

ProQinase™ TIE2 Y897S

TEK tyrosine kinase, endothelial (venous malformations, multiple cutaneous and mucosal)

Recombinant Human Active Protein Kinase

HGNC Symbol: TEK

Synonyms: TEK, TIE-2

Product No.: 1086-0000-1

Lot: 001

Description: Human TIE2, C-terminal fragment, amino acids Q₇₇₁-A₁₁₂₄ (as in NCBI/Protein entry NP_000450.2) with a Y897S mutation, N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: TIE2 Y897S Lot 001, was confirmed as TIE2 with a Y897S mutation by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 68,903 Da

Expression: Baculovirus infected Sf9 cells

Purification: GST-Affinity Chromatography

Activation: in vitro autoactivation

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

Storage temperature: -80°C
Avoid repeated freeze-thaw cycles!

Protein concentration: 0.113 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

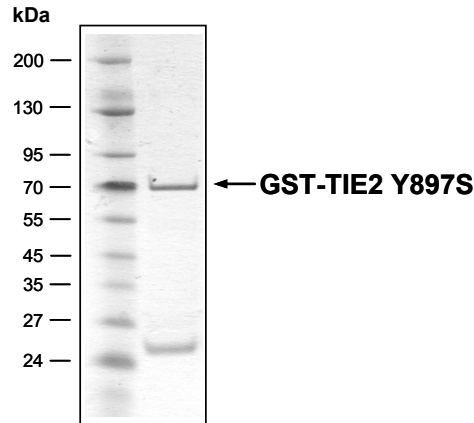
Biochemical Parameters:

Specific activity: 12 pmol/µg×min
ATP-K_M: 0.71 µM

Additional assay technology: TIE2 Y897S Lot 001

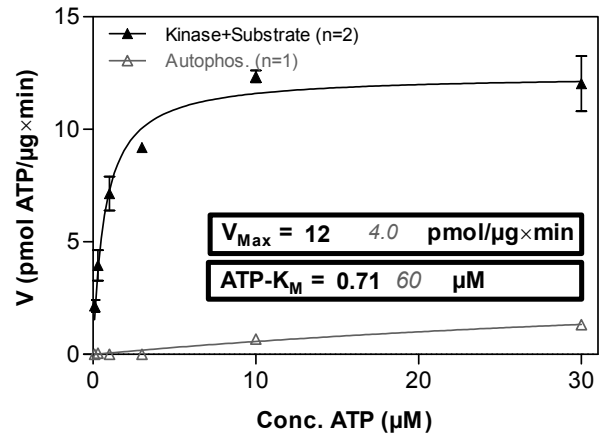
was also successfully tested by Reaction Biology for the use with the ADP-Glo™ Kinase assay from ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details

TIE2 Y897S Lot 001:
Coomassie stain



2.0 µg GST-TIE2 Y897S

TIE2 Y897S 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} (Sigma P-1152), 40 µg / ml
 - TIE2 Y897S: 1.0 µg / ml
- Filter binding assay
 - MSFC membrane (Millipore)

Recombinant Proteins

ProQinase™ TIE2 Y897S

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TIE2 Y897S Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSM	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHG	RDSLEVL FQG	240
241	PLAMLQLKRA	NVQRRMAQAF	QNVREEPAVQ	FNSGTLALNR	KVKNNPDPTI	YPVLDWNDIK	300
301	FQDVIGEGNF	GQVLKARIKK	DGLRMDAAIK	RMKEYASKDD	HRDFAGELEV	LCKLGHPNI	360
361	INLLGACEHR	GSLYLAIEYA	PHGNLLDFLR	KSRVLET DPA	FAIANSTAST	LSSQQLLHFA	420
421	ADVARGMDYL	SQKQFIHRDL	AARNILVGEN	YVAKIAD FGL	SRGQEVYVVK	TMGRLPVRWM	480
481	AIESLNYSVY	TTNSDVWSYG	VLLWEIVSLG	GTPYCGMTCA	ELYEKLPQGY	RLEKPLNCDD	540
541	EVYDLMRQCW	REKPYERPSF	AQILVSLNRM	LEERKTYVNT	TLYEKFTYAG	IDCSAEAEA	600

1-218: GST Red: HIS6-tag Green: 3C blue:TIE2 fragment S: Y897S mutation

TIE2 wt ¹ amino acid sequence							
1	MDSLASLVLC	GVSLLLSGTV	EGAMDLILIN	SLPLVSDAET	SLTCIASGWR	PHEPITIGRD	60
61	FEALMNQH QD	PLEVTQDVTR	EWAKKVVWKR	EKASKINGAY	FCEGRV RGEA	IRIRTMKMRQ	120
121	QASFLPATLT	MTVDKGDNVN	ISFKKVL I K	EDAVIYKNGS	FIHSVPRHEV	PDILEVHLP H	180
181	AQPQDAGVYS	ARYIGGNLFT	SAFTRLIVRR	CEAQKWGPEC	NHLCTACMNN	GVCHEDTGEC	240
241	ICPPGFMGRT	CEKACELHTF	GRTCKERCSG	QEGCKSYVFC	LDPDYGCSCA	TGWKGLQCNE	300
301	ACHPGFYGPD	CKLRCSCNNG	EMCDRFQ GCL	CSPGWQGLQC	EREGIPRMTP	KIVDLPDHIE	360
361	VNSGKFNPIC	KASGWPLPTN	EEMTLVKPDG	TVLHPKDFNH	TDHFSVAIFT	IHRILPPDSG	420
421	VWVCSVNTVA	GMVEKPFNIS	VKVLPKPLNA	PNVIDTGHNF	AVINISSEPY	FGDGIKSKK	480
481	LLYKPVNHYE	AWQHIQVTNE	IVTLNYLEPR	TEYELCVQLV	RRGEGGEGHP	GPVRRFTTAS	540
541	IGLPPPRGLN	LLPKSQTTLN	LTWQPIFPSS	EDDFYVEVER	RSVQKSDQQN	IKVPGNLTSV	600
600	LLNNLHPREQ	YVVRARVNTK	AQGEWSEDLT	AWTSLDILPP	QPENIKISNI	THSSAVISWT	660
661	ILDGYSISSI	TIRYKVQ GKN	EDQHVDVKIK	NATITQYQLK	GLEPETAYQV	DIFAENNIGS	720
721	SNPAFSHEL V	TLPE SQAPAD	LGGGKMLLIA	ILGSAGMTCL	TVLLAFLIIL	QLKRANVQRR	780
781	MAQAFQNVRE	EPAVQFN SGT	LALNRKVKNN	PDPTIYPVLD	WNDIKFQDVI	GEGNFGQVLK	840
841	ARIKDG LRM	DAAIKRMKEY	ASKDDHRDFA	GELEVLCKLG	HHPNIINLLG	ACEHRGYLYL	900
901	AIEYAPHGNL	LDFLRKS RVL	ETDPAF AIAN	STATLSSQQ	LLHFAADVAR	GMDYLSQKQF	960
961	IHRDLAARNI	LVGENYVAKI	ADFGLSRGQE	VYVKKTMGRL	PVRWMAIESL	NYSVYTTNSD	1020
1021	VWSYGVLLWE	IVSLGGTPYC	GMTCAELYEK	LPQGYRLEKP	LNCDDDEVYDL	MRQCWREKPY	1080
1081	ERPSFAQILV	SLNRMLEERK	TYVNTTLYEK	FTYAGIDCSA	EEAA		1140

blue: TIE2 sequence expressed in fusionprotein Red: variant in fusionprotein

¹NCBI/Protein accession number NP_000450.2

Recombinant Proteins