

EGF-R L861Q

epidermal growth factor receptor

Recombinant Human Active Protein Kinase

Synonyms: ERBB, ERBB1

Product No.: 1036-0000-1

Lot: 002

Description: Human EGF-R, C-terminal fragment, amino acids H₆₇₂-A₁₂₁₀ (as in GenBank entry NM_005228.3), L861Q mutant, N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: EGF-R L861Q Lot 001, was confirmed as EGF-R by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 89,216 Da

Expression: Baculovirus infected Sf9 cells

Purification: GST-Affinity Chromatography

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!

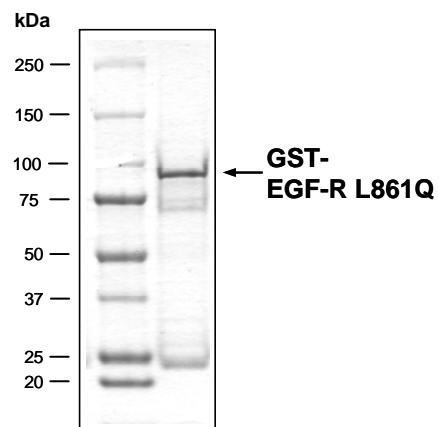
Protein concentration: 0.166 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

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(Glu:Tyr)_{4:1} (Sigma P-0275), 5.0 µg / ml
 - EGF-R L861Q: 1.0 µg / ml
- Filter binding assay
 - MSFC membrane (Millipore)

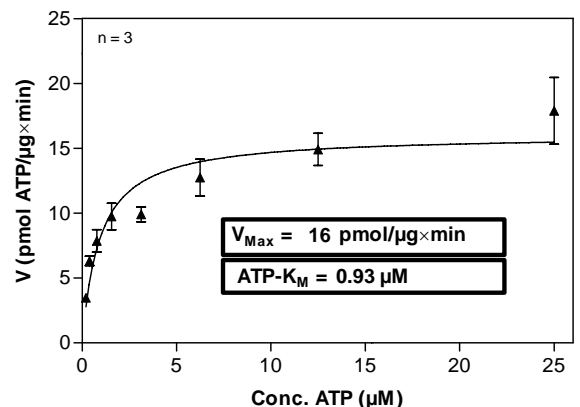
Specific activity: 16 pmol/µg×min

**EGF-R L861Q Lot 002:
Coomassie stain**



2.0 µg GST-EGF-R L861Q

**EGF-R L861Q Lot 002:
Determination of V_{max} and K_m value for ATP**



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EGF-R L861Q Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRLL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQ SMA	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFEDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMGHHHHHGG	RDSL E V L F Q G	240
241	PLAMGHIVRK	RTLRRLLQER	ELVEPLTPSG	EAPNQALLRI	LKETEFKKIK	VLSSGAFGTV	300
301	YKGLWIPEGE	KVKIPVAIKE	LREATSPKAN	KEILDEAYVM	ASVDNPHVCR	LLGICLTSTV	360
361	QLITQLMPFG	CLLDYVREHK	DNIGSQYLLN	WCVQIAKGMN	YLED RRLVHR	DLAARNVLVK	420
421	TPQHVKITDF	GLAKQLGAE E	KEYHAEGGKV	PIKWMALESI	LHRIYTHQSD	VWSYGVTVWE	480
481	LMTFGSKPYD	GIPASEISSI	LEKGERLPQP	PICTIDVYMI	MVKCWMIDAD	SRPKFRELI I	540
541	EF SKMARDPQ	RYLVIQ GDER	MHLPSPTDSN	FYRALMDEED	MDDVVD ADEY	LIPQQGFFSS	600
600	PSTSRTPLLS	SLSATSNNST	VACIDRNLQ	SCPIKEDSFL	QRYSSDPTGA	LTEDSIDDTF	660
661	LPVPEYINQS	VPKRPAGSVQ	NPVYHNQPLN	PAPSRDPHYQ	DPHSTAVGNP	EYLN TVQPTC	720
721	VNSTFD SPAH	WAQKGS HQIS	LDNPDYQ QDF	FPKEAKPNGI	FKGSTAENAE	YLRVAPQSSE	780
781	FIGA						840

1-218: GST Red: HIS6-tag Green: 3C blue:EGF-R fragment RED: L861Q mutation

EGF-R wt ORF (taken from GenBank entry NM_005228.3)							
1	MRPSGTAGAA	LLALLAALCP	ASRALEEKKV	CQGTSNKLTQ	LGT FEDHFLS	LQRMFN NCEV	60
61	VLGNLEITYV	QRNYDLSFLK	TIQEVAGYVL	IALNTVERIP	LENLQIIRGN	MYYENS YALA	120
121	VLSNYDANKT	GLKELPMRNL	QEILHGAVRF	SNNPALCNVE	SIQWRDIVSS	DFLSNM SMDF	180
181	QNH LGSCQKC	DPSCPNGSCW	GAGEENCQKL	TKIICAQQCS	GRCRGKSPSD	CCHNQCAAGC	240
241	TGPRES DCLV	CRKFRDEATC	KDTC PPLMLY	NPTTYQMDVN	PEGKYSFGAT	CVKKCPRNYV	300
301	VTDHGSCVRA	CGADSYEMEE	DGVRKCKKCE	GPCRKVCNGI	GIGEFKDSLS	INATNIK HFK	360
361	NCTSISGDLH	ILPVAFRGDS	FTHTPPLDPQ	ELDILKTVKE	ITGFLLIQAW	PENRTDLHAF	420
421	ENLEIIRGRT	KQHGQFSLAV	VSLNITSLGL	RSLKEISDGD	VIISGNKNLC	YANTINWKKL	480
481	FGTSGQKTKI	ISNRGENSCK	ATGQVCHALC	SPEGCWGPEP	RDCVSCRNVS	RGRECVDKCN	540
541	LLEGE PREFV	ENSECIQCHP	ECLPQAMNIT	CTGRGP DNCI	QCAHYIDGPH	CVKTC PAVGM	600
600	GENNTLVWKY	ADAGHVCHLC	HPNCTYGCTG	PGLEGCP TNG	PKIPSIATGM	VGALL LLLLVV	660
661	ALGIGLFMRR	RHIVRKRTL R	RLLQERELVE	PLTPSGEAPN	QALLRILKET	EFKKIKVLGS	720
721	GAFGTVYKGL	WIPEGEKVKI	PVAIKELREA	TSPKANKEIL	DEAYVMASVD	NPHVCRL LGI	780
781	CLTSTVQLIT	QLMPFGCLLD	YVREHKDNIG	SQYLLNWCVQ	IAGMNYLED	RRLVHRDLAA	840
841	RNVLVKTPQH	VKITDFGLAK	LLGAE EKEYH	AEGGKVP IKW	MALESILHRI	YTHQSDVWSY	900
901	GVTVWELMTF	GSKPYDGIPA	SEISSILEKG	ERLPQPPICT	IDVYMIMVKC	WMIDADSRPK	960
961	FRELII EFSK	MARDPQRYLV	IQGDERMHL P	SPTDSNFYRA	LMDEEDMDDV	VDADEYLI PQ	1020
1021	QGGFSSPSTS	RTPLLSSLSA	TSNNSTVACI	DRNGLQSCPI	KEDSFLQ RYS	SDPTGALTED	1080
1081	SIDDTFLPVP	EYINQSVPKR	PAGSVQNPVY	HNQPLNPAPS	RDPHYQDPHS	TAVGNPEYLN	1140
1141	TVQPTCVNST	FDSPA HWAQK	GSHQISLDNP	DYQQDFFPKE	AKPNGIFKGS	TAENAEYLRV	1200
1201	APQSSEFIGA						1260

blue: fragment of EGF-R expressed in fusionprotein RED: L861 mutation site

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