

ProQinase™ MAP4K4

Mitogen-activated protein kinase kinase kinase kinase 4

Recombinant Human Active Protein Kinase

HGNC Symbol: MAP4K4

Synonyms: HGK, MEKKK4, NIK

Product No.: 1493-0000-1

Lot: 008

Description: Human MAP4K4, amino acids A₇-W₁₃₂₀ (as in [NCBI/Protein](#) entry XP_005264112.1), N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: MAP4K4 Lot 008, was confirmed as MAP4K4 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 178,606 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: 0.133 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

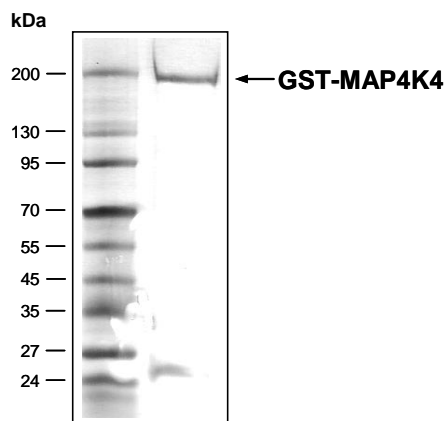
Biochemical Parameters:

Specific kinase activity (P_i transfer): 2.4 pmol/µg × min
ATP-K_M: 0.8 µM

Additional assay technology:

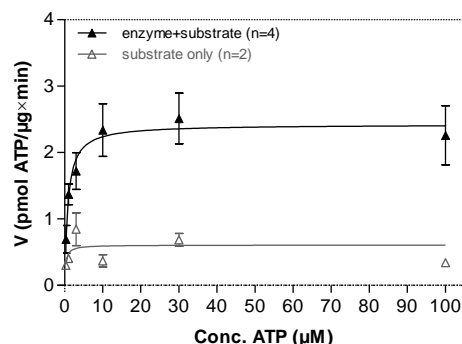
MAP4K4 Lot 008 was also successfully tested by Reaction Biology for the use with the ADP-Glo™ Kinase assay from Promega ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details

MAP4K4 Lot 008: Coomassie stain



2.0 µg GST-MAP4K4

MAP4K4 Lot 008: 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: RBER-GSK3, 80 µg/ml
 - MAP4K4: 0.4 µg/ml
- Filter binding assay
MSFC membrane (Millipore)

ProQinase™ MAP4K4

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GST-MAP4K4 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFKDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPOID	KYLKSSKYIA	WPLQGWAQTF	GGGDHPPKSD	PMGHHHHHG	RDSLEVLFCG	240
241	MAKSLVDID	LSSLRDPAGI	FELVEVVGNG	TYGQVYKGRH	VKTGQLAAIK	VMDVTEDEEE	300
301	EIKLEINMLK	KYSHHRNIAT	YGFYFIKSP	PGHDDQLWL	MEFCGAGSIT	DLVKNTKGNT	360
361	LKEDWIAYIS	REILRGLAHL	HIHHVIHRDI	KGQNVLLTEN	AEVKLVDFGV	SAQLDRTVGR	420
421	RNTFIGTPYW	MAPEVIACDE	NPDATYDYS	DLWSCGITAI	EMAEGAPPLC	DMHPRALFL	480
481	I PRNPPRLK	SKKWSKKFFS	FIEGCLVKNY	MQRPSTEQLL	KHPFIRDQPN	ERQVRIQLKD	540
541	HIDRTRKKRG	EKDETEYEYS	GSEEEEEVFP	EQEGEPSSIV	NVPGESTLRR	DFLRLQOENK	600
601	ERSEALRQQ	LLQEQQLREQ	EYKQQLLAE	RQKRIEQQKE	QRRRLEEQQR	REREARQQE	660
661	ERQRREQQE	KRLEELERR	RKEEEERRA	EEEKRRVERE	QEYIRRRQEE	EQRHLEVLQQ	720
721	QLLQEQAMLL	ECRWREMEEH	RQAERLQROL	QQEQAYLLSL	QHDHRRPHPO	HSQQPPPPQQ	780
781	ERSKPSFHAP	EPKAHYEPAD	RAREVEDRFR	KTNHSSPEAQ	SKQTGRVLEP	PVPSRSESEFS	840
841	NGNSESVHPA	LQRPAPQVQ	WSHLASLKN	VSPVSRSHSF	SDPSPKFAHH	HLRSQDPCPP	900
901	SRSEVLSQSS	DSKSEAPDPT	QKAWSRSDSD	EVPPRPVVRT	TSRSPVLSRR	DSPLQSGSQ	960
961	NSQAGQRNST	SSIEPRLLE	RVEKLVPRPG	SGSSSGSSNS	GSQPGSHPGS	QSGSGERFRV	1020
1021	RSSSKSEGST	SQLEENAVK	PEDKKEVFRP	LKPAGEVDLT	ALAKELRAVE	DVRPPHKVTD	1080
1081	YSSSSEESGP	TDEEDDDVEQ	EGADESTFRP	EDTRAASSLN	LSNGETESVK	TMIVHDDVES	1140
1141	EPAMTPSKEG	TLIVRQTQSA	SSTLQKHKSS	SSFTPFIDPR	LLQISPSSTG	TVTSVVGFS	1200
1201	DGMRPEAIRQ	DPTRKGSVVN	VNPTNTRPQS	DTPEIRKYKK	RFNSEILCAA	LWGVNLLVGT	1260
1261	ESGLMLLDRS	GQGVYPLIN	RRRFQQMDVL	EGLNVLVITIS	GKKDKLRVY	LSWLRNKILH	1320
1321	NDPEVEKKQG	WTTVGDLEGC	VHYKVVKYER	IKFLVIALKS	SVEVYAWAPK	PYHKFMAFKS	1380
1381	FGELVHKPLL	VDLTVEEGQR	LKVIYGSCAG	FHAVDSDSGS	VYDIYLPTHI	QCSIKPHAI	1440
1441	ILPNTDGMEL	LVCYEDEGVY	VNTYGRITKD	VFLQWGEMPT	SVAYIRSNQT	MGWGEKAI	1500
1501	RSVETGHLDG	FVFMHKRAQL	KFLCERNRDKV	FASVRSVSGS	SQVYFMTLGR	TSLLSW	1560

1-218: GST Red: HIS6-tag Green: 3C cleavage site blue: MAP4K4

MAP4K4 wt ¹ Amino Acid Sequence							
1	MANDSPAKSL	VDIDLSSLRD	PAGIFELVEV	VGNQTYGQVY	KGRHVKTGQL	AAIKVMDVTE	60
61	DEEEEIKLEI	NMLKKYSHHR	NIATYYGAFI	KKSPPGHDDQ	LWLVMEFCGA	GSITDLVKNT	120
121	KGNTLKEDWI	AYISREILRG	LAHLHIHVI	HRDIKQNVL	LTENAIEVKLV	DFGVSAQLDR	180
181	TVGRRNTFIG	TPYWMAPEVI	ACDENPDATY	DYRSDLWSCG	ITAIEMAEAG	PPLCDMHPMR	240
241	ALFLIPRNPP	PRLKSKKWSK	KFFSFIEGCL	VKNYMQRPST	EQLLKHPFIR	DQPNERQVRI	300
301	QLKDHIRTR	KKRGEKDETE	YEYSGSEEEE	EEVPEQEGEP	SSIVNVPGES	TLRRDFLRLQ	360
361	QENKERSEAL	RRQQLLQEQ	LREQEYKQ	LLAERQKRIE	QQKEQRRRLE	EQRRREREAR	420
421	RQQEREQRR	EQEEKRLEE	LERRRKEEEE	RRRAEEKRR	VEREQEYIRR	QLEEEQRHLE	480
481	VLOQQLLQEQ	AMLLECRWRE	MEEHRQAERL	QRQLQEQAY	LLSLQHDHRR	PHPQHSQPP	540
541	PPQERSKPS	FHAPEPKAHY	EPADRAREVE	DRFRKTNHSS	PEAQSKQTR	VLEPPVPSRS	600
601	ESFSNGNSE	VHPALQPAE	PQVQWSHLAS	LKNVSPVSR	SHSFDPSPK	FAHHHLRSQD	660
661	PCPPSRSEVL	SQSSDSKSEA	PDPTQKAWSR	SDSDEVPPRV	PVRTTSRSPV	LSRRDSPLQG	720
721	SGQONSQAGQ	RNSTSSIEPR	LLWERVEKLV	PRPGSGSSG	SSNSGSPQGS	HPGSQSGSGE	780
781	RFRVRSSSKS	EGSPSQRLN	AVKKPEDKKE	VFRPLKPAGE	VDLTALAKEL	RAVEDVRPPH	840
841	KVTDYSSSSE	ESGTTDEEDD	DVEQEGADES	TSGPEDTRAA	SSLNLSNGET	ESVKTMIVHD	900
901	DVESEAMPMT	SKEGTLIVRQ	TQSASSTLQK	HKSSSFTPE	IDPRLQLISP	SSGTTVTSVV	960
961	GFSCDGMRPE	AIRQDPTRKG	SVVNVNPTNT	RPQSDTPEIR	KYKKRFNSEI	LCAALWGVNL	1020
1021	LVGTESGLML	LDRSGQGVY	PLINRRRFQ	MDVLEGLNVL	VTISGKKDKL	RVYYLSWLRN	1080
1081	KILHNDPEVE	KKQGWTTVGD	LEGCVHYKV	KYERIKFLVI	ALKSSVEVYA	WAPKPYHKFM	1140
1141	AFKSFQELVH	KPLLVDLTV	EGQLKVIY	SCAGFHAVDV	DSGSVYDIYL	PTHIQCSIKP	1200
1201	HAIILPNTD	GMELLVCYED	EGVYVNTYGR	ITKDVVLQWG	EMPTSVAYIR	SNQTMGWGK	1260
1261	AIEIRSVETG	HLDGVMFHKR	AQRLKFLCER	NDKVFFASVR	SGSSQVYFM	TLGRTSLLSW	1320

blue: MAP4K4 sequence expressed in recombinant protein

¹NCBI/Protein accession number XP_005264112.1