

ProQinase™ EPHB2

EPH receptor B2

Recombinant Human Active Protein Kinase

HGNC Symbol: EPHB2

Synonyms: HEK5, TYRO5, DRT, EPHT3, ERK

Product No.: 0194-0000-3

Lot: 006

Description: Human EPHB2, C-terminal fragment, amino acids G₅₇₀-V₉₈₇ (as in [NCBI/Protein](#) entry NP_004433.1), N-terminal GST-HIS₆ fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

Product identity: EPHB2 Lot 006, was confirmed as Kinase by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 79,283 Da

Expression host: Sf9 insect cells

Purification: GST-Affinity Chromatography

Activation: in-vitro auto activation

Storage buffer: 50 mM TRIS-HCl pH 8.0, 100 mM NaCl, 5 mM DTT, 4 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: 0.131 µg/µl

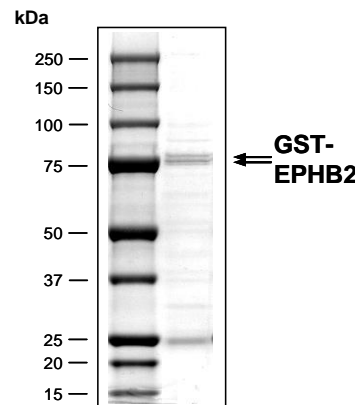
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

Biochemical Parameters:

Specific kinase activity (P_i transfer): 10 pmol/µg × min

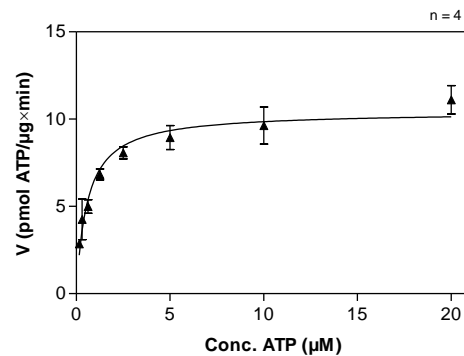
ATP-K_M: 0.6 µM

EPHB2 Lot 006: Coomassie stain



2.0 µg GST-EPHB2

EPHB2 Lot 006: 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} 20 µg/ml
 - Kinase: 2 µg/ml
- Filter binding assay
MSFC membrane (Millipore)

Additional assay technology:

EPHB2 Lot 006 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-EPHB2 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDKVLTQSM	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI	PQID KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMG HHHHHG	RRRASVAAGI	240
241	LVPRG PSGLD	GIYARGIQAS	MGARGRQCDG	YLQNSPLSMG	FERADSEYTD	KLQHYTSGHM	300
301	TPGMKIYIDP	FTYEDPNEAV	REFAKEIDIS	CVKIEQVIGA	GEFGEVCSGH	LKLPGKREIF	360
361	VAIKTLKSGY	TEKQRRD FLS	EASIMGQ FDH	PNVIHLE GVV	TKSTPVMII	EFMENG SLDS	420
421	FLRQNDGQ FT	VIQLVG MRLG	IAAGMKY LAD	MNYVHR DLAA	RNILVNS NLV	CKVSDF GLSR	480
481	FLEDDT SDPT	YTSALG GKIP	IRWTAPE AIQ	YRKFTS ASDV	WSYGIV MWEV	MSYGER PYWD	540
541	MTNQD VINAI	EQDYRL PPPM	DCPSAL HQLM	LDCWQ KDRNH	RPKFGQ IVNT	LDKMIR NPNS	600
601	LKAMA PLSSG	INLPL LDRTI	PDYTS FNVD	ERLEAI KMGQ	YKESF ANAGF	TSFDV VSQMM	660
661	MEDIL RVGVT	LAGHQ KKILN	SIQVM RAQMN	QIQS VEV			720

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: EPHB2 fragment **boxed**: variation from RefSeq

EPHB2 wt ¹ Amino Acid Sequence							
1	MALRRLGAAL	LLLPLLA	AVE ETLMDSTTAT	AELGWMVHPP	SGWEEVSGYD	ENMNTIRTYQ	60
61	VCNVFESSQN	NWLRTKF	FIR RGARIHVEM	KFSVRDCSSI	PSVPGSCKET	FNLYYYYEADF	120
121	DSATKTFPNW	MENPWVK	VDT IAADSFQV	DLGGRVMKIN	TEVRSFGPVS	RSGFYLAQD	180
181	YGGCMSLIAV	RVFYRKCP	PRI IQNGAIFQET	LSGAESTSLV	AARGSCIANA	EEVDVPIKLY	240
241	CNGDGEWLVP	IGRCMCKAG	F EAVENGTVCR	GCPSGTFKAN	QGDEACTHCP	INSRTTSEGA	300
301	TNCVCRNGYY	RADLDPLD	MMP CTTIPSAPQA	VISSVNETSL	MLEWTPPRDS	GGREDLVYNI	360
361	ICKSCGSGRG	ACTRCGDN	VQ YAPRQLGLTE	PRIYISDLLA	HTQYTFEIQ	A VNGVTDQSPF	420
421	SPQFASVNIT	TNQAAPSAV	S IMHQVSRVTD	SITLSWSQPD	QPNGVILDYE	LQYYEKELSE	480
481	YNATAIKSPT	NTVTVQGL	KA GAIYVFQVRA	RTVAGYGRYS	GKMYFQTMTE	AEYQTSIQEK	540
541	LPLIIGSSAA	GLVFLIAV	VV IAIVCNRRRG	FERADSEYTD	KLQHYTSGHM	TPGMKIYIDP	600
601	FTYEDPNEAV	REFAKEIDIS	CVKIEQVIGA	GEFGEVCSGH	LKLPGKREIF	VAIKTLKSGY	660
661	TEKQRRD FLS	EASIMGQ FDH	PNVIHLE GVV	TKSTPVMII	EFMENG SLDS	FLRQNDGQ FT	720
721	VIQLVG MRLG	IAAGMKY LAD	MNYVHR DLAA	RNILVNS NLV	CKVSDF GLSR	FLEDDT SDPT	780
781	YTSALG GKIP	IRWTAPE AIQ	YRKFTS ASDV	WSYGIV MWEV	MSYGER PYWD	MTNQD VINAI	840
841	EQDYRL PPPM	DCPSAL HQLM	LDCWQ KDRNH	RPKFGQ IVNT	LDKMIR NPNS	LKAMA PLSSG	900
901	INLPL LDRTI	PDYTS FNVD	EWLEAI KMGQ	YKESF ANAGF	TSFDV VSQMM	MEDIL RVGVT	960
961	LAGHQ KKILN	SIQVM RAQMN	QIQS VEV				1020

blue: EPHB2 sequence expressed in recombinant protein **Red**: variant in recombinant protein

¹NCBI/Protein accession number NP_004433.1