

## MAP3K1

mitogen-activated protein kinase kinase kinase 1

Recombinant Human Active Protein Kinase

HGNC Symbol: MAP3K1

Synonyms: MAPKKK1, MEKK, MEKK1

Product No.: 1103-0000-1

Lot: 001

**Description:** Human MAP3K1, C-terminal fragment, amino acids M<sub>1193</sub>-W<sub>1512</sub> (as in [NCBI/Protein](#) entry NP\_005912.1), N-terminal 4xFLAG, C-terminal HIS<sub>8</sub> fusion protein, expressed in Sf9 insect cells

**Product identity:** MAP3K1 Lot 001, was confirmed as MAP3K1 by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW**<sub>Fusion Protein</sub>: 41,387 Da

**Expression host:** Sf9 insect cells

**Purification:** Immobilized Metal 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, 20 % glycerol

**Storage temperature:** -80°C

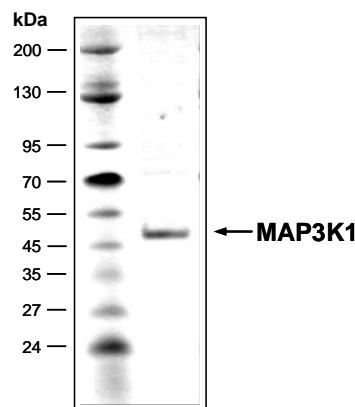
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

**Protein concentration:** 0.483 µg/µl  
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

### Biochemical Parameters:

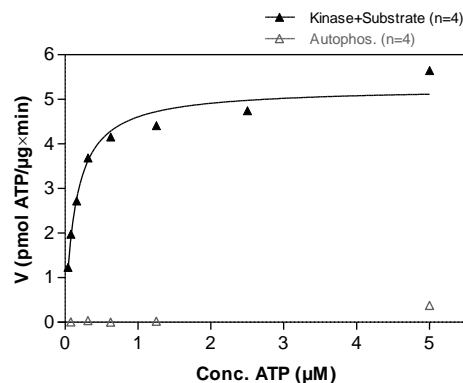
Specific kinase activity (P<sub>i</sub> transfer): 5.3 pmol/µg × min  
ATP-K<sub>M</sub>: 0.14 µM

### MAP3K1 Lot 001: Coomassie stain



2.0 µg MAP3K1

### MAP3K1 Lot 001: 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-RS-Peptide 100 µg/ml
  - MAP3K1: 1 µg/ml
- Filter binding assay  
MSPH membrane (Millipore)

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MAP3K1 Recombinant Fusion Protein Amino Acid Sequence							
1	M	DKDDDDDKD	YKDDDDDKDYK	DDDDDKDYKDD	DDKSGGGSMA	MSASQDALPI VPQLQVNGE	60
61	D	IIIIQQDTP	ETLPGHTKAK	QPYREDTEWL	KGQQIGLGAF	SSCYQAQDVG TGTLMVAKQV	120
121	T	YVRNTSSEQ	EEVVEALREE	IRMMSHLNHP	NIIRMLGATC	EKSNYNLFIE WMAGGSVAHL	180
181	L	SKYGAFKES	VVINYTEQLL	RGLSYLHENQ	IIHRDVKGAN	LLIDSTGQRL RIADFGAAAR	240
241	L	ASKGTGAGE	FQGQLLGTIA	FMAPEVLRGQ	QYGRSCDVWS	VGCAIEMAC AKPPWNAEKH	300
301	S	NHLALIFKI	ASATTAPSIP	SHLSPGLRDV	ALRCLELQPC	DRPPSRELLK HPVFRTTWKL	360
361	L	E	HHHHHHHH				420

1-218: GST Pink: 4x FLAG-tag Red: HIS6-tag blue: MAP3K1 fragment

MAP3K1 wt <sup>1</sup> Amino Acid Sequence							
1	M	AAAAGNRAS	SSGFPGARAT	SPEAGGGGGA	LKASSAPAAA	AGLLREAGSG GRERADWRRR	60
61	Q	LRKVRVEL	DQLPEQPLFL	AASPPASSTS	PSPEPADAAG	SGTGFQPVAV PPPHGAASRG	120
121	G	AHLTESVAA	PDSGASSPAA	AEPGEKRAPA	AEPSPAAAAPA	GREMENKETL KGLHKMDDRP	180
181	E	ERMIREKLEK	ATCMPAWKHE	WLERRNRRGP	VVVKPIPVKG	DGSEMNLHAA ESPGEVQASA	240
241	A	SPASKGRRS	PSPGNPSGR	TVKSESPGVR	RKRVSPPVFQ	SGRITPPRRA PSPDGFSPYS	300
301	P	EETNRRVVK	VMRARLYLQ	QIGPNSFLIG	GDSPDNKYRV	FIGPQNCSCA RGTFCIHLLF	360
361	V	MLRVFQLEP	SDPMLWRKTL	KNFEVESLFQ	KYHSRRSSRI	KAPSRNTIQK FVSRMSNSHT	420
421	L	SSSSTSTSS	SENSIKDEEE	QMCPICLLGM	LDEESLTVCE	DGCRNKLHHH CMSIWAECCR	480
481	R	NREPLICPL	CRSKWRSHDF	YSHELSSPVD	SPSSLRAAQ	QTVQQPLAG SRRNQESNFN	540
541	L	THYGTQQIP	PAYKDLAEPW	IQVFGMELVG	CLFSRNWVNR	EMALRRLSHD VSGALLLANG	600
600	E	STGNSGGSS	GSSPSGGATS	GSSQTSISGD	VVEACCSVLS	MVCADPVYKV YVAALKTLRA	660
661	M	LVYTPCHSL	AERIKLQRL	QPVVDITLVK	CADANSRTSQ	LSISTLLELC KGQAGELAVG	720
721	R	EILKAGSIG	IGGVYVLLNC	ILGNQTESNN	WQELLGRLCL	IDRLLLEFPA EFYPHIVSTD	780
781	V	SQAEPVEIR	YKLLLSLLTF	ALQSIDNSHS	MVGKLSRRIY	LSSARMVTTV PHVFSKLEEM	840
841	L	SVSSSTHFT	RMRRRLMAIA	DEVEIAEAIQ	LGVEDTLDGQ	QDSFLQASVP NNYLETTENS	900
901	S	PPECTVHLEK	TGKGLCATKL	SASSEDISER	LASISVGPSS	STTTTTTTE QPKPMVQTKG	960
961	R	PHSQCLNSS	PLSHHSQLMF	PALSTPSSST	PSVPAGTAD	VSKHRLQGFI PCRIPSASPQ	1020
1021	T	QRKFSLQFH	RNCPENKDS	KLSPVFTQSR	PLPSSNIHRP	KPSRPTPGNT SKQGDPSKNS	1080
1081	M	TLDLNSSSK	CDDSFCCSSN	SSNAVIPSDE	TVFTPVEEKC	RLDVNTEINS SIEDLLEASM	1140
1141	P	SSDTTVTFK	SEVAVLSPEK	AENDDTYKDD	VNHNQCKEK	MEAEELAA IAMAMSASQD	1200
1201	A	LPIVPQLQV	ENGEDI IIII	QDTPETLPGH	TKAKQPYRED	TEWLKGOQIG LGAFSSCYQA	1260
1261	Q	DVGTGTLMA	VKQVTVRNT	SSEQEEVVEA	LREEIRMMSH	LNHPNIRML GATCEKSNYN	1320
1321	L	FIEWMAGGS	VAHLLSKYGA	FKESVINYT	EQLLRGLSYL	HENQIIHRDV KGANLLIDST	1380
1381	G	QRLRIADFG	AAARLASKGT	GAGEFQQLL	GTIAFMAPEV	LRGQYGRSC DVWSVGCII	1440
1441	E	MACAKPPWN	AEKHSNHLAL	IFKIASATTA	PSIPSHLSPG	LRDVALRCLE LQPDRPPSR	1500
1501	E	LLKHPVFRT	TW				1560

blue: MAP3K1 sequence expressed in recombinant protein

<sup>1</sup>NCBI/Protein accession number NP\_005912.1

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