

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:

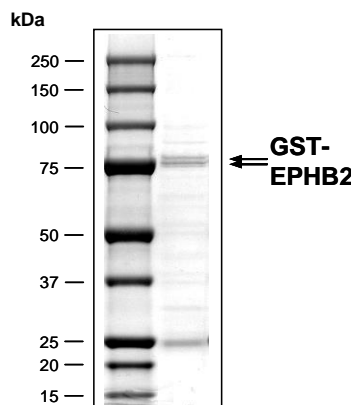
Specific kinase activity (P_i transfer): 10 pmol/µg × min

ATP-K_M: 0.6 µM

Additional assay technology:

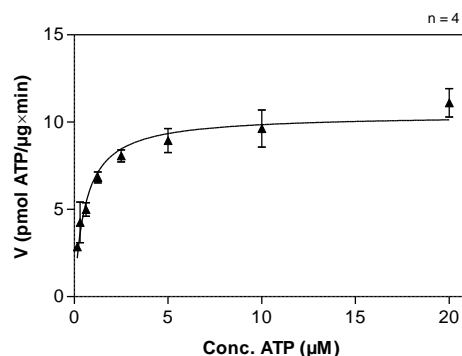
EPHB2 Lot 006 was also successfully tested by ProQinase for the use with the ADP-Glo™ Kinase assay from Promega
ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details

**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)

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GST-EPHB2 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	KRIEAIPOID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMG HHHHHG	RRRASVAAGI	240
241	LVPRG PSGLD	GIYARGIQAS	MGARGRQCDG	YLQNSPLSMG	FERADSEYTD	KLQHYTSGHM	300
301	TPGMKIYIDP	FTYEDPNEAV	REFAKEIDIS	CVKIEQVIGA	GEFGEVCSGH	LKLPKREIF	360
361	VAIKTLKSGY	TEKQRRDFLS	EASIMGQFDH	PNVIHLEGVV	TKSTPVMII	EFMENGSLDS	420
421	FLRQNDGQFT	VIQLVGMLRG	IAAGMKYLAD	MNYVHRDLAA	RNILVNSNLV	CKVSDFGLSR	480
481	FLEDDTSDPT	YTSALGGKIP	IRWTAPEAIQ	YRKFTSASDV	WSYGIVMWEV	MSYGERPYWD	540
541	MTNQDVINAI	EQDYRLPPPM	DCPSALHQLM	LDCWQKDRNH	RPKFGQIVNT	LDKMIRNPNS	600
601	LKAMAPLSSG	INLPLLDRTI	PDYTSFNTVD	ERLEAIKMGQ	YKESFANAGF	TSFDVVSQMM	660
661	MEDILRVGVT	LAGHQKILN	SIQVMRAQMN	QIQSVEV			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	LLLPLLAAVE	ETLMDSTTAT	AELGWMVHPP	SGWEEVSGYD	ENMNTIRTYQ	60
61	VCNVFESSQN	NWLRTKFIIR	RGAHRIHVEM	KFSVRDCSSI	PSVPGSCKET	FNLYYYYEADF	120
121	DSATKTFPNW	MENPWVKVDT	IAADESFSQV	DLGGRVMKIN	TEVRSFGPVS	RSGFYLAQFD	180
181	YGGCMSLIAV	RVFYRKCPRI	IQNGAIFQET	LSGAESTSLV	AARGSCIANA	EEVDVPIKLY	240
241	CNGDGEWLVP	IGRCMCKAGF	EAVENGTVCR	GCPSGTFKAN	QGDEACTHCP	INSRTTSEGA	300
301	TNCVCRNGYY	RADLDPLDMP	CTTIPSAPQA	VISSVNETSL	MLEWTPPRDS	GGREDLVYNI	360
361	ICKSCGSGRG	ACTRCGDNVQ	YAPRQLGLTE	PRIYISDLLA	HTQYTFEIQ	VNGVTDQSPF	420
421	SPQFASVNIT	TNQAAPSAVS	IMHQVSRVTD	SITLSWSQPD	QPNGVILDYE	LQYYEKELSE	480
481	YNATAIKSPT	NTVTVQGLKA	GAIYVFQVRA	RTVAGYGRYS	GKMYFQTMTE	AEYQTSIQEK	540
541	LPLIIGSSAA	GLVFLIAVVV	IAIVCNRRRG	FERADSEYTD	KLQHYTSGHM	TPGMKIYIDP	600
601	FTYEDPNEAV	REFAKEIDIS	CVKIEQVIGA	GEFGEVCSGH	LKLPKREIF	VAIKTLKSGY	660
661	TEKQRRDFLS	EASIMGQFDH	PNVIHLEGVV	TKSTPVMII	EFMENGSLDS	FLRQNDGQFT	720
721	VIQLVGMLRG	IAAGMKYLAD	MNYVHRDLAA	RNILVNSNLV	CKVSDFGLSR	FLEDDTSDPT	780
781	YTSALGGKIP	IRWTAPEAIQ	YRKFTSASDV	WSYGIVMWEV	MSYGERPYWD	MTNQDVINAI	840
841	EQDYRLPPPM	DCPSALHQLM	LDCWQKDRNH	RPKFGQIVNT	LDKMIRNPNS	LKAMAPLSSG	900
901	INLPLLDRTI	PDYTSFNTVD	EWLEAIKMGQ	YKESFANAGF	TSFDVVSQMM	MEDILRVGLT	960
961	LAGHQKILN	SIQVMRAQMN	QIQSVEV				1020

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

¹[NCBI/Protein](#) accession number NP_004433.1

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