

ProQinase™ MET F1200I

met proto-oncogene

Recombinant Human Active Protein Kinase

HGNC Symbol: MET

Synonyms: c-MET, HGFR

Product No.: 0978-0000-1

Lot: 001

Description: Human MET C-terminal fragment, amino acids K₉₅₆-S₁₃₉₀ (as in NCBI/Protein entry NP_000236.2), F1200I mutant, N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: MET F1200I Lot 001 product identity was confirmed by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 78,753 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 10 µl are not advisable. Avoid repeated freeze-thaw cycles!

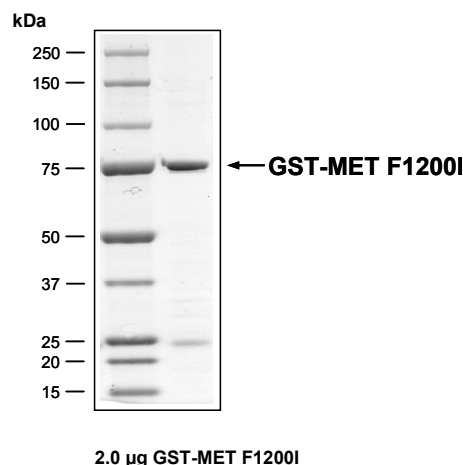
Protein concentration: 0.307 µg/µl

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

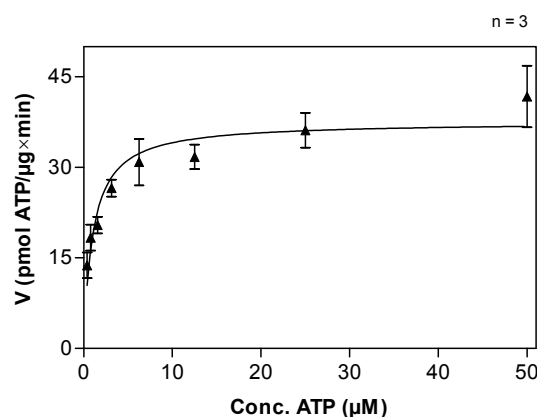
Biochemical Parameters:

Specific kinase activity (P_i transfer): 38 pmol/µg × min
ATP-K_M: 1.0 µM

MET F1200I Lot 001: Coomassie stain



MET F1200I 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: TRK-C derived peptide 20 µg/ml
 - Kinase: 1.0 µg/ml
- Filter binding assay
 - MSPH membrane (Millipore)

Additional assay technology: MET F1200I 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



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MET F1200I 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	KRIEAIPIQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GICSIEEFKK	RKQIKDLGSE	LVRYPARVHT	PHLDRLVSAR	SVSPTTEMVS	300
301	NESVDYRATF	PEDQFPNSSQ	NGSCRQVQYP	LTDMSPIILTS	GSDSISSPLL	QNTVHIDLSA	360
361	LNPELVQAVQ	HVVIGPSSLI	VHFNEVIGRG	HFGCVYHGTL	LDNDGKKIHC	AVKSLNRITD	420
421	IGEVSQLFTE	GIIMKDFSHP	NVLSLLGICL	RSEGSPLVVL	PYMKHGDRLN	FIRNETHNPT	480
481	VKDILGFGLQ	VAKGMKYLAS	KKIVHRDLAA	RNCMLDEKFT	VKVADFGLAR	DMYDKEYYSV	540
541	HNKTKGAKLPV	KWMALESLOT	QKFTTKSDVW	SFGVLLWELM	TRGAPPYPDV	NTFDITVYLL	600
600	QGRRLQLPEY	CPDPLYEVML	KCWHPKAEMR	PSFSELVSRI	SAIFSTFIGE	HYVHVNTATV	660
661	NVKCVAPYPS	LLSSEDNADD	EVDTRPASFW	ETS			720

1-218: GST Red: HIS6-tag Pink: Thrombin cleavage site blue: MET fragment boxed: F1200I

MET wt ¹ Amino Acid Sequence							
1	MKAPAVLAPG	ILVLLFTLVQ	RSNGECKEAL	AKSEMNVNMK	YQLPNFTAET	PIQNVILHEH	60
61	HIFLGATNYI	YVLNEEDLQK	VAEYKTGPVL	EHDPDFPCQD	CSSKANLSGG	VWKDNINMAL	120
121	VVDYYDDQL	ISCGSVNRGT	CQRHVFPNNH	TADIQSEVHC	IFSPQIEEPS	QCPDCVVSAL	180
181	GAKVLSSVKD	RFINFFVGNT	INSSYFPDHP	LHSISVRRLK	ETKDGFMFLT	DQSYIDVLPE	240
241	FRDSYPIKYV	HAFESNNFIY	FLTVQRETLD	AQTFHTRIIR	FCSINSLGHS	YMEMPLECIL	300
301	TEKRKKRSTK	KEVFNILQAA	YVSKPGAQLA	RQIGASLNDD	ILFGVFAQSK	PDSAEPMDRS	360
361	AMCAFPIKYV	NDFFNKIVNK	NNVRCLQHFY	GNPNEHCFNR	TLLRNSSGCE	ARRDEYRTEF	420
421	TTALQRVDLF	MGQFSEVLLT	SISTFIKGD	TIANLGTSEG	RFMQVVVSRS	GPSTPHVNFL	480
481	LDSPVSPPEV	IVEHTLNQNG	YTLVITGKKI	TKIPLNGLGC	RHFQSCSQCL	SAPPFVQCGW	540
541	CHDKCVRSEE	CLSGTWTQOI	CLPAIYKVFP	NSAPLEGGTR	LTICGWDFGF	RRNNKFDLKK	600
600	TRVLLGNESC	TLTLSESTMN	TLKCTVGPAM	NKHFNMSIII	SNGHGTQYS	TFSYVDPVIT	660
661	SISPKYGPMA	GGTLLTLTGN	YLNNGNSRHI	SIGGKTCTLK	SVSNSILECY	TPAQTISTEF	720
721	AVKLKIDLAN	RETSIFSRYE	DPIVYEIHPT	KSFISGGSTI	TGVGKNLNSV	SVPRMVINVH	780
781	EAGRNFVAC	QHRNSSEIIC	CTTPSLQQLN	LQLPLKTKAF	FMLDGILSKY	FDLIYVHNPNV	840
841	FKPFEKPVMI	SMGNENVLEI	KGNDIDPEAV	KGEVLKVGNK	SCENIHLHSE	AVLCTVPNDL	900
901	LKLNSSELNIE	WKQAISSTVL	GKVVIVQPDQ	FTGLIAGVVS	ISTALLLLLG	FFLWLKKRKQ	960
961	IKDLGSELVR	YDARVHTPHL	DRLVSARSVS	PTTEMVSNES	VDYRATFPED	QFPNSSQNGS	1020
1021	CRQVQYPLTD	MSPILTSQDS	DISSPLLQNT	VHIDLSALNP	ELVQAVQHV	IGPSSLIHVF	1080
1081	NEVIGRGHFG	CVYHGTLN	DGKKIHC	SLNRITDIGE	VSQFLTEGII	MKDFSHPNVL	1140
1141	SLLGICLRSE	GSPLVLPYM	KHGDLRNFIR	NETHNPTVKD	LIGFGLQVAK	GMKYLASKKF	1200
1201	VHRDLAARNC	MLDEKFTVKV	ADFGLARDMY	DKEYYSVHNK	TGAKLPVKWM	ALESLOTQKF	1260
1261	TTKSDVWSFG	VLLWELMTRG	APPYPDVNTF	DITVYLLQGR	RLQPEYCPD	PLYEVMLKCV	1320
1321	HPKAEMRPSF	SELVSRIAS	FSTFIGEHYV	HVNATYVNVK	CVAPYPSLLS	SEDNADDEVD	1380
1381	TRPASFWETS						1440

blue: MET sequence expressed in fusion protein Red: variant in fusion protein

¹NCBI/Protein accession number NP_000236.2

Please notice:

Variant amino acid numbering beginning with Ser755 when referring to GenBank accession J02958 (additional 18 aa exon between S755/G756, frequently found in the literature)