

## NIK

mitogen-activated protein kinase kinase kinase 14

Recombinant Human Active Protein Kinase

HGNC Symbol: MAP3K14

Synonyms: FTDCR1B, HS, HSNIK

Product No.: 0344-0000-1

Lot: 006

**Description:** Human NIK, full length, amino acids M<sub>1</sub>-P<sub>947</sub> (as in [NCBI/Protein](#) entry NP\_003945.2), N-terminal GST-HIS<sub>6</sub> fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

**Product identity:** NIK, Lot 006, was confirmed as NIK by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW**<sub>Fusion Protein</sub>: 133,437 Da

**Expression host:** Sf9 insect cells

**Purification:** GST-Affinity Chromatography

**Activation:** This kinase was not activated by special procedures

**Storage buffer:** 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 20 % glycerol

**Storage temperature:** -80°C

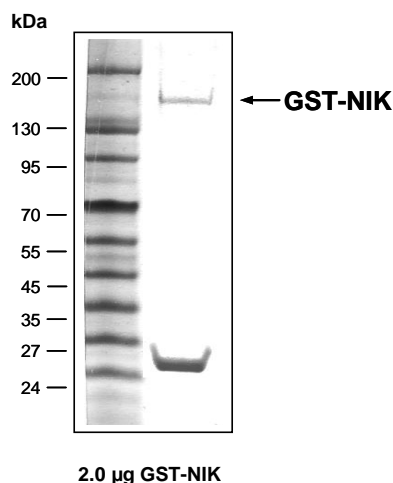
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

**Protein concentration:** 0.131 µg/µl  
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

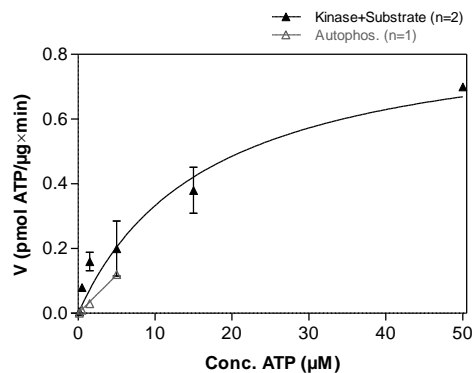
### Biochemical Parameters:

Specific kinase activity (P<sub>i</sub> transfer): 1 pmol/µg × min  
ATP-K<sub>M</sub>: 17 µM

### NIK Lot 006: Coomassie stain



### NIK Lot 006: Determination of V<sub>max</sub> and K<sub>M</sub> value for ATP



### Determination of K<sub>M</sub> value & Specific activity:

- Assay conditions:
  - 60 mM HEPES-NaOH, pH 7.5
  - 3 mM MgCl<sub>2</sub>
  - 3 mM MnCl<sub>2</sub>
  - 3 µM Na-orthovanadate
  - 1.2 mM DTT
  - 50 µg/ml PEG<sub>20,000</sub>
  - ATP (variable)
  - Substrate: CDC25C-derived peptide, 20 µg/ml
  - Kinase: 2 µg/ml
- Filter binding assay  
MSPH membrane (Millipore)

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GST-NIK Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSM	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GICSRMAVME	MACPGAPGSA	VGQKELPKA	KEKTPPLGKK	QSSVYKLEAV	300
301	EKSPVFCGKW	EILNDVITKG	TAKEGSEAGP	AAISIIAQAE	CENSQEFSP	FSEIFIFIAGS	360
361	KQYSQSESLD	QIPNNVAHAT	EGKMARVCWK	GKRRSKARKK	RKKKSSKSLA	HAGVALAKPL	420
421	PRTPEQESCT	IPVQEDDESPL	GAPYVRNTPQ	FTKPLKEPGL	GQLCFKQLGE	GLRPALPRSE	480
481	LHKLISPLQC	LNHVWKLHHP	QDGGPLPLPT	HPFPYSRLPH	PFPHPLQPW	KHPLESFLG	540
541	KLACVDSQKP	LPDPHLSKLA	CVDSPKPLPG	PHLEPSCLSR	GAHEKFSVEE	YLVHALQGSV	600
601	SSGQAHSLTS	LAKTWAARGS	RSREPSPKTE	DNEGVLLETK	LKPVVDEYRE	EVHWATHQLR	660
661	LGRGLFGEVH	RMEDKQTGFQ	CAVKKVRLEY	FRAEELMACA	GLTSPRIVPL	YGAVREGPWV	720
721	NIFMELLEGG	SLGQLVKEQG	CLPEDRALY	LGQALEGLEY	LHSRRILHGD	VKADNVLLSS	780
781	DGSHAALCDF	GHAVCLQPDG	LGKSLLTGDY	IPGTETHMAP	EVVLGRSCDA	KVDVWSSCCM	840
841	MLHMLNGCHP	WTQFFRGPLC	LKIASPPPPV	REIPPSCAPL	TAQAIQEGLR	KEPIHRVSAA	900
901	ELGGKVNRL	QQVGGLKSPW	RGEYKEPRHP	PPNQANYHQT	LHAQPRELSP	RAPGPRPAEE	960
961	TTGRAPKLQP	PLPPEPPEPN	KSPPLTLSKE	ESGMWEPLPL	SSLEPAPARN	PSSPERKATV	1020
1021	PEQELQQLEI	ELFLNSLSQP	FSLEEQEQIL	SCLSIDSLSL	SDDSEKNPSK	ASQSSRDTLS	1080
1081	SGVHSWSSQA	EARSSSWNMV	LARGRPTDTP	SYFNGVKVQI	QSLNGEHLHI	REFHRVKVGD	1140
1141	IATGISSQIP	AAAFSLVTKD	GQPVRYDMEV	PDSGIDLQCT	LAPDGSFAWS	WRVKHGQLEN	1200
1201	RP						1260

1-218: GST Red: HIS6-tag Pink: Thrombin cleavage site blue: NIK

NIK wt <sup>1</sup> Amino Acid Sequence							
1	MAVMEMACPG	APGSAVGQQK	ELPKAKEKTP	PLGKKQSSVY	KLEAVEKSPV	FCGKWEILND	60
61	VITKGTAKEG	SEAGPAAISI	IAQAECENSQ	EFSPTFSERI	FIAGSKQYSQ	SESLDQIPNN	120
121	VAHATEGKMA	RVCWKGKRRS	KARKKRRKKS	SKSLAHAGVA	LAKPLPRTPE	QESCTIPVQE	180
181	DESPLGAPYV	RNTPQFTKPL	KEPGLGQLCF	KQLGEGLRPA	LPRSELHKLI	SPLQCLNHVW	240
241	KLHHPQDGGP	LPLPTHFPFY	SRLPHFPFFH	PLQPWKPHPL	ESFLGKLACV	DSQKPLDPH	300
301	LSKLACVDS	KPLPGPHLEP	SCLSRGAHEK	FSVEEYLVHA	LQGSVSSGQA	HSLTSLAKTW	360
361	AARGSRREP	SPKTEDNEGV	LLTEKLPVD	YEYREEVHWA	THQLRLGRGS	FGEVHRMEDK	420
421	QTGFQCAVKK	VRLEVFRAEE	LMACAGLTSP	RIVPLYGAVR	EGPWVNI FME	LLEGGSLGQL	480
481	VKEQGCLPED	RALYYLGQAL	EGLEYLHRR	ILHGDVKADN	VLLSSDGSHA	ALCDFGHAVC	540
541	LQPDGLGKSL	LTGDYIPGTE	THMAPEVVLG	RSCDAKVDVW	SSCCMMLHML	NGCHPWTQFF	600
601	RGPLCLKIAS	EPPPVR EIPP	SCAPLTAQAI	QEGLRKEPIH	RVSAAELGGK	VNRLQOVGG	660
661	LKSPWRGEYK	EPRHPPPNQA	NYHQT LHAQP	RELSPRAPGP	RPAEETTGRA	PKLQPP LPE	720
721	PPEPNKSPPL	TLSKEESGMW	EPLPLSSLEP	APARNPSSPE	RKATVPEQEL	QQLEIELFLN	780
781	SLSQPFSL E	QEQLSCLSI	DSL SLD DSE	KNPSKASQSS	RDTLSSGVHS	WSSQAEARSS	840
841	SWNMVLARGR	PTDTPSYFNG	VKVQIQSLNG	EHLHIREFHR	VKVGDIATGI	SSQIPAAAFS	900
901	LVTKDGQPVR	YDMEVPDSGI	DLQCTLAPDG	SFAWSRVK H	GQLENRP		960

blue: NIK sequence expressed in recombinant protein

<sup>1</sup>NCBI/Protein accession number NP\_003945.2

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