

ProQinase™ MEKK2

mitogen-activated protein kinase kinase kinase 2

Recombinant Human Active Protein Kinase

HGNC Symbol: MAP3K2

Synonyms: MAPKKK2, MEKK2B

Product No.: 0583-0000-1

Lot: 005

Description: Human MEKK2, full length, amino acids M₁-H₆₁₉ (as in [NCBI/Protein](#) entry NP_006600.3), N-terminal GST-HIS₆ fusion protein with a Thrombin and 3C cleavage site, expressed in Sf9 insect cells

Product identity: MEKK2 Lot 005, was confirmed as MEKK2 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 100,450 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: 0.158 µg/µl

(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

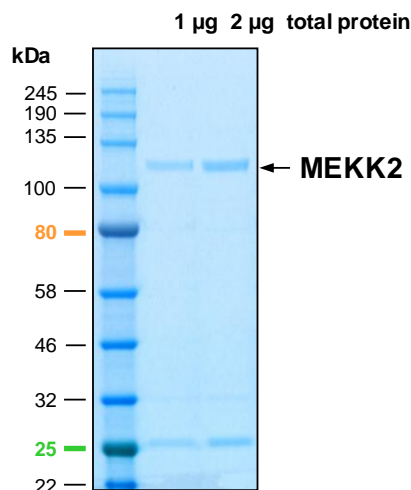
Specific kinase activity (P_i transfer): 1.6 pmol/µg × min

ATP-K_M: 0.7 µM

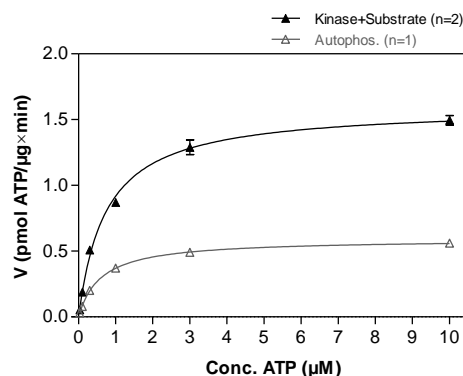
Additional assay technology:

MEKK2 Lot 005 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

MEKK2 Lot 005: Coomassie stain



MEKK2 Lot 005: 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: Casein 20 µg/ml
 - Kinase: 4 µg/ml
- Filter binding assay
- MSFC membrane (Millipore)

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GST-MEKK2 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPOID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GIYARDSLEV	LFQGPLAMDD	QQALNSIMQD	LAVLHKASRP	ALSLQETRKA	300
301	KSSSPKKQND	VRVKFEHRGE	KRILQFPRPV	KLEDLRKAK	IAFGQSMDLH	YTNNELVIPL	360
361	TTQDDLDKAV	ELLDRSIHMK	SLKILLVING	STQATNLEPL	PSLEDLDNTV	FGAERKKRLS	420
421	IIGPSTRDRS	SPPPGYIPDE	LHQVARNGSF	TSINSEGEFI	PESMDQMLDP	LSLSSPENSG	480
481	SGSCPSLDSP	LDGESYPKSR	MPRAQSYPDN	HQEFSDYDNP	IFEKFGKGGT	YPRRYHVSYP	540
541	HQEYNDGRKT	FPRARRTQGT	SLRSPVSFSP	TDHSLSTSSG	SSIFTPEYDD	SRIRRRGSDI	600
601	DNPTLTVMID	SPPSRSPRAP	TNWRLGKLLG	QGAFGRVYLC	YDVDTGRELA	VKQVQFDPDS	660
661	PETSKEVNAL	ECEIQLLKNL	LHERIVQYVG	CLRDPQEKTL	SIFMEYMPGG	SIKDQLKAYG	720
721	ALTENVTRKY	TRQILEGVHY	LHSNMIVHRD	IKGANILRDS	TGNVKLGDFG	ASKRLQITICL	780
781	SGTGMKSVTG	TPYWMSPEVI	SGEGYGRKAD	IWSVACTVVE	MLTEKPPWAE	FEAMAAIFKI	840
841	ATQPTNPKLP	PHVSDYTRDF	LKRIFVEAKL	RPSADELLRH	MFVHYH		900

1-218: GST Red: HIS6-tag Pink: Thrombin cleavage site Green: 3C cleavage site blue: MEKK2

MEKK2 wt ¹ Amino Acid Sequence							
1	MDDQQALNSI	MQDLAVLHKA	SRPALSLQET	RKAKSSSPKK	QNDVRVKFEH	RGEKRILQFP	60
61	RPVKLEDLRS	KAKIAFGQSM	DLHYTNNELV	IPLTTQDDLD	KAVELLDLRSI	HMKSLKILLV	120
121	INGSTQATNL	EPLPSLEDLD	NTVFGAERKK	RLSIIGPSTR	DRSSPPPGYI	PDELHQVARN	180
181	GSFTSINSEG	EFIPESMDQM	LDPLSLSSPE	NSGSGSCPSL	DSPLDGESYP	KSRMPRAQSY	240
241	PDNHQEFSDY	DNPIFEKFGK	GGTYPRRYHV	SYHHQEYNDG	RKTFPRARRT	QGTSLRSPVS	300
301	FSPTDHSLSL	SSGSSIFTPE	YDDSRIRRRG	SDIDNPTLTV	MDISPPSRSP	RAPTNRWLK	360
361	LLGQGAFRV	YLCYDVDTGR	ELAVKQVQFD	PDSPETSKEV	NALECEIQLL	KNLLHERIVQ	420
421	YYGCLRDPQE	KTLSIFMEYM	PGSISKDLK	AYGALTENVV	RKYTRQILEG	VHYLHSNMIV	480
481	HRDIKGANIL	RDSTGNVCLG	DFGASKRLQT	ICLSGTGMKS	VTGTPYWMSP	EVISGEGYGR	540
541	KADIWSVACT	VVEMLTEKPP	WAEFEAMAAI	FKIATQPTNP	KLPPHVSDYT	RDFLKRIFVE	600
601	AKLRPSADEL	LRHMFVHYH					660

blue: MEKK2 sequence expressed in recombinant protein

¹NCBI/Protein accession number NP_006600.3