

ProQinase™ ASK1

mitogen-activated protein kinase kinase kinase 5

Recombinant Human Active Protein Kinase

HGNC Symbol: MAP3K5

Synonyms: MAPKKK5

Product No.: 1998-0000-1

Lot: 003

Description: Human ASK1, internal fragment, amino acids M₁-V₉₇₁ (as in [NCBI/Protein](#) entry NP_005914.1), N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: ASK1 Lot 003, has been verified by mass spectrometry LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 64170 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

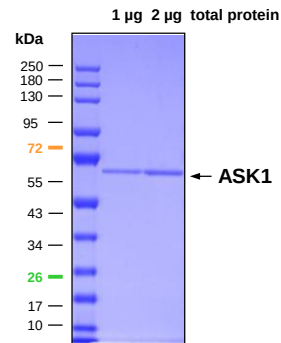
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10 µl are not advisable. Avoid repeated freeze-thaw cycles!

Protein concentration: 0.622 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

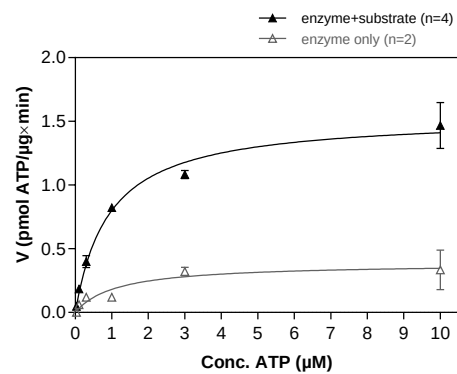
Biochemical Parameters:

Specific kinase activity (P_i transfer): 1.5 pmol/µg*min
ATP-K_M: 0.9 µM

ASK1 Lot 003: Coomassie stain



ASK1 Lot 003: Determination of V_{max} and K_M value for ATP



- 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-S6P 40 µg/ml
 - Kinase: 2 µg/ml

Assay technology:
Radiometric filter binding assay
MSFC membrane (96 well plate, Millipore)

Sequence information

GST-ASK1 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLIERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPEML	KMFKDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHGG	RDSLEVLFGG	240
241	PMVNTITEEK	GRSTEEGDCE	SDLLEYDYEY	DENGDRVVLG	KGTYGIVYAG	RDLSNQVRIA	300
301	IKEIPERDSR	YSQPLHEEIA	LHKHLKHKNI	VQYLGFSFSEN	GFIKIFMEQV	PGGSLSALLR	360
361	SKWGPLKDNE	QTIGFYTKQI	LEGLKYLHDN	QIVHRDIKGD	NVLINTYSGV	LKISDFGTSK	420
421	RLAGINPCTE	TFTGTLQYMA	PEIIDKGPRG	YGKAADIWSL	GCTIEMATG	KPPFYELGEP	480
481	QAAMFKVGMF	KVHPEIPESM	SAEAKAFILK	CFEPDPDKRA	CANDLLVDEF	LKVSSKKKKT	540
541	QPKLSALSAG	SNEYLRSISL	PVPV				600

1-218: GST **Red**: HIS6-tag **Green**: 3C cleavage site **blue**: ASK1 fragment

ASK wt ¹ Amino Acid Sequence							
1	MSTEADegIT	FSVPPFAPSG	FCTIPEGGIC	RRGGAAAVGE	GEEHQLPPPP	PGSFWNVESA	60
61	AAPGIGCPAA	TSSSSATRGR	GSSVGGGSRR	TTVAYVINEA	SQGQLVVAES	EALQSLREAC	120
121	ETVGATLETI	HFGKLDfGET	TVLDRFYNAD	IAVVEMSDAF	RQPSLFYHLG	VRESFSMANN	180
181	IILYCDTNSD	SLQSLKEIIC	QKNTMCTGNY	TFVPYMITPH	NKVYCCDSSF	MKGLTELMQP	240
241	NFELLGLPIC	LPLVDRFIQL	LKVAQASSSQ	YFRESILNDI	RKARNLYTGK	ELAAELARIR	300
301	QRVDNIEVLT	ADIVINLLLS	YRDIQDYDSI	VKLVTLEKL	PTFDLASHHH	VKFHYAFALN	360
361	RRNLPGDRAK	ALDIMIPMVQ	SEGQVASDMY	CLVGRIYKDM	FLDSNFTDTE	SRDHGASWFK	420
421	KAFESEPTLQ	SGINYAVLLL	AAGHQFESSF	ELRKVGKLS	SLLGKKGNLE	KLQSYWEVGF	480
481	FLGASVLAND	HMRVIQASEK	LFKLKTPAWY	LKSIVETILI	YKHfVKLTTE	QPVAKQELVD	540
541	FWMDFLVEAT	KTDVTVVRFP	VLILEPTKIY	QPSYLSINNE	VEEKTISIWH	VLPDDKKGIH	600
601	EWNFSASSVR	GVSISKFEER	CCFLYVLHNS	DDFQIYFCTE	LHCKKFFEMV	NTITEEKGRS	660
661	TEEGDCESDL	LEYDYEYDEN	GDRVVLGKGT	YGIVYAGRDL	SNQVRIAIKE	IPERDSRYSQ	720
721	PLHEEIALHK	HLKHKNIQYQ	LGSFSENGFI	KIFMEQVPGG	SLSALLRSKW	GPLKDNEQTI	780
781	GFYTKQILEG	LKYLHDNQIV	HRDIKGDNVL	INTYSGVLKI	SDFGTSKRLA	GINPCTETFT	840
841	GTLQYMAPEI	IDKGPRGYGK	AADIWSLGCT	IIEMATGKPP	FYELGEPQAA	MFKVGMFKVH	900
901	PEIPESMSAE	AKAFILKCFE	PDPDKRACAN	DLLVDEFLKV	SSKKKKTQPK	LSALSAGSNE	960
961	YLRISLSPVP	VLVEDTSSSS	EYGSVSPDTE	LKVDPFsFKT	RAKSCGERDV	KGIRTLFLGI	1020
1021	PDENFEDHSA	PPSPEEKDSG	FFMLRKDSER	RATLHRILTE	DQDKIVRNLM	ESLAQGAEEP	1080
1081	KLKWEHITTL	IASLREFVRS	TDRKIIATTL	SKLKLDFD	SHGISQVQV	LFGFQDAVNK	1140
1141	VLRNHNKPH	WMFALDSIIR	KAVQTAITIL	VPFLRPfHSL	ASESDTADQE	DLDVEDDHEE	1200
1201	QPSNQTVRRP	QAVIEDAVAT	SGVSTLSSTV	SHDSQSAHRS	LNVLQGRMKI	ETNRLLEELV	1260
1261	RKEKELQALL	HRAIEEKDQE	IKHLKLKSQP	IEIPELPVFH	LNSSGTNTED	SELTDLWRVN	1320
1321	GAEDETISRF	LAEDYTLLDV	LYYVTRDDLK	CLRLRGMLC	TLWKAIIDFR	NKQT	1380

blue: ASK1 sequence expressed in recombinant protein¹[NCBI/Protein](https://www.ncbi.nlm.nih.gov/Protein) accession number NP_005914.1