

## EPHA3

EPH receptor A3

Recombinant Human Active Protein Kinase

HGNC Symbol: EPHA3

**Synonyms:** ETK; ETK1; EphA3; HEK; HEK4; TYRO4; EPH receptor A3

**Product No.:** 0351-0000-1

**Lot:** 001

**Description:** Human EPHA3, C-terminal fragment, amino acids G<sub>569</sub>-V<sub>983</sub> (as in [NCBI/Protein](#) entry NP\_005224.2), N-terminal GST-HIS<sub>6</sub> fusion protein with a Thrombin cleavage site, expressed in Sf9 insect cells

**Product identity:** EPHA3 Lot 001, was confirmed as EPHA3 by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW<sub>Fusion Protein</sub>:** 75,680 Da

**Expression host:** Sf9 insect cells

**Purification:** GST-Affinity Chromatography

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

**Storage buffer:** 50 mM HEPES pH 7.5, 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:** 0.140 µg/µl  
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

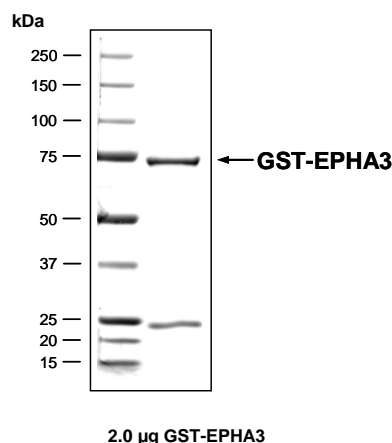
### Biochemical Parameters:

Specific kinase activity (P<sub>i</sub> transfer): 184 pmol/µg × min  
ATP-K<sub>M</sub>: 19.6 µM

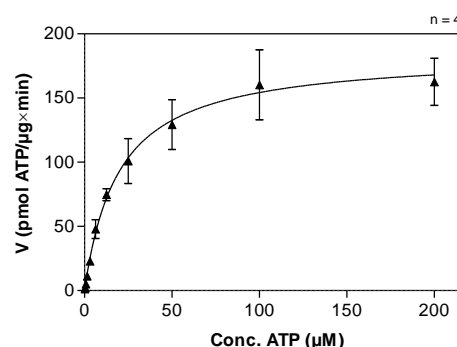
### Additional assay technology:

EPHA3 Lot 001 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

### EPHA3 Lot 001: Coomassie stain



### EPHA3 Lot 001: Determination of V<sub>max</sub> and K<sub>M</sub> value for ATP



- 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(Glu:Tyr)<sub>4:1</sub>, 20 µg/ml  
Kinase: 4 µg/ml
- Filter binding assay  
MSFC membrane (Millipore)

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GST-EPHA3 Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPOID	KYLKSSKYIA	WPLQGWAQTF	GGGDHPPKSD	PMG <b>HHHHHG</b>	RRRASVAAGI	240
241	<b>LVPRGSPGLD</b>	<b>GICSRGYKSK</b>	<b>HGADEKRLHF</b>	<b>GNGHLKLPGL</b>	<b>RTYVDPHTYE</b>	<b>DPTQAVHEFA</b>	300
301	<b>KELDATNISI</b>	<b>DKVVGAGEFG</b>	<b>EVCSGRLLKP</b>	<b>SKKEISVAIK</b>	<b>TLKVGYTEKQ</b>	<b>RRDFLGEASI</b>	360
361	<b>MGQFDHPNII</b>	<b>RLEGVVTKSK</b>	<b>PVMIVTEYME</b>	<b>NGSLDSFLRK</b>	<b>HDAQFTVIQL</b>	<b>VGMLRGIASG</b>	420
421	<b>MKYLSDMGYV</b>	<b>HRDLAARNIL</b>	<b>INSNLVCKVS</b>	<b>DFGLSRVLED</b>	<b>DPEAAYTTRG</b>	<b>GKIPIRWTSP</b>	480
481	<b>EAIAYRKFTS</b>	<b>ASDVWSYGIV</b>	<b>LWEVMSYGER</b>	<b>PYWEMSNDQV</b>	<b>IKAVDEGYRL</b>	<b>PPMDCPAAS</b>	540
541	<b>YQLMLDCWQK</b>	<b>DRNNRPKFEQ</b>	<b>IVSILDKLIR</b>	<b>NPGSLKIITS</b>	<b>AAARPSNLLL</b>	<b>DQSNVDITTF</b>	600
601	<b>RTTGDWLNQV</b>	<b>R<b>TAHCKEIFT</b></b>	<b>GVEYSSCDTI</b>	<b>AKISTDDMKK</b>	<b>VGVTVVGPOK</b>	<b>KIISSEIKALE</b>	660
661	<b>TQSKNGPVPV</b>						720

1-218: GST **Red**: HIS6-tag **Pink**: Thrombin cleavage site **blue**: EPHA3 fragment **boxed**: variation from RefSeq

EPHA3 wt <sup>1</sup> Amino Acid Sequence							
1	MDCQLSILL	LSCSVLDSFG	ELIPQPSNEV	NLLDSKTIQG	ELGWISYP SH	GWEEISGVDE	60
61	HYTPIRTYQV	CNVMDHSQNN	WLRTNWWPRN	SAQKIYVELK	FTLRDCNSIP	LVLGTCKETF	120
121	NLYYMESDDD	HGVKFRHQF	TKIDTIAADE	SFTQMDLGD	ILKLNTEIRE	VGPNVKKGFY	180
181	LAFQDVGACV	ALVSVRVYFK	KCPFTVKNLA	MFPDTPMDS	QSLVEVRGSC	VNNSKEEDPP	240
241	RMYCSTEGEW	LVPIGKCSN	AGYEERGFMC	QACRPGFYKA	LDGNMKCAK	PPHSSTQEDG	300
301	SMNCRCENNY	FRADKDPSPM	ACTRPPSSPR	NVISNINETS	VILDWSWPLD	TGGRKDVTFN	360
361	IICKKCGWNI	KQCEPCSPNV	RFLPRQFGLT	NTTIVTVDLL	AHTNYTFEID	AVNGVSELSS	420
421	PPRQFAAVSI	TTNQAAPSPV	LTIKKDRTSR	NSISLSWQEP	EHPNGIILDY	EVKYYEKQEQ	480
481	ETSYTILRAR	GTNVTISSLK	PDTIYVFQIR	ARTAAGYGTN	SRKFEFETSP	DSFSISGESS	540
541	QVVMIAISAA	VAIILLTVVI	YVLIGRFGCY	<b>KSKHGAEKR</b>	<b>LHFGNGHLKL</b>	<b>PGLRTYVDPH</b>	600
601	<b>TYEDPTQAVH</b>	<b>EFAKELDATN</b>	<b>ISIDKVVAG</b>	<b>EFGEVCSGRL</b>	<b>KLPSKKEISV</b>	<b>AIKTLKVGYT</b>	660
661	<b>EKQRDFLGE</b>	<b>ASIMQFDHP</b>	<b>NIIRLEGVVT</b>	<b>KSKPVMIVTE</b>	<b>YMENGLDSF</b>	<b>LRKHDAQFTV</b>	720
721	<b>IQLVGMLRGI</b>	<b>ASGMKYLSDM</b>	<b>GYVHRDLAAR</b>	<b>NILINSNLVC</b>	<b>KVSDFGLSRV</b>	<b>LEDDPEAAYT</b>	780
781	<b>TRGGKIPIRW</b>	<b>TSPEAIAYRK</b>	<b>FTSASDVWSY</b>	<b>GIVLWEVMSY</b>	<b>GERPYWEMSN</b>	<b>QDVIKAVDEG</b>	840
841	<b>YRLPPPMDCP</b>	<b>AA<b>LY</b>QMLDC</b>	<b>WQKDRNNRPK</b>	<b>FEQIVSILDK</b>	<b>LIRNPGSLKI</b>	<b>ITSAAARPSN</b>	900
901	<b>LLLDQSNVDI</b>	<b>TTFRTTGDWL</b>	<b>NGV<b>W</b>TAHCKE</b>	<b>IFTGVEYSSC</b>	<b>DTIAKISTDD</b>	<b>MKKVGVTVVG</b>	960
961	<b>PQKKIISIK</b>	<b>ALETQSKNGP</b>	<b>VPV</b>				1020

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

<sup>1</sup>NCBI/Protein accession number NP\_005224.2  
W924R: SNP variation see [NCBI/dbSNP](#) ID: rs35124509

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