

# EML4 ALK F1174L

Echinoderm microtubule-associated protein-like 4 anaplastic lymphoma kinase fusionprotein

Recombinant Human Active Protein Kinase

HGNC Symbol: n/a

Synonyms: n/a

Product No.: 1247-0000-1

Lot: 003

**Description:** Human pathological fusionprotein EML4 ALK, full length, amino acids M<sub>1</sub>-P<sub>1059</sub> (as in NCBI/Protein entry BAF73611.1) with a F1174L mutation, N-terminal GST-HIS<sub>6</sub> fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

**Product identity:** EML4 ALK F1174L Lot 003, was confirmed as EML4 ALK by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW<sub>Fusion Protein</sub>:** 145.413 Da

**Expression:** Baculovirus infected Sf9 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, 15 mM reduced glutathione, 20% glycerol

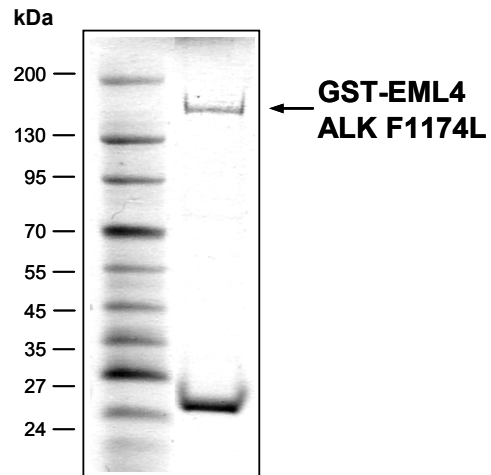
**Storage temperature:** -80°C  
Avoid repeated freeze-thaw cycles!

**Protein concentration:** 0.111 µg/µl (Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

**Biochemical Parameters:**

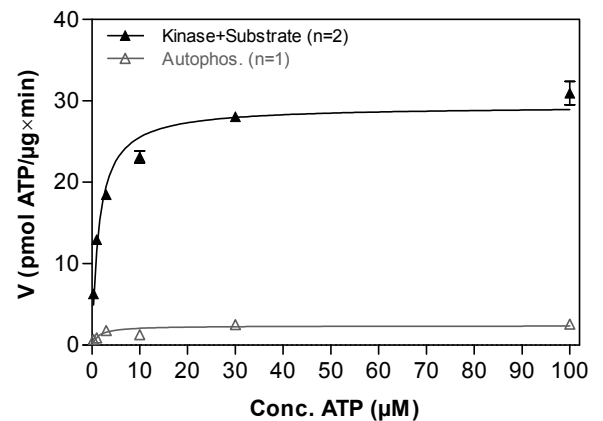
Specific activity: 29 pmol/µg×min  
ATP-K<sub>M</sub>: 1.5 µM

**EML4 ALK F1174L Lot 003:  
Coomassie stain**



2.0 µg GST-EML4 ALK F1174L

**EML4 ALK F1174L Lot 003:  
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: TRK-C derived peptide
  - EML4 ALK F1174L: 2.0 µg / ml
- Filter binding assay
  - MSPH membrane (Millipore)

**Additional assay technology:** EML4 ALK F1174L Lot 003

was also successfully tested by ProQinase for the use with the ADP-Glo™ Kinase assay from Promega

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EML4 ALK F1174L Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLPEML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAIPOID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHGG	RDSLEVLFGQ	240
241	PLAMDGFAGS	LDDSSAAST	SDVQDRLSAL	ESRVQQQEDE	ITVLKAALAD	VLRRLAISED	300
301	HVASVKKSVS	SKGQPSRAV	IPMSCITNGS	GANRKPSHTS	AVSIAGKETL	SSAAKSGTEK	360
361	KKEKPOGQRE	KKEESHSDNQ	SPQIRASPS	QPSSQPLQIH	RQTPESKNAT	PTKSIKRSP	420
421	AEKSHNSWEN	SDDSRNKLK	IPSTPKLIPK	VTKTADKHKD	VIINQEGEYI	KMFMRGRPIT	480
481	MFIPSDVDNY	DDIRTELPPE	KLKLEWAYGY	RGKDCRANVY	LLPTGEIVYF	IASVVVLFNY	540
541	EERTQRHYLG	HTDCVKCLAI	HPDKIRIATG	QIAGVDKDG	PLQPHVRVWD	SVTLSTLQII	600
601	GLGTFERGVG	CLDFSKADSG	VHLCVIDDSN	EHMLTVWDWQ	RKAKGAEIKT	TNEVVLAVEF	660
661	HPTDANTIIT	CGKSHIFFWT	WSGNSLTRKQ	GIFGKYEKPK	FVQCLAFLGN	GDVLTGDSGG	720
721	VMLIWSKTTV	EPTPGKGPV	YRRKHQELQA	QMELQSPPEY	KLSKLRTSTI	MTDYNPNYCF	780
781	AGKTSSISDL	KEVPRKNITL	IRGLGHGAFG	EVYEQVSGM	PNDPSPLQVA	VKTLPEVCSE	840
841	QDELDFLMEA	LIISKLNHQ	IVRCIGVSLQ	SLPRFILLEL	MAGGDLKSFL	RETRPRPSQP	900
901	SSLAMDLLH	VARDIACGCQ	YLEENHFIHR	DIAARNCLLT	CPGPGRVAKI	GDFGMARDIY	960
961	RASYRKGCC	AMLVWKMP	EAFMEGIFTS	KTDTWSFGVL	LWEIFSLGYM	PYPSKSNQEV	1020
1021	LEFYTSGGRM	DPPKNCPPV	YRIMTQCWQH	QPEDRPNFAI	ILERIEYCTQ	DPDVINTALP	1080
1081	IEYGPLVEEE	EKVPVRPKDP	EGVPLLVSQ	QAKREEERSP	AAPPPLPTTS	SGKAACKPTA	1140
1141	AEVSVRVRPG	PAVEGGHVM	AFSQNSRNK	LHRVHGSRNK	PTSLWNPTYG	SWFTEKPTTK	1200
1201	NNPIAKKEPH	ERGNLGLS	CTVPPNVATG	RLPGASLLE	PSSLTANMKE	VPLFRLRHFP	1260
1261	CGNVNYGYQQ	QGLPLEAATA	PGAGHYEDTI	LKSKNSMNQP	GP		1320

1-218: GST    Red: HIS6-tag    Green: 3C    blue:EML4ALK    F1174L ALK mutation

EML4 ALK wt <sup>1</sup> amino acid sequence							
1	MDGFAGSLDD	SISAASTSDV	QDRLSALESR	VQQQEDEITV	LKAALADVLR	RLAISEDHVA	60
61	SVKKSVS	QPSRAVIM	SCITNGSGAN	RKPSHTSAVS	IAGKETLSSA	AKSGTEKKKE	120
121	KPOGQREKKE	ESHSNDQSPQ	IRASPSQPS	SQPLQIHRQT	PESKNATPTK	SIKRPSPAEK	180
181	SHNSWENSDD	SRNKLKIPS	TPKLIKVTK	TADKHKDVII	NQEGEYIKMF	MRGRPITMFI	240
241	PSVDNYDDI	RTELPPEK	LEWAYGYRGK	DCRANVYLLP	TGEIVYFIAS	VVVLFNYEER	300
301	TQRHYLGHTD	CVKCLAIHPD	KIRIATGQIA	GVDKDGRLPQ	PHVRVWDSVT	LSTLQIIGLG	360
361	TFERGVGCLD	FSKADSGVHL	CVIDDSNEHM	LTVWDWQKKA	KGAEIKTTNE	VVLAVEFHPT	420
421	DANTIITCGK	SHIFFWTWSG	NSLTRKQGF	KGYEKPFVQ	CLAFLGNDV	LTGDSGGVML	480
481	IWSKTTVEPT	PGKGPVYRR	KHQELQAMQM	ELQSPEYKLS	KLRTSTIMTD	YNPNYCFAGK	540
541	TSSISDLKEV	PRKNITLIRG	LGHGAFGEVY	EGQVSGMPND	PSPLQVAVKT	LPEVCSEQDE	600
601	LDFLMEALII	SKFNHQNIVR	CIGVSLQSLP	RFILLELMAG	GDLKSFLRET	RPRPSQPSSL	660
661	AMLDLLHVAR	DIACGCQYLE	ENHFIHRDIA	ARNCLLTCPG	PGRVAKIGDF	GMARDIYRAS	720
721	YYRKGCCAML	PVKWMPPEAF	MEGIFTSKTD	TWSFGVLLWE	IFSLGYMPYP	SKSNQEVLEF	780
781	VTSGGRMDPP	KNCPGPVYRI	MTQCWQHQP	DRPNFAIILE	RIEYCTQDPD	VINTALPIEY	840
841	GPLVEEEKV	PVRPKDPEGV	PPLLVSQOAK	REEERSPAAP	PPLPTTSSGK	AAKKPTAAEV	900
901	SVRVRPGPAV	EGGHVMAFS	QSNPPSELHR	VHGSRNKPTS	LWNPTYGSWF	TEKPTKKNP	960
961	IAKKEPHERG	NLGLSCTV	PPNVATGRLP	GASLLEPSS	LTANMKEVPL	FRLRHFP	1020
1021	VNYGYQQQL	PLEAATAPGA	GHYEDTILKS	KNSMNQPGP			1080

**bold letters:** expressed part of EML4 (blue) and ALK (green)    **RED** letters: variant in Fusionprotein

<sup>1</sup>NCBI/Protein accession number BAF73611.1

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