

## MLK4

mitogen-activated protein kinase kinase kinase 21

Recombinant Human Active Protein Kinase

HGNC Symbol: MAP3K21

Synonyms: KIAA1804

Product No.: 1078-0000-1

Lot: 002

**Description:** Human MLK4, internal fragment, amino acids A<sub>98</sub>-L<sub>451</sub> (as in [NCBI/Protein](#) entry NP\_115811.2), N-terminal GST-HIS<sub>6</sub> fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

**Product identity:** MLK4 Lot 002, was confirmed as MLK4 by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW**<sub>Fusion Protein</sub>: 68,091 Da

**Expression host:** Sf9 insect cells/E.coli

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

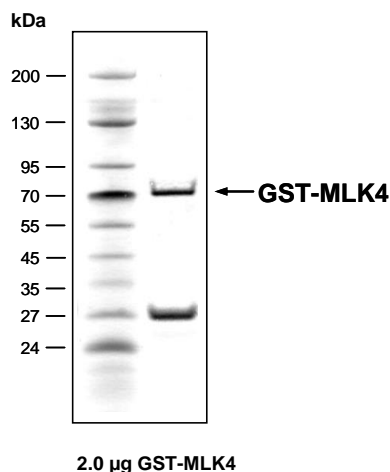
### Biochemical Parameters:

Specific kinase activity (P<sub>i</sub> transfer): 28 pmol/µg × min  
ATP-K<sub>M</sub>: 2.5 µM

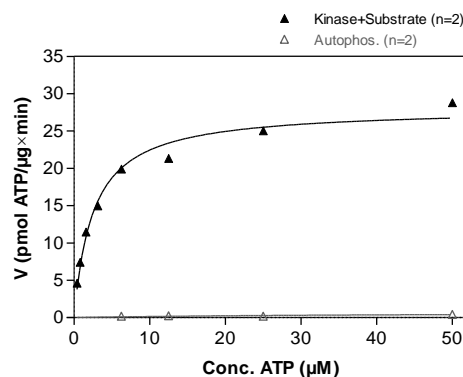
### Additional assay technology:

MLK4 Lot 002 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

### MLK4 Lot 002: Coomassie stain



### MLK4 Lot 002: 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: bio-lkBa, 8 µg/ml
  - MLK4: 1 µg/ml
- Filter binding assay  
MSPH membrane (Millipore)

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GST-MLK4 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDKVLTQSM	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI	PQID KYLKSSKYIA	WPLQG	WQATF GGGDHPPKSD	PMG	HHHHHG RDS	240
241	PLAMV	APCRP AASPAPPSR	PSSPVHVAFE	RLEL	KELIGA GGFGQVYRAT	WQGQEVAVKA	300
301	ARQDPEQDAA	AAAE	SVRREA RLFAMLRHPN	I IELRGVCLQ	QPHLC	LVLEF ARGGALNRAL	360
361	AAANAAPDPR	APG	PRRARRI PPHVLVNWAV	QIARG	MLYLH EEA	FVPILHR DLKSSNILL	420
421	EKIEHDDICN	KTLKITDFGL	AREW	HRTTKM STAGTYAWMA	PEVIK	SSLFS KGSDIWSYGV	480
481	LLWELLTGEV	PYRGIDGLAV	AYGVA	VNKL LPIPSTCPEP	FAKLM	KECWQ QDPHIRPSFA	540
541	LILEQLTAIE	GAVM	TEMPQE SFHSMQDDWK	LEIQ	QMFDEL RTKEKELRSR	EEELTRAA	600

1-218: GST Red: HIS6-tag Green: 3C cleavage site blue: MLK4 fragment

MLK4 wt <sup>1</sup> Amino Acid Sequence							
1	MALRGAAGAT	DTPVSSAGGA	PGGSASSSST	SSGGSASAGA	GLWAALYDYE	ARGEDELSLR	60
61	RGQLVEVLSQ	DAAVSGDEGW	WAGQVQRRLG	IFPANYV	APC RPAAS	PAPP SRPSSPVHVA	120
121	FERLEL	KELI GAGGFGQVYR	ATWQ	GQEVAV KAARQDPEQD	AAAA	AESVRR EARLFAMLRH	180
181	PNIIELRGVC	LQQPHLC	LVLEF EFARGGALNR	ALAAANAAPD	PRAPG	PRRAR RIPPHVLVNW	240
241	AVQIARGMLY	LHEEAFV	PIL HRDLKSSNIL	LLEKIEHDDI	CNKTL	KITDF GLAREWHR	300
301	KMSTAGTYAW	MAPEVIK	SSL FSKGSDIWSY	GVLLWELLTG	EV	PYRGIDGL AVAYGVA	360
361	LTLPIPSTC	P E	PF	AKLMKEC WQD	PHIRPS FALILEQLTA	IEGAVMTEMP	420
421	WKLEIQMFD	ELRTKEKELR	SREEELTRAA	LQQKSQEELL	KRREQ	QLAER EIDVLERELN	480
481	ILIFQLNQE	K PKVKKRKGKF	KRSRLKLDG	HRISLPSDFQ	HKITV	QASPN LDKRRSLNSS	540
541	SSSPSSPTM	MPRLRAIQLT	SDES	NKTWGR NTVFRQEEFE	DV	KRNFKKKG CTWGPNSIQM	600
600	KDRTDCKERI	RPLSDGNSPW	STILIK	NQKT MPLASLFVDQ	PGS	CEEPKLS PDGLEHRKPK	660
661	QIKLPSQAYI	DLPLGKDAQR	ENPAE	AESWE EAASANAATV	SIEM	TPTNSL SRSPQRKTE	720
721	SALYGCTVLL	ASVALGLDLR	ELHKAQA	AEE PLPKEEKKK	R EGIF	QRASKS RRSASPPTSL	780
781	PSTCGEASSP	PSLPLSSALG	ILSTPSFSTK	CLLQMDSEDP	LVDS	APVTC D SEMLTPDFCP	840
841	TAPGSGREPA	LMPRLDTDCS	VSRNLPSSFL	QQT	CGNVPYC ASSK	HRPSSH RRTMSDGNPT	900
901	PTGATIIISAT	GASALPLCPS	PAPSHL	PRE VSPKKHSTVH	IVP	QRRPASL RSRSDL	960
961	PQTAVS	QLAQ TACV	VGRPGP HPTQFLAAKE	RTKSHVPSLL	DAD	VEGQSRD YTVPLCRMRS	1020
1021	KTSRPSIYEL	EKEFLS					1080

blue: MLK4 sequence expressed in recombinant protein

<sup>1</sup>[NCBI/Protein](https://www.ncbi.nlm.nih.gov/protein/NP_115811.2) accession number NP\_115811.2