

ProQinase™ TIE2 R849W

TEK receptor tyrosine kinase

Recombinant Human Active Protein Kinase

HGNC Symbol: TEK

Synonyms: CD202b, TIE-2

Product No.: 1085-0000-1

Lot: 001

Description: Human TIE2, C-terminal fragment, amino acids Q₇₇₁-A₁₁₂₄ (as in [NCBI/Protein](#) entry NP_000450.1) with a R849W mutation, N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: TIE2 R849W Lot 001, was confirmed as TIE2 by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 69,009 Da

Expression host: Sf9 insect cells

Purification: GST-Affinity Chromatography

Activation: in vitro auto activation

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: 0.177 µg/µl

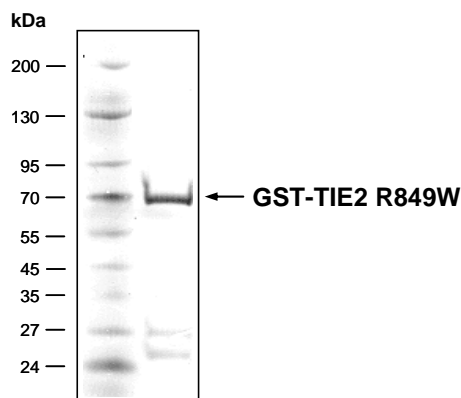
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

Specific kinase activity (P_i transfer): 88 pmol/µg × min

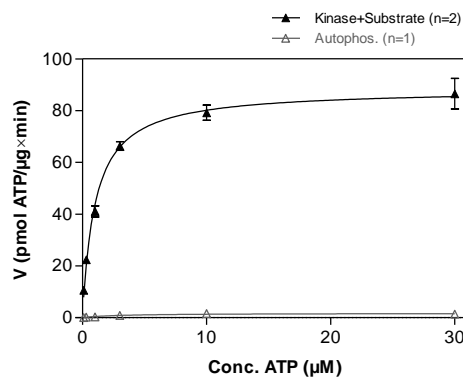
ATP-K_M: 1 µM

TIE2 R849W Lot 001:
Coomassie stain



2.0 µg GST-TIE2 R849W

TIE2 R849W 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: Poly(Ala,Glu,Lys,Tyr)_{6:2:5:1}, 40 µg/ml
 - Kinase: 1 µg/ml
- Filter binding assay
 - MSFC membrane (Millipore)

Additional assay technology:

TIE2 R849W Lot 001 was also successfully tested by Reaction Biology 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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GST-TIE2 R849W Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDKVLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPIQID	KYLKSSKYIA	WPLQGQWQATF	GGGDHPPKSD	PMG HHHHHG	RDS LEVLFQG	240
241	PLAMLQLKRA	NVQRMAQAF	QNVREPAVQ	FNSGTLALNR	KVKNNPDPTI	YPVLDWNDIK	300
301	FQDVIGEGNF	GQVLKARIK	DGLWMDAAIK	RMKEYASKDD	HRDFAGELEV	LCKLGHHPNI	360
361	INLLGACEHR	GYLylaIEYA	PHGNLLDFLR	KSRVLETDPA	FAIANSTAST	LSSQQLLHFA	420
421	ADVARGMDYL	SQKQFIHRDL	AARNILVGEN	YVAKIADFGL	SRGQEVYVKK	TMGRLPVRWM	480
481	AIESLNYSVY	TTNSDVWSYG	VLLWEIVSLG	GTPYCGMTCA	ELYEKLPGY	RLEKPLNCDD	540
541	EYVDLMRQCV	REKPYERPSF	AQILVSLNRM	LEERKTYVNT	TLYEKFTYAG	IDCSAEAEA	600

1-218: GST **Red**: HIS6-tag **Green**: 3C cleavage site **blue**: TIE2 fragment **boxed**: R849W mutation

TIE2 wt ¹ Amino Acid Sequence							
1	MDSLASLVLC	GVSLLLSGTV	EGAMDLILIN	SLPLVSDAET	SLTCIASGWR	PHEPITIGRD	60
61	FEALMNQHOD	PLEVTQDVTR	EWAKKVVWKR	EKASKINGAY	FCEGRVRGEA	IRIRTMKMRQ	120
121	QASFLPATLT	MTVDKGDNVN	ISFKKVLIKE	EDAVIYKNGS	FIHSVPRHEV	PDILEVHLPH	180
181	AQPQDAGVYS	ARYIGGNLFT	SAFTRLIVRR	CEAQKWGPEC	NHLCTACMNN	GVCHEDTGEC	240
241	ICPPGFMGRT	CEKACELHTF	GRTCKERCSG	QEGCKSYVFC	LPDPYGCSCA	TGWKGLQCNE	300
301	ACHPGFYGPD	CKLRCSNNG	EMCDRFQGL	CSPGWQLQC	EREGIPRMTF	KIVDLPDIE	360
361	VNSGKFNPI	KASGWPLPTN	EEMTLVKPDG	TVLHPKDFNH	TDHFSVAIFT	IHRILPPDSG	420
421	VWVCSVNTVA	GMVEKPFNIS	VKVLPKPLNA	PNVIDTGHN	AVINISSEPY	FGDGPIKSKK	480
481	LLYKPVNHYE	AWQHIQVTNE	IVTLNLYEPR	TEYELCVQLV	RRGEGGEGHP	GPVRRFTTAS	540
541	IGLPPPRGLN	LLPKSQTTLN	LTWQPIFPSS	EDDFYVEVER	RSVQKSDQQN	IKVPGNLTST	600
600	LLNNLHPREQ	YVVRARVNTK	AQGEWSEDLT	AWTSLDILPP	QPENIKISNI	THSSAVISWT	660
661	ILDGYSISSI	TIRYKVVQGN	EDQHVDVKK	NATITQYQLK	GLEPETAYQV	DIFAENNIGS	720
721	SNPAFSHEL	TLPEAQAPAD	LGGGKMLLIA	ILGSAGMTCL	TVLLAFLIIL	QLKRANVQRR	780
781	MAQAFQNVRE	EPAVQFNST	LALNRKVKN	PDPTIYPVLD	WNDIKFQDVI	GEGNFGQVLK	840
841	ARIKKGDLRM	DAAIKRMKEY	ASKDDHRDFA	GELEVLCKLG	HHPNIINLLG	ACEHRGYLYL	900
901	AIEYAPHGNL	LDFLRKSRL	ETDPAFAIAN	STASTLSSQQ	LLHFAADVAR	GMDYLSQKQF	960
961	IHRDLAARNI	LVGENYVAKI	ADFGLSRGQE	VYVKKTMGR	PVRWMAIESL	NYSVYTTNSD	1020
1021	VWSYGVLLWE	IVSLGGTPYC	GMTCAELYEK	LPQGYRLEKP	LNCDDDEVYDL	MRQCWREKPY	1080
1081	ERPSFAQILV	SLNRMLEERK	TYVNTTLYEK	FTYAGIDCSA	EEAA		1140

blue: TIE2 sequence expressed in recombinant protein **Red**: variant in recombinant protein

¹[NCBI/Protein](#) accession number NP_000450.1