

ProQinase™ MET Y1230A

met proto-oncogene

Recombinant Human Active Protein Kinase

HGNC Symbol: MET

Synonyms: c-MET, HGFR

Product No.: 0944-0000-1

Lot: 002

Description: Human MET C-terminal fragment, amino acids K₉₅₆-S₁₃₉₀ (as in NCBI/Protein entry NP_000236.2), Y1230A mutant, N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: MET Y1230A Lot 002 product identity was confirmed by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 78,695 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

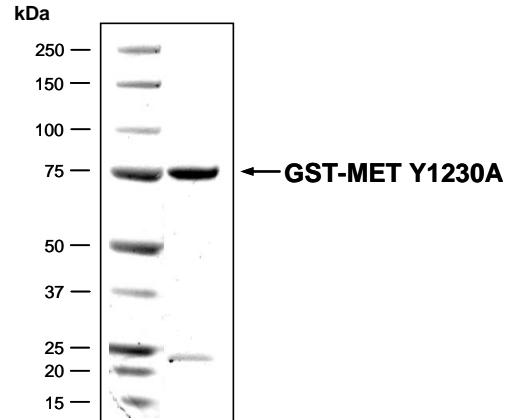
Storage temperature: -80°C

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

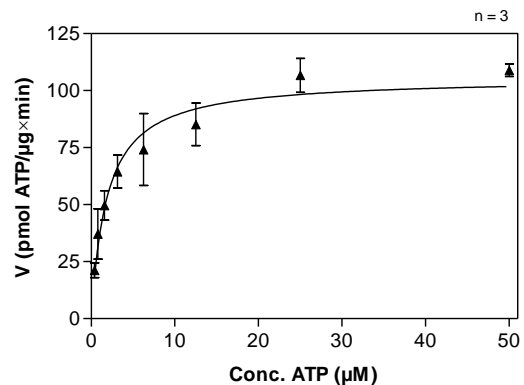
Biochemical Parameters:
Specific kinase activity (P_i transfer): 106 pmol/µg x min
ATP-K_M: 1.9 µM

MET Y1230A Lot 002:
Coomassie stain



2.0 µg GST-MET Y1230A

MET Y1230A Lot 002:
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 Y1230A Lot 002 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



ProQinase™ MET Y1230A

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MET Y1230A Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSM	IIRYIADKH	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHG	RRRASVAAGI	240
241	LVPRGSPGLD	GICSIEEFKK	RKQIKDLGSE	LVRVDARVHT	PHLDRLVSAR	SVSPTTEMVS	300
301	NESVDYRATF	PEDQFPNSSQ	NGSCRQVQYP	LTDMSPI LTS	GSDISSPLL	QNTVHIDL SA	360
361	LNPELVQAVQ	HVVIGPSSLI	VHFNEVIGRG	HFGCVYHGTL	LDNDGKKIHC	AVKSLNRITD	420
421	IGEVSQFLTE	GIIMKDFSH	NVLSLLGICL	RSEGSPLVVL	PYMKHGDLRN	FIRNETHNPT	480
481	VKDLIGFGLQ	VAKGMKYLAS	KKFVHRDLAA	RNCMLDEKFT	VKVADFLGAR	DMADKEYYSV	540
541	HNKTGAKLPEV	KWMALESLOT	QKFTTKSDVW	SFGVLLWELM	TRGAPPYDPV	NTFDITVYLL	600
600	QGRRLLOPEY	CPDPLYEVML	KCWHPKAEMR	PSFSELVSRI	SAIFSTFIGE	HYVHV NATYV	660
661	NVKCVAPYPS	LLSSEDNADD	EVDTRPASFW	ETS			720

1-218: GST Red: HIS6-tag Pink: Thrombin cleavage site blue: MET fragment boxed: Y1230A

MET wt ¹ Amino Acid Sequence							
1	MKAPAVLAPG	ILVLLFTLVQ	RSNGECKEAL	AKSEMNVNMK	YQLPNFTAET	PIQNVILHEH	60
61	HIFLGATNYI	YVLNEEDLQK	VAEYKTGPVL	EHPDCFPQD	CSSKANLSSG	VWKDINMAL	120
121	VVDYYDDQL	ISCGSVNRGT	CQRHVFPNH	TADIQSEVHC	IFSPQIEEPS	QCPCVVSAL	180
181	GAKVLSSVKD	RFINFFVGNT	INSSYFPDHP	LHSISVRLK	ETKDGFMFLT	DQSYIDLVE	240
241	FRDSYPIKYV	HAFESNNFIY	FLTVQRETL	AQTFHTRIIR	FCSINSLGHS	YMEMPLCIL	300
301	TEKRKRSTK	KEVFNILQAA	YVSKPGAQLA	RQIGASLNDD	ILFGVFAQSK	PDSAEPMDRS	360
361	AMCAFPKIVY	NDFFNKIVNK	NNVRCLQHFY	GNHEHCFNR	TLLRNSSGCE	ARRDEYRTEF	420
421	TTALQRVDLF	MGQFSEVLLT	SISTFIKGD	TIANLGTSEG	RFMQVVVRS	GPSTPHVNF	480
481	LDSHPVSPV	IVEHTLNQNG	YTLVITGKKI	TKIPLNGLC	RHFQSCSQCL	SAPPFVQCGW	540
541	CHDKCVRSEE	CLSGTWTQQI	CLPAIYKVF	NSAPLEGGTR	LTICGWDFGF	RRNNKFDLKK	600
600	TRVLLGNESC	TLTLESTMN	TLKCTVGPAM	NKHFNSIII	SNGHGTQYS	TFSYVDPVIT	660
661	SISPKYGPMA	GGTLLTLTGN	YLNNGNSRHI	SIGGKTCTLK	SVNSILECY	TPAQTISTEF	720
721	AVKLKIDLAN	RETSIFS YRE	DPIVYEIHPT	KSFISGGSTI	TGVGKNLNSV	SVPRMVINVH	780
781	EAGRNFVAC	QHRNSEIIC	CTTPSLQQLN	LQLPLKTKAF	FMLDGILSKY	FDLIYVHNPV	840
841	FKPFEKPVMI	SMGNENVLEI	KGNDIDPEAV	KGEVLKVGNK	SCENIHLHSE	AVLCTVPNDL	900
901	LKLNSELNIE	WKQAISSTVL	GKVIVQPDQN	FTGLIAGVVS	ISTALLLLLG	FFLWLKKRKQ	960
961	IKDLGSELVR	YDARVHTPHL	DRLVSARSVS	PTEMVSNES	VDYRATFPED	QFPNSSONGS	1020
1021	CRQVQYPLTD	MSPILTS GDS	DISSPLLQNT	VHIDLSALNP	ELVQAVQHV	IGPSSLIVHF	1080
1081	NEVIGRGHFG	CVYHGTL LDN	DGKKIHCAVK	SLNRITDIGE	VSQFLTEGII	MKDFSHPNVL	1140
1141	SLLGICLRSE	GSPLVLPYM	KHGD LNFIR	NETHNPTVKD	LIGFGLQVAK	GMKYLASKKF	1200
1201	VHRDLAARNC	MLDEKFTVKV	ADFG LARDMY	DKEYYSVHNC	TGAKLPVKWM	ALESLOTQKF	1260
1261	TTKSDVVSFG	VLLWELMTRG	APPYDPVNTF	DITVYLLQGR	RLLQPEYCPD	PLYEVMLKCW	1320
1321	HPKAEMRPSF	SELVSRI SAI	FSTFIGEHYV	HVNATYVNVK	CVAPYPSLLS	SEDNADDEV	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)

This product was manufactured at Reaction Biology in Freiburg, Germany, and is for in vitro research use only, not for use in humans or animals.
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