enzyme dilution buffer and assayed in 25 mM Tris (pH 7.5), 10 mM $MgCl_2$, 0.5 mM EGTA, 0.5 mM Na_3VO_4, 5 mM β−glycerophosphate, 2.5 mM DTT, 0.01% Triton® X-100, 200 µM ATP, 50 µg/mL IKKtide peptide substrate (KKKKERLLDDRHDSGLDSMKDEE) and trace [^{32}P]-γ-ATP for 10 minutes at 30°C.

Gel Information for IKBKB (IKK beta)

Page Description: The SDS-PAGE and/or Native PAGE were run on 4-20% Tris-Glycine Novex® gels (Catalog #: EC6025BOX).

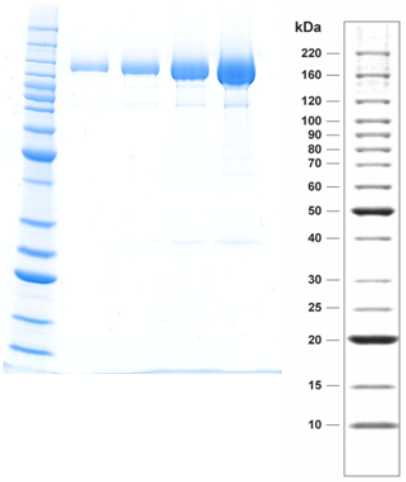
Lane 1: Invitrogen™ BenchMark™ Protein Ladder (Catalog #: 10747-012).

Lane 2: 0.5 µg IKBKB (IKK beta)

Lane 3: 1.0 µg IKBKB (IKK beta)

Lane 4: 2.5 µg IKBKB (IKK beta)

Lane 5: 5.0 µg IKBKB (IKK beta)



Purity:

90% as determined by a Coomassie® blue stained SDS-PAGE gel.

Molecular Weight:

114.0 kDa. Calculated from the protein sequence(s).

Mass Spectrometry:

IKBKB (IKK beta) was subjected to proteolytic digest followed by mass spec analysis. The resulting MS/MS data verified IKBKB (IKK beta) identity by comparison against the amino acid sequence(s) of the recombinant protein.

Protein sequence alignment with reference sequence(s)

GenBank Accession Number: NP_001547

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   1 MAPILGYWKI KGLVQPTRLL LEYLEEKYEE HLYERDEGDK WRNKKFELGL EFPNLPYYID GDVKLTQSMA IIRYIADKHN MLGGCPKERA EISMLEGAVL GST TAG
   1 MAPILGYWKI KGLVQPTRLL LEYLEEKYEE HLYERDEGDK WRNKKFELGL EFPNLPYYID GDVKLTQSMA IIRYIADKHN MLGGCPKERA EISMLEGAVL IVGN IKBKB
   1 M--------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- NP_001547

 101 DIRYGVSRIA YSKDFETLKV DFLSKLPEML KMFEDRLCHK TYLNGDHVTH PDFMLYDALD VVLYMDPMCL DAFPKLVCFK KRIEAIPQID KYLKSSKYIA
 101 DIRYGVSRIA YSKDFETLKV DFLSKLPEML KMFEDRLCHK TYLNGDHVTH PDFMLYDALD VVLYMDPMCL DAFPKLVCFK KRIEAIPQID KYLKSSKYIA
   2 ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

 201 WPLQGWQATF GGGDHPPKSD LVPR
 201 WPLQGWQATF GGGDHPPKSD LVPRHNQTSL YKKAGTMSWS PSLTTQTCGA WEMKERLGTG GFGNVIRWHN QETGEQIAIK QCRQELSPRN RERWCLEIQI
   2 ---------- ---------- ---------- -------SWS PSLTTQTCGA WEMKERLGTG GFGNVIRWHN QETGEQIAIK QCRQELSPRN RERWCLEIQI

 224
 301 MRRLTHPNVV AARDVPEGMQ NLAPNDLPLL AMEYCQGGDL RKYLNQFENC CGLREGAILT LLSDIASALR YLHENRIIHR DLKPENIVLQ QGEQRLIHKI
  65 MRRLTHPNVV AARDVPEGMQ NLAPNDLPLL AMEYCQGGDL RKYLNQFENC CGLREGAILT LLSDIASALR YLHENRIIHR DLKPENIVLQ QGEQRLIHKI

 224
 401 IDLGYAKELD QGSLCTSFVG TLQYLAPELL EQQKYTVTVD YWSFGTLAFE CITGFRPFLP NWQPVQWHSK VRQKSEVDIV VSEDLNGTVK FSSSLPYPNN
 165 IDLGYAKELD QGSLCTSFVG TLQYLAPELL EQQKYTVTVD YWSFGTLAFE CITGFRPFLP NWQPVQWHSK VRQKSEVDIV VSEDLNGTVK FSSSLPYPNN

 224
 501 LNSVLAERLE KWLQLMLMWH PRQRGTDPTY GPNGCFKALD DILNLKLVHI LNMVTGTIHT YPVTEDESLQ SLKARIQQDT GIPEEDQELL QEAGLALIPD
 265 LNSVLAERLE KWLQLMLMWH PRQRGTDPTY GPNGCFKALD DILNLKLVHI LNMVTGTIHT YPVTEDESLQ SLKARIQQDT GIPEEDQELL QEAGLALIPD

 224
 601 KPATQCISDG KLNEGHTLDM DLVFLFDNSK ITYETQISPR PQPESVSCIL QEPKRNLAFF QLRKVWGQVW HSIQTLKEDC NRLQQGQRAA MMNLLRNNSC
 365 KPATQCISDG KLNEGHTLDM DLVFLFDNSK ITYETQISPR PQPESVSCIL QEPKRNLAFF QLRKVWGQVW HSIQTLKEDC NRLQQGQRAA MMNLLRNNSC

 224
 701 LSKMKNSMAS MSQQLKAKLD FFKTSIQIDL EKYSEQTEFG ITSDKLLLAW REMEQAVELC GRENEVKLLV ERMMALQTDI VDLQRSPMGR KQGGTLDDLE
 465 LSKMKNSMAS MSQQLKAKLD FFKTSIQIDL EKYSEQTEFG ITSDKLLLAW REMEQAVELC GRENEVKLLV ERMMALQTDI VDLQRSPMGR KQGGTLDDLE

 224
 801 EQARELYRRL REKPRDQRTE GDSQEMVRLL LQAIQSFEKK VRVIYTQLSK TVVCKQKALE LLPKVEEVVS LMNEDEKTVV RLQEKRQKEL WNLLKIACSK
 565 EQARELYRRL REKPRDQRTE GDSQEMVRLL LQAIQSFEKK VRVIYTQLSK TVVCKQKALE LLPKVEEVVS LMNEDEKTVV RLQEKRQKEL WNLLKIACSK

 224
 901 VRGPVSGSPD SMNASRLSQP GQLMSQPSTA SNSLPEPAKK SEELVAEAHN LCTLLENAIQ DTVREQDQSF TALDWSWLQT EEEEHSCLEQ AS.
 665 VRGPVSGSPD SMNASRLSQP GQLMSQPSTA SNSLPEPAKK SEELVAEAHN LCTLLENAIQ DTVREQDQSF TALDWSWLQT EEEEHSCLEQ AS
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* highlighted residues denote differences from the reference protein sequence(s).

Anita Targosz, Associate Director Date: 22/Aug/2017