

## ProQinase™ EPHA4

EPH receptor A4

Recombinant Human Active Protein Kinase

HGNC Symbol: EPHA4

Synonyms: TYRO1, HEK8, SEK

Product No.: 0334-0000-1

Lot: 001

**Description:** Human Kinase, C-terminal fragment, amino acids S<sub>570</sub>-V<sub>986</sub> (as in [NCBI/Protein](#) entry NP\_004429.1), N-terminal GST-HIS<sub>6</sub> fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

**Product identity:** EPHA4, Lot 001, was confirmed as human EPHA4 by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW<sub>Fusion Protein</sub>:** 76,656 Da

**Expression host:** Sf9 insect cells

**Purification:** GST-Affinity Chromatography

**Activation:** This kinase was not activated by special procedures

**Storage buffer:** 50 mM TRIS-HCl pH 8.0, 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:** 0.628 µg/µl  
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

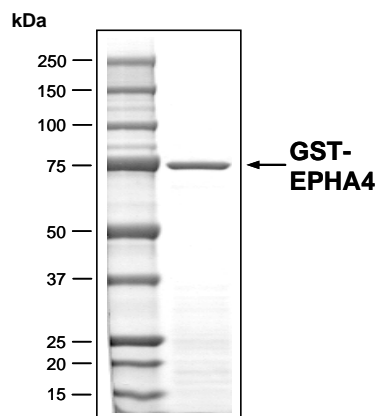
### Biochemical Parameters:

Specific kinase activity (P<sub>i</sub> transfer): 18.9 pmol/µg × min  
ATP-K<sub>M</sub>: 29 µM

### Additional assay technology:

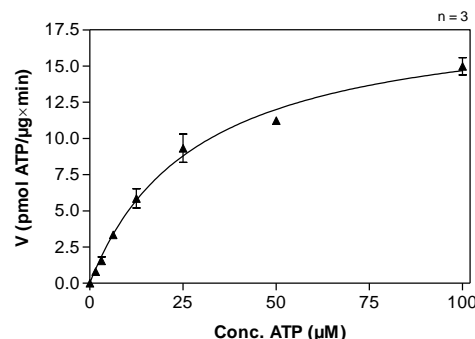
EPHA4 Lot 001 was also successfully tested by Reaction Biology for the use with the ADP-Glo™ Kinase assay from Promega ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details

### EPHA4 Lot 001: Coomassie stain



SP001, 2.0 µg

### EPHA4 Lot 001: Determination of V<sub>max</sub> and K<sub>M</sub> value for ATP



### Determination of K<sub>M</sub> value & Specific activity:

- Assay conditions:
  - 60 mM HEPES-NaOH, pH 7.5
  - 3 mM MgCl<sub>2</sub>
  - 3 mM MnCl<sub>2</sub>
  - 3 µM Na-orthovanadate
  - 1.2 mM DTT
  - 50 µg/ml PEG<sub>20,000</sub>
  - ATP (variable)
  - Substrate: Poly(E/Y)<sub>4:1</sub> 2.5 µg/ml
  - Kinase: 4 µg/ml
- Filter binding assay
  - MSFC membrane (Millipore)

# ProQinase™ EPHA4

Product No.: 0334-0000-1

GST-EPHA4 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSM	IIRYIADKH	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP	KMFEDRLCH	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI	PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMG <b>HHHHHG</b>	240
241	<b>LVPRG</b> SPGLD	GIC <b>SRSRRRS</b>	<b>KYSKAKQ</b> EAD	<b>EEKHLN</b> QGV	<b>TYVDPFTY</b> ED	<b>PNQAVR</b> FAK	300
301	<b>EIDASCI</b> KIE	<b>KVIGVGE</b> FGE	<b>VC</b> GRLKVP	<b>G</b> KREICVAIKT	<b>LKAGYTD</b> KQR	<b>RDFLSE</b> ASIM	360
361	<b>GQFDHP</b> NIH	<b>LEGVVTK</b> CKP	<b>VMIITE</b> YEMEN	<b>GSLDAFL</b> RKN	<b>DGRFTVI</b> QLV	<b>GMLRGI</b> GSGM	420
421	<b>KYLSDM</b> SYVH	<b>RDLAARN</b> ILV	<b>NSNLVCK</b> VSD	<b>FGMSRV</b> LEDD	<b>PEAAYT</b> TRGG	<b>KIPIRWT</b> APE	480
481	<b>AIAYRK</b> F TSA	<b>SDVWSY</b> GIVM	<b>WEVMSY</b> GERP	<b>YWDSMN</b> QDVI	<b>KAIEEGY</b> RLP	<b>PPMDR</b> PIALH	540
541	<b>QLMLDC</b> WQKE	<b>RSDRPK</b> FGQI	<b>VNMLDK</b> LIRN	<b>PNSLKR</b> TGTE	<b>SSRPNT</b> ALLD	<b>PSSPEF</b> SAVV	600
601	<b>SVGDWL</b> QAIK	<b>MDRYKDN</b> FTA	<b>AGYTTLE</b> AVV	<b>HVNQED</b> LARI	<b>GITAITHQ</b> NK	<b>ILSSVQ</b> AMRT	660
661	<b>QM</b> QMHGRMV	<b>PV</b>					720

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: EPHA4 fragment **boxed**: variation from RefSeq

EPHA4 wt <sup>1</sup> Amino Acid Sequence									
1	MAGIFYFALF	SCLFGICDAV	TGSRVYPANE	VTLLDSRSVQ	GELGWIASPL	EGGWEEVSIM	60		
61	DEKNTPIR	TY QVCNVMEPSQ	NNWLRTDWIT	REGAQRVYIE	IKFTLRDCNS	LPGVMGTCKE	120		
121	TFNLYYY	ESD	NDKERFIREN	QFVKIDTIAA	DESFTQVDIG	DRIMKLNTEI	RDVGPLSKKG	180	
181	FYLA	FQDVGA	CIALVSVRVF	YKCCPLTVRN	LAQFPDITG	ADTSSLVEVR	GSCVNNSEEK	240	
241	DVPKMY	CGAD	GEWLVP	IGNC	LCNAGHEERS	GECQACKIGY	YKALSTDATC	AKCPPHSYSV	300
301	WEGAT	SCTCD	RGFFRADNDA	ASMPCTRPPS	APLNLSNVN	ETSVNLEWSS	PQNTGGRQDI	360	
361	SYNVVCK	KCG	AGDPSKCRPC	GSGVHYTPQQ	NGLKTTKVS	I	TDLLAHTNYT	FEIWA	420
421	KYNPNPD	QSV	SVTVTTNQAA	PSSIALVQAK	EVTRYSVALA	WLEPDRPNV	ILEYEVKYYE	480	
481	KDQNER	SYRI	VRTAARNTDI	KGLNPLTSYV	FHVRARTAAG	YGDFSEPLEV	TTNTVPSRII	540	
541	GDGANST	VLL	VSVSGSVLV	VILIAAFVIS	<b>RRRSKYSKAK</b>	<b>QEADEEKHLN</b>	<b>QGVRTYVDPF</b>	600	
601	<b>TYEDPNQAVR</b>	<b>EFAKEIDASC</b>	<b>IKIEKVIGVG</b>	<b>EFGEVCSGRL</b>	<b>KVPGKREICV</b>	<b>AIKTLKAGYT</b>	660		
661	<b>DKQRRDFLSE</b>	<b>ASIMQFDHP</b>	<b>NIIHLEGVVT</b>	<b>KCKPVMITE</b>	<b>YMENGLDAF</b>	<b>LRKNDGRFTV</b>	720		
721	<b>IQLVGMLRGI</b>	<b>GSGMKYLSDM</b>	<b>SYVHRDLAAR</b>	<b>NILVNSNLVC</b>	<b>KVSDFGMSRV</b>	<b>LEDDPEAAYT</b>	780		
781	<b>TRGGKIP</b> IRW	<b>TAPEAIAYRK</b>	<b>FTSASDVWSY</b>	<b>GIVMWEVMSY</b>	<b>GERPYWMSN</b>	<b>QDVIKAI</b> EEG	840		
841	<b>YRLPPPMD</b> CP	<b>IALHQLMLDC</b>	<b>WQKERSDRPK</b>	<b>FGQIVNMLDK</b>	<b>LIRNPN</b> SLKR	<b>TGTESSRPNT</b>	900		
901	<b>ALLDPSSPEF</b>	<b>SAVVSVDWL</b>	<b>QAIKMDRYKD</b>	<b>NFTAAGYTTL</b>	<b>EAVVHV</b> NQED	<b>LARIGITAIT</b>	960		
961	<b>HQNKILSSVQ</b>	<b>AMRTQM</b> QMH	<b>GRMVPV</b>				1020		

**blue**: EPHA4 sequence expressed in recombinant protein **Red**: variant in recombinant protein

<sup>1</sup>[NCBI/Protein](#) accession number NP\_004429.1