

Certificate of Analysis

MST4

MASK like serine/threonine kinase

Recombinant Human Active Protein Kinase

HGNC Symbol: MST4

Synonyms: MASK

Product No.: 0286-0000-1

Lot: 001

Description: Human MST4, full length, amino acids M₁-P₄₁₆ (as in NCBI/Protein entry NP_057626.1), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: MST4 Lot 001, was confirmed as MST4 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 76,424 Da

Expression: Baculovirus infected Sf9 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: 0.209 µg/µl

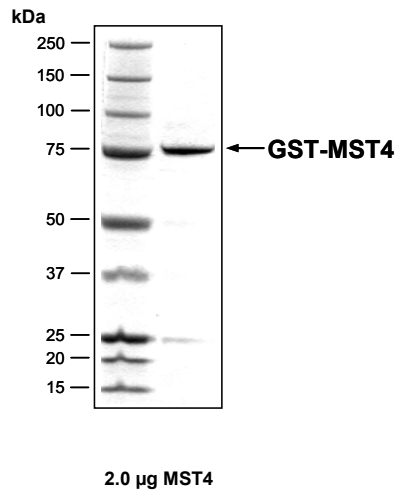
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

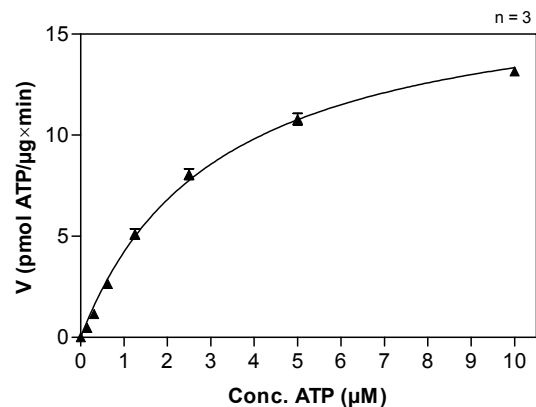
Specific kinase activity (P_i transfer): 18 pmol/µg×min

ATP-K_M: 3.1 µM

MST4 Lot 001: Coomassie stain



MST4 Lot 001: 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: Myelin Basic Protein, 2.0 µg/ml
 - MST4: 2.0 µg/ml
- Filter binding assay
 - MAFC membrane (Millipore)

Additional assay technology: MST4 Lot 001

was also successfully tested by ProQinase for the use with the ADP-Glo™ Kinase assay from



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GST-MST4 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHGG	RRRASVAAGI	240
241	LVPRGSPGLD	GIYARGIQAS	MAHSPVAVQV	PGMQNNIADP	EELFTKLERI	GKGSFGEVFK	300
301	GIDNRTQQVV	AIKIIDLEEA	EDEIEDIQQE	ITVLSQCDSS	YVTKYYGSYL	KGSKLWIIME	360
361	YLGGSALDL	LRAGPFDEFQ	IATMLKEILK	GLDYLHSEKK	IHRDIKAANV	LLSEQGDVKL	420
421	ADFGVAGQLT	DTQIKRNTFV	GTPFWMAPEV	IQQSAYDSKA	DIWSLGITAI	ELAKGEPPNS	480
481	DMHMPRVLFL	IPKNNPPTLV	GDFTKSFKEF	IDACLNKDPS	FRPTAKELLK	HKFIVKNSKK	540
541	TSYLTELIDR	FKRWKAEGHS	DDESSEGS	SESTSRENNT	HPEWSFTTVR	KKPDPKKVQN	600
601	GAEQDLVQTL	SCLSMIITPA	FAELKQDEN	NASRNQAIEE	LEKSIABAEA	ACPGITDKMV	660
	KKLIEKFQKC	SADESP					

1-218: GST **Red:** HIS6-tag **Pink:** Thrombin cleavage site **blue:** MST4

MST4 wt ¹ Amino Acid Sequence							
1	MAHSPVAVQV	PGMQNNIADP	EELFTKLERI	GKGSFGEVFK	GIDNRTQQVV	AIKIIDLEEA	60
61	EDEIEDIQQE	ITVLSQCDSS	YVTKYYGSYL	KGSKLWIIME	YLGGSALDL	LRAGPFDEFQ	120
121	IATMLKEILK	GLDYLHSEKK	IHRDIKAANV	LLSEQGDVKL	ADFGVAGQLT	DTQIKRNTFV	180
181	GTPFWMAPEV	IQQSAYDSKA	DIWSLGITAI	ELAKGEPPNS	DMHMPRVLFL	IPKNNPPTLV	240
241	GDFTKSFKEF	IDACLNKDPS	FRPTAKELLK	HKFIVKNSKK	TSYLTELIDR	FKRWKAEGHS	300
301	DDESSEGS	SESTSRENNT	HPEWSFTTVR	KKPDPKKVQN	GAEQDLVQTL	SCLSMIITPA	360
361	FAELKQDEN	NASRNQAIEE	LEKSIABAEA	ACPGITDKMV	KKLIEKFQKC	SADESP	420

blue: MST4 sequence expressed in fusionprotein

¹NCBI/Protein accession number NP_057626.1