

NEK6

NIMA (never in mitosis gene a)-related kinase 6

Recombinant Human Active Protein Kinase

HGNC Symbol: NEK6

Synonyms: SID6-1512

Product No.: 0209-0000-1

Lot: 001

Description: Human NEK6, amino acids M₈-T₃₁₃ (as in NCBI/Protein entry NP_055212.2), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: NEK6 Lot001, was confirmed as NEK6 by mass spectroscopy LC-ESI-MS/MS (Protagen AG, Germany)

Theoretical MW_{Fusion Protein}: 68,041 Da

Expression: Baculovirus infected Sf9 cells

Purification: GST-Affinity Chromatography

Activation: This kinase was not activated by special procedures

Storage buffer: 50 mM TRIS-HCl pH 8.0, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 20% glycerol

Storage temperature: -80°C

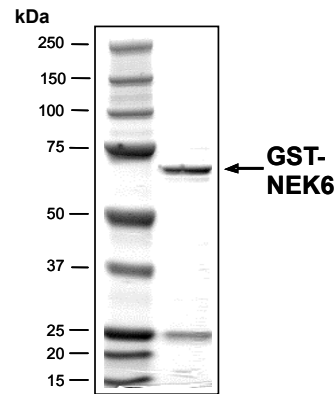
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

Protein concentration: 0.129 µg/µl (Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

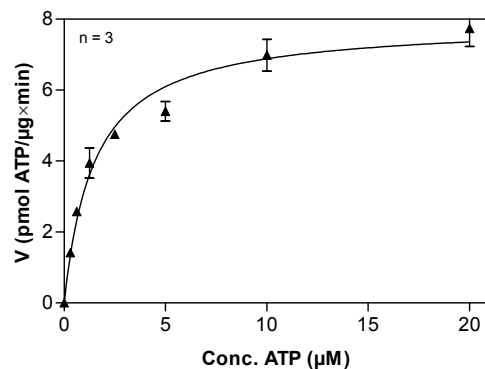
Specific kinase activity (P_i transfer): 7.9 pmol/µg×min
ATP-K_M: 1.5 µM

**NEK6 Lot 001:
Coomassie stain**



2.0 µg GST-NEK6

**NEK6 Lot 001:
Determination of V_{max} and K_M value for ATP**



Determination of K_M value & Specific activity:

- Assay conditions:
 - 60 mM HEPES-NaOH, pH 7.5
 - 3 mM MgCl₂
 - 3 mM MnCl₂
 - 3 µM Na-orthovanadate
 - 1.2 mM DTT
 - 50 µg / ml PEG_{20,000}
 - ATP (variable)
 - Substrate: RB-CTF, 40µg/ml
 - Kinase: 2.0 µg / ml
- Filter binding assay
 - MSFC membrane (Millipore)

Additional assay technology: NEK6 Lot 001

was also successfully tested by ProQinase for the use with the ADP-Glo™ Kinase assay from Promega. ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details



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NEK6 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQ SMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GIYARGIQAS	MGARGRQCDG	YLQNSPLMPH	GGSSNNLCHT	LGPVHPPDPQ	300
301	RHPNTLSFRC	SLADFQIEKK	IGRGQFSEVY	KATCLLDRKT	VALKKVQIFE	MMDAKARQDC	360
361	VKEIGLLKQL	NHPNIIKYLD	SFIEDNELNI	VLELADAGDL	SQMIKYFKKQ	KRLIPERTVW	420
421	KYFVQLCSAV	EHMHSRRVMH	RDIK PANVFI	TATGVVKLGD	LGLGRFFSSE	TTAAHSLVGT	480
481	PYYMSPERIH	ENGYNFKSDI	WSLGCLLYEM	AALQSPFYGD	KMNLFSLCQK	IEQCDYPLP	540
541	GEHYSEKLRE	LVSMCICPDP	HQRPDIGYVH	QVAKQMHIWM	SSTKGEFQHT	GGRY	600

1-218: GST Red: HIS6-tag Pink: Thrombin blue: NEK6 fragment

NEK6 wt ¹ amino acid sequence							
1	MAGQPGHMPH	GGSSNNLCHT	LGPVHPPDPQ	RHPNTLSFRC	SLADFQIEKK	IGRGQFSEVY	60
61	KATCLLDRKT	VALKKVQIFE	MMDAKARQDC	VKEIGLLKQL	NHPNIIKYLD	SFIEDNELNI	120
121	VLELADAGDL	SQMIKYFKKQ	KRLIPERTVW	KYFVQLCSAV	EHMHSRRVMH	RDIK PANVFI	180
181	TATGVVKLGD	LGLGRFFSSE	TTAAHSLVGT	PYYMSPERIH	ENGYNFKSDI	WSLGCLLYEM	240
241	AALQSPFYGD	KMNLFSLCQK	IEQCDYPLP	GEHYSEKLRE	LVSMCICPDP	HQRPDIGYVH	300
301	QVAKQMHIWM	SST					360

blue: NEK6 sequence expressed in fusionprotein

¹NCBI/Protein accession number NP_055212.2