

EGF-R d747-752/P753S

epidermal growth factor receptor

Recombinant Human Active Protein Kinase

Synonyms: ERBB, ERBB1

Product No.: 1039-0000-1

Lot: 001

Description: Human EGF-R, C-terminal fragment, amino acids H₆₇₂-A₁₂₁₀ (as in GenBank entry NM_005228.3), amino acids 747-752 deleted, P753S mutant, N-terminal GST-HIS₆ fusion protein with a 3C cleavage site, expressed in Sf9 insect cells

Product identity: EGF-R d747-752/P753S Lot 001, was confirmed as EGF-R by mass spectroscopy LC-ESI-MS/MS

Theoretical MW_{Fusion Protein}: 88,503 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!

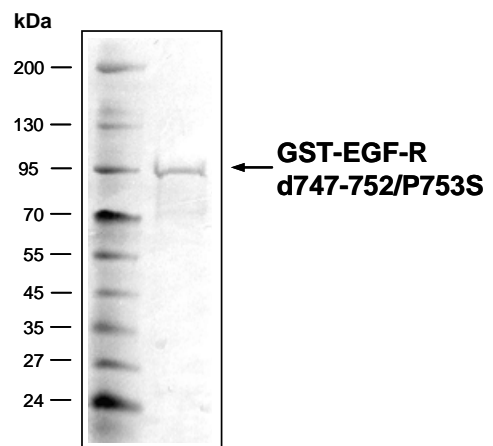
Protein concentration: 0.103 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

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 d747-752/P753S: 1.0 µg / ml
- Filter binding assay
MSFC membrane (Millipore)

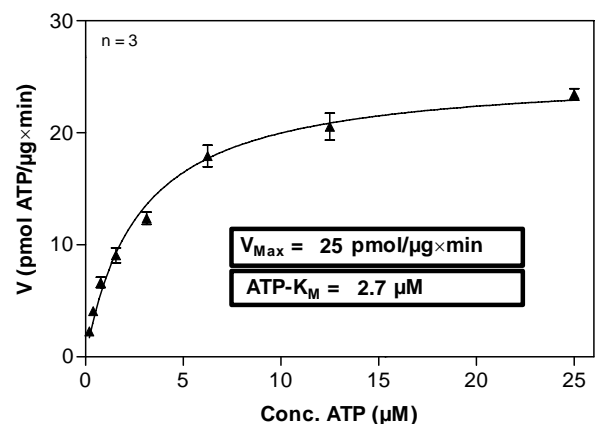
Specific activity: 25 pmol/µg×min

**EGF-R d747-752/P753S Lot 001:
Coomassie stain**



1.0 µg GST-EGF-R d747-752/P753S

**EGF-R d747-752/P753S Lot 001:
Determination of V_{max} and K_m value for ATP**



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EGF-R d747-752/P753S Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	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	PMGHHHHHHG	RDSLEVL FQG	240
241	PLAMGHIVRK	RTLRRLLQER	ELVEPLTPSG	EAPNQALLRI	LKETEFKKIK	VLGSGAFGTV	300
301	YKGLWIPEGE	KVKIPVAIKE	<u>SKANKEILDE</u>	AYVMASVDNP	HVCRLLGICL	TSTVQLITQL	360
361	MPFGCLLDYV	REHKDNIGSQ	YLLNWCVQIA	KGMNYLEDNR	LVHRDLAARN	VLVKT PQHVK	420
421	ITDFGLAKLL	GAEEKEYHAE	GGKVPIKWMA	LESILHRIYT	HQSDVWSYGV	TVWELMTFGS	480
481	KPYDGIPASE	ISSILEKGER	LPQPPICTID	VYMIMVKCWM	IDADSRPKFR	ELIIEFSKMA	540
541	RDPQRYLVIQ	GDERMHL PSP	TDSNFYRALM	DEEDMDDVVD	ADEYLIPQQG	FFSSPSTSRT	600
600	PLSSLSATS	NNSTVACIDR	NGLQSCPIKE	DSFLQRYSSD	PTGALTEDSI	DDTFLPVP EY	660
661	INQSVPKRPA	GSVQNPVYHN	QPLNPAPSRD	PHYQDPHSTA	VGNPEYLN TV	QPTCVNSTFD	720
721	SPAHW A QKS	HQISLDNPDY	QQDFFPKEAK	PNGIFKGSTA	ENAEYLRVAP	QSSEFIGA	780

1-218: GST **Red: HIS6-tag** **Green: 3C** **blue: EGF-R fragment** underlined: aa flanking the deletion **red:** point mutation

EGF-R wt ORF (taken from GenBank entry NM_005228.3)							
1	MRPSGTAGAA	LLALLAALCP	ASRALEEKKV	CQGTSNKLTQ	LGTFEDHFLS	LQRMFN NCEV	60
61	VLGNLEITYV	QRNYDLSFLK	TIQEVAGYVL	IALNTVERIP	LENLQIIRGN	MYYENS YALA	120
121	VLSNYDANKT	GLKELPMRNL	QEILHGAVRF	SNNPALCNVE	SIQWRDIVSS	DFLSNMSMDF	180
181	QNH LGSCQKC	DPSCPNGSCW	GAGEENCQKL	TKIICAQQCS	GRCRGKSPSD	CCHNQCAAGC	240
241	TGPRES DCLV	CRKFRDEATC	KDTC PPLMLY	NPTTYQMDVN	PEGKYSFGAT	CVKKCPRNYV	300
301	VDHGSCVRA	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 PEFV	ENSECIQCHP	ECLPQAMNIT	CTGRGPDNCI	QCAHYIDGPH	CVKTC PAVGM	600
600	GENNTLVWKY	ADAGHVCHLC	HPNCTYGCTG	PGLEGCP TNG	PKIPSIATGM	VGALLLLL LVV	660
661	ALGIGLFMR R	RHIVRKRTL R	RLLQERELVE	PLTPSGEAPN	QALLRILKET	EFKKIKVLGS	720
721	GAFGTVYKGL	WIPEGEKVKI	PVAIKELREA	TSPKANKEIL	DEAYVMASVD	NPHVCRL LGI	780
781	CLTSTVQLIT	QLMPFGCLLD	YVREHKDNIG	SOYLLNWCVQ	IAKGMNYLED	RRLVHRDLAA	840
841	RNVLVKTPQH	VKITDFGLAK	LLGAEEKEYH	AEGGVPIKW	MALESILHRI	YTHQSDVWSY	900
901	GVTVWELMTF	GSKPYDGIPA	SEISSILEKG	ERLPQPPICT	IDVYMIMVKC	WMIDADSRPK	960
961	FRELIIEFSK	MARDPQRYLV	IQDERMHL P	SPTDSNFYRA	LMDEEDMDDV	VDADEYLIPQ	1020
1021	QGFFSSPSTS	RTPLLSSLSA	TSNNSTVACI	DRNGLQSCPI	KEDSFLQRY S	SDPTGALTED	1080
1081	SIDDTFLPVP	EYINQSVPKR	PAGSVQNPVY	HNQPLNPAPS	RDPHYQDPHS	TAVGNPEYLN	1140
1141	TVQPTCVNST	FDSPAHW A QK	GSHQISLDNP	DYQQDFFPKE	AKPNGIFKGS	TAENAEYLRV	1200
1201	APQSSEFIGA						1260

blue: fragment of EGF-R expressed in fusionprotein **RED:** 747-752 deletion, P753 mutation site

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