Certificate of Analysis

REACTION BIOLOGY

Recombinant Proteins

ProQinase™ 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 M_1-P_{416} (as in NCBI/Protein entry NP_057626.1), N-terminal GST-HIS_6 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

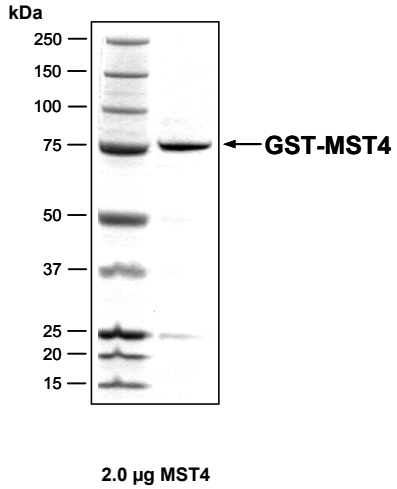
Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 20% glycerol

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)

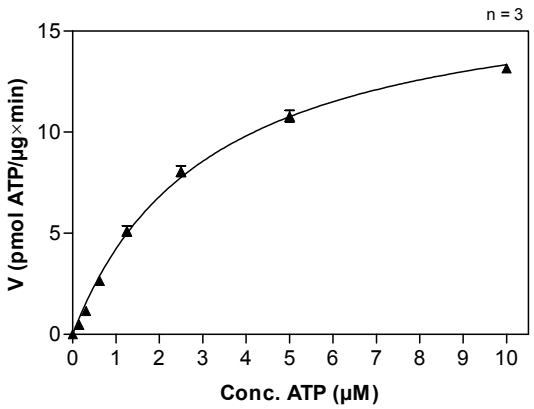
Biochemical Parameters:
Specific kinase activity (P_i transfer): 18 pmol/µg×min
ATP-K_M: 3.1 µM

MST4 Lot 001:
Coomassie stain



2.0 µg MST4

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 $MgCl_2$
  3 mM $MnCl_2$
  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 Reaction Biology for the use with the ADP-Glo™ Kinase assay from ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details

Promega

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ProQinase™ MST4

Product No.: 0286-0000-1

GST-MST4 Recombinant Fusion Protein Amino Acid Sequence

```
1    MSPILGYWKI KGLVQPTRLL LEYLEEKYEE HLYERDEGDK WRNKKFELGL EFPNLPYYID  60
61   GDVKLTQSMA IIRYIADKHN MLGGCPKERA EISMLEGAVL DIRYGVSRIA YSKDFETLKV  120
121  DFLSKLPEML KMFEDRLCHK TYLNGDHVTH PDFMLYDALD VVLYMDPMCL DAFPKLVCFK  180
181  KRIEAIPQID KYLKSSKYIA WPLQGWQATF GGGDHPPKSD PMGHHHHHHG RRRASVAAGI  240
241  LVPRGSPGLD GIYARGIQAS MAHSPVAVQV PGMQNNIADP EELFTKLERI GKGSFGEVFK  300
301  GIDNRTQQVV AIKIIDLEEA EDEIEDIQQE ITVLSQCDSS YVTKYYGSYL KGSKLWIIME  360
361  YLGGGSALDL LRAGPFDEFQ IATMLKEILK GLDYLHSEKK IHRDIKAANV LLSEQGDVKL  420
421  ADFGVAGQLT DTQIKRNTFV GTPFWMAPEV IQQSAYDSKA DIWSLGITAI ELAKGEPPNS  480
481  DMHPMRVLFL IPKNNPPTLV GDFTKSFKEF IDACLNKDPS FRPTAKELLK HKFIVKNSKK  540
541  TSYLTELIDR FKRWKAEGHS DDESDSEGSD SESTSRENNT HPEWSFTTVR KKPDPKKVQN  600
601  GAEQDLVQTL SCLSMIITPA FAELKQQDEN NASRNQAIEE LEKSIAVAEA 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 YLGGGSALDL LRAGPFDEFQ  120
121  IATMLKEILK GLDYLHSEKK IHRDIKAANV LLSEQGDVKL ADFGVAGQLT DTQIKRNTFV  180
181  GTPFWMAPEV IQQSAYDSKA DIWSLGITAI ELAKGEPPNS DMHPMRVLFL IPKNNPPTLV  240
241  GDFTKSFKEF IDACLNKDPS FRPTAKELLK HKFIVKNSKK TSYLTELIDR FKRWKAEGHS  300
301  DDESDSEGSD SESTSRENNT HPEWSFTTVR KKPDPKKVQN GAEQDLVQTL SCLSMIITPA  360
361  FAELKQQDEN NASRNQAIEE LEKSIAVAEA ACPGITDKMV KKLIEKFQKC SADESP      420
```

blue: MST4 sequence expressed in fusionprotein

[1]NCBI/Protein accession number NP_057626.1

Recombinant Proteins

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