

TRK-A G667C

Tropomyosin receptor kinase A

Recombinant Human Active Protein Kinase

HGNC Symbol: NTRK1

Synonyms: MTC; TRK; TRK1; TRKA

Product No.: 1537-0000-1

Lot: 008

Description: Human TRK-A, C-terminal fragment, amino acids G₄₄₃-G₇₉₆ (as in NCBI/Protein entry NP_002520.2) with a G667C point mutation, N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: TRK-A G667C Lot 008, was confirmed as TRK-A by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 69,726 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: 0.13 µg/µl

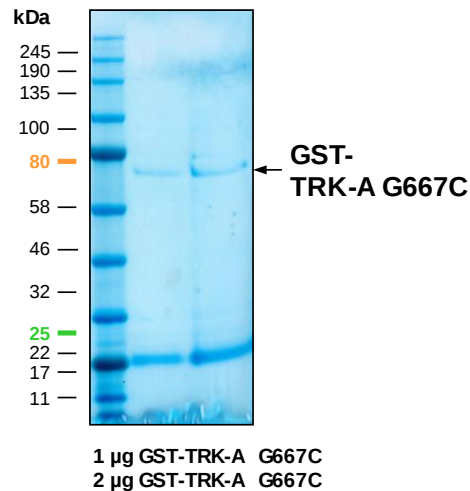
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

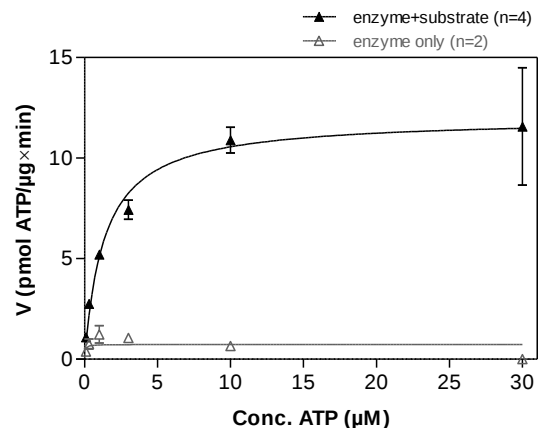
Specific kinase activity (Pi transfer): 12 pmol/µg×min

ATP-K_M: 1.4 µM

TRK-A G667C Lot 008: Coomassie stain



TRK-A G667C Lot 008: 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 (R₁₁-VYSTDYRLFNPS, 80 µg/ml)
 - TRK-A G667C: 1.0 µg/ml
- Filter binding assay
 - MSPH membrane (Millipore)

Additional assay technology: TRK-A G667C Lot 008

was also successfully tested by ProQinase for the use with the ADP-Glo™ Kinase assay from Promega. ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details



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TRK-A G667C 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	KRIEAIPOID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHGG	RRRASVAAGI	240
241	LVPRGSPGLD	GIYARGIQAS	MGRRNKFGIN	RPAVLAPEDG	LAMSLHFMTL	GGSSLSPTG	300
301	KGSGLQGHII	ENPQYFSDAC	VHHIKRRDIV	LKWELGEGAF	GKVFLAECHN	LLPEQDKMLV	360
361	AVKALKEASE	SARQDFQREA	ELLTMLQHQH	IVRFFGVCTE	GRPLLMVFY	MRHGDINRFL	420
421	RSHGPDALL	AGGEDVAPGP	LGLGQLLAVA	SQVAAGMVYL	AGLHVFVRDL	ATRNCILVGQG	480
481	LVVKICDFGM	SRDIYSTDY	RVGGRTMLPI	RWMPPEISILY	RKFTTESDVW	SFGVVLWEIF	540
541	TYGKQWPYQL	SNTEAIDCIT	QGRELERPRA	CPPEVYAIMR	GCWQREPQQR	HSIKDVHARL	600
601	QALAQAPPVY	LDVLG					660

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: TRK-A fragment **boxed**: G667C point mutation

TRK-A wt ¹ Amino Acid Sequence							
1	MLRGRRGQL	GWHSWAAGPG	SLLAWLILAS	AGAAPCPDAC	CPHGSSGLRC	TRDGALDSLH	60
61	HLPGAENLTE	LYIENQQHLQ	HLELRDLRGL	GELRNLTIVK	SGLRFVAPDA	FHFTPRLSRL	120
121	NLSFNALESL	SWKTVQGLSL	QELVLSGNPL	HCSCALRWLQ	RWEEEGLGGV	PEQKLQCHGQ	180
181	GPLAHMPNAS	CGVPTLVQV	PNASVDVGDD	VLLRCQVEGR	GLEQAGWILT	ELEQSATVMK	240
241	SGGLPSLGLT	LANVTSDLNR	KNVTCWAEND	VGRAEVSQV	NVSFPASVQL	HTAVEMHHWC	300
301	IPFSVDGQPA	PSLRWLFNGS	VLNETSFIFT	EFLEPAANET	VRHGCLRLNQ	PTHVNNGNYT	360
361	LLAANPFGQA	SASIMAAFMD	NPFEFNPEDP	IPVSFSPVDT	NSTSGDPVEK	KDETFPGVSV	420
421	AVGLAVFACL	FLSTLLLVLN	KCGRRNKFGI	NRPAVLAPED	GLAMSLHFMT	LGGSSLSPTG	480
481	GKSGLQGHI	IENPQYFSDA	CVHHIKRRDI	VLKWELGEGA	FGKVFLAECH	NLLPEQDKML	540
541	VAVKALKEAS	ESARQDFQRE	AELLTMLQHQ	HIVRFFGVCT	EGRPLLMVFE	YMRHGDINRFL	600
601	LRSHGPDACL	LAGGEDVAPG	PLGLGQLLAV	ASQVAAGMVY	LAGLHVFVRD	LATRNCILVGQ	660
661	GLVVKIGDFG	MSRDIYSTDY	YRVGGRTMLP	IRWMPPEISIL	YRKFTTESDV	WSFGVVLWEI	720
721	FTYGKQWPYQ	LSNTEAIDCI	TQGRELERPR	ACPPEVYAIM	RGCWQREPQQ	RHSIKDVHAR	780
781	LQALAQAPPV	YLDVLG					840

blue: TRK-A sequence expressed in fusionprotein

¹NCBI/Protein accession number NP_002520.2