

## ProQinase™ PIK3CB L1049R/PIK3R1

phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit beta  
phosphoinositide-3-kinase regulatory subunit 1

Recombinant Human Active Lipid Kinase

HGNC Symbol: PIK3CB

**Synonyms:** P110BETA, PI3K, PI3KBETA, PI3K-beta, PIK3C1

**Lipid Kinase Family:** PI3K Class I

(according to: Phylogenomics of phosphoinositide lipid kinases: perspectives on the evolution of second messenger signaling and drug discovery: James R Brown & Kurt R Auger; BMC Evolutionary Biology 11, 4-14 (2011))

**Product No.:** 1585-1165-1

**Lot:** 005

**Description:** Human PIK3CB, full length, amino acids M<sub>1</sub>-S<sub>1070</sub> (as in NCBI/Protein entry NP\_006210.1) with a L1049R mutation, N-terminal GST-HIS6 fusion protein with a 3C cleavage site and PIK3R1 full length, amino acids M<sub>1</sub>-R<sub>724</sub> (as in NCBI/Protein entry NP\_852664.1), N-terminal fused to a MYC-tag, expressed in Sf9 insect cells

**Product identity:** PIK3CB L1049R/PIK3R1 Lot 005, was confirmed as PIK3CB by mass spectroscopy LC-ESI-MS/MS

**Theoretical MW<sub>PIK3CB L1049R</sub>:** 151,310 Da

**Theoretical MW<sub>PIK3R1</sub>:** 85,371 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, 15 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:** 0.122 µg/µl

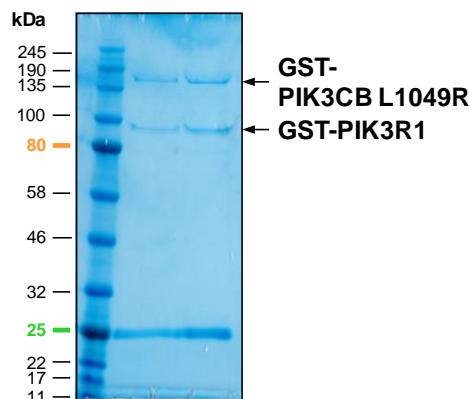
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

**Biochemical Parameters:**

Specific kinase activity (P<sub>i</sub> transfer): 2397 pmol/µg × min

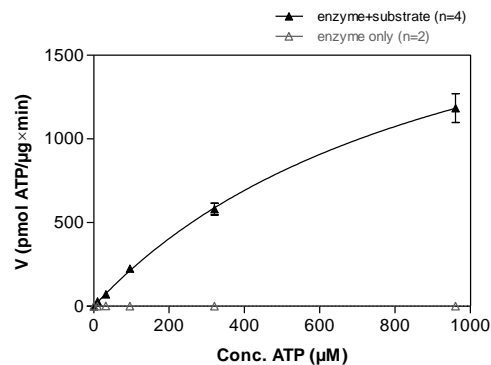
ATP-K<sub>M</sub>: 985 µM

**PIK3CB L1049R/PIK3R1 Lot 005:  
Coomassie stain**



1 µg PIK3CB L1049R/PIK3R1  
2 µg PIK3CB L1049R/PIK3R1

**PIK3CB L1049R/PIK3R1 Lot 005:  
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 µM Na-orthovanadate
  - 1.2 mM DTT
  - 50 µg/ml PEG<sub>20,000</sub>
  - ATP (variable)
  - Substrate: PIP<sub>2</sub>: 50 µM / PS: 950 µM
  - PIP<sub>2</sub>: 08:0 PI(4,5)P<sub>2</sub> (1,2-Dioctanoyl-sn-Glycero-3-(Phosphoinositol-4,5-Bisphosphate))
  - PS: 1-Palmitoyl-2-Oleoyl-sn-Glycero-3-[Phospho-L-Serine]
  - Kinase: 4 µg/ml
- ADP-Glo™ assay (Promega)

# ProQinase™ PIK3CB L1049R/PIK3R1

Product No.: 1585-1165-1

GST-PIK3CB L1049R Recombinant Fusion Protein Amino Acid Sequence							
1	MSPILGYWKI	KGLVQPTRL	LEYLEEKYEE	HLYERDEGDK	WRNKKFELGL	EFPNLPYYID	60
61	GDVKLTQSM	IIRYIADKHN	MLGGCPKERA	EISMLEGAVL	DIRYGVSRIA	YSKDFETLKV	120
121	DFLSKLP EML	KMFKDRLCHK	TYLNGDHVTH	PDFMLYDALD	VVLYMDPMCL	DAFPKLVCFK	180
181	KRIEAI PQID	KYLKSSKYIA	WPLQGWQATF	GGGDHPPKSD	PMG <b>HHHHHG</b>	RDS <b>LEVLFGQ</b>	240
241	<b>PLAMVMCFSF</b>	<b>IMPPAMADIL</b>	<b>DIWAVDSQIA</b>	<b>SDGSIPVDFL</b>	<b>LPTGIYIQLE</b>	<b>VPREATISYI</b>	300
301	<b>KQMLWKQVHN</b>	<b>YPMFNLLMDI</b>	<b>DSYMFACVNO</b>	<b>TAVYEELEDE</b>	<b>TRRLCDVRPF</b>	<b>LPVLKLVTRS</b>	360
361	<b>CDPGEKLD SK</b>	<b>IGVLIGKGLH</b>	<b>EFDSLKDPEV</b>	<b>NEFRRKMRKF</b>	<b>SEEKILSLVG</b>	<b>LSWMDWLKQT</b>	420
421	<b>YPPEHEPSIP</b>	<b>ENLEDKLYGG</b>	<b>KLIVAVHFEN</b>	<b>QODVFSFQVS</b>	<b>PNMNPIKVNE</b>	<b>LAIQKRLTIH</b>	480
481	<b>GKEDEVSPYD</b>	<b>YVLQVSGRVE</b>	<b>YVFGDHPLIQ</b>	<b>FQYIRNCVMN</b>	<b>RALPHFILVE</b>	<b>CCKIKKMYEQ</b>	540
541	<b>EMIAIEAAIN</b>	<b>RNSSNLPLPL</b>	<b>PPKKTRII SH</b>	<b>VWENNNPFQI</b>	<b>VLVKGNKLTN</b>	<b>EETVKVHVRA</b>	600
601	<b>GLFHGTSELLC</b>	<b>KTIVSSEVSG</b>	<b>KNDHIWNEPL</b>	<b>EFDINICDLP</b>	<b>RMARLCFAVY</b>	<b>AVLQKVKTKK</b>	660
661	<b>STKTINPSKY</b>	<b>QTIKAGKVH</b>	<b>YPVAWNTMV</b>	<b>FDKQGLRTG</b>	<b>DIILHSWSSF</b>	<b>PDELEMLNP</b>	720
721	<b>MGTVQTNPYT</b>	<b>ENATALHVKF</b>	<b>PENKKQPYYY</b>	<b>PPFDKIEKA</b>	<b>AEIASSDSAN</b>	<b>VSSRGGKFL</b>	780
781	<b>PVLKEILDRD</b>	<b>PLSQLCENEM</b>	<b>DLIWTLRQDC</b>	<b>REIFPQSLPK</b>	<b>LLLSIKWNKL</b>	<b>EDVAQLQALL</b>	840
841	<b>QIWPKLPPRE</b>	<b>ALELLDFNYP</b>	<b>DQYVREYAVG</b>	<b>CLRQMSDEEL</b>	<b>SOYLLQLVQV</b>	<b>LKYEPFLDCA</b>	900
901	<b>LSRFLLERAL</b>	<b>GNRRIGQFLF</b>	<b>WHLRSEVHIP</b>	<b>AVSVQFGVIL</b>	<b>EAYCRGSGVH</b>	<b>MKVLQSKQVEA</b>	960
961	<b>LNLKKTLSNL</b>	<b>IKLNAVKLN R</b>	<b>AKGKEAMHTC</b>	<b>LKQSA YREAL</b>	<b>SDLQSP LNPC</b>	<b>VILSELYVEK</b>	1020
1021	<b>CKYMDSKMKP</b>	<b>LWL VYNNKVF</b>	<b>GEDSVGVIFC</b>	<b>NGDDL RQDML</b>	<b>TLQMLRLMDL</b>	<b>LWKEAGLDR</b>	1080
1081	<b>MLPYGCLATG</b>	<b>DRSGLIEVVS</b>	<b>TSETIADIQL</b>	<b>NSSNVA AAAA</b>	<b>FNKDALLNL</b>	<b>KEYNSGDDL D</b>	1140
1141	<b>RAIEEFTLSC</b>	<b>AGYCVASYVL</b>	<b>GIGDRHSDNI</b>	<b>MVKKTGQLFH</b>	<b>IDFGHILGNF</b>	<b>KSKFGIKRER</b>	1200
1201	<b>VPFILTYDFI</b>	<b>HVIQOGKTGN</b>	<b>TEKFGRFRQC</b>	<b>CEDAYLILRR</b>	<b>HGNL FITLFA</b>	<b>LMLTAGLPEL</b>	1260
1261	<b>TSVKDIQYLK</b>	<b>DSLALGKSEE</b>	<b>EALKQFKQKF</b>	<b>DEA<b>RRESWTT</b></b>	<b>KVNWMAHTVR</b>	<b>KDYRS</b>	1320

1-218: GST **Red:** HIS6-tag **Green:** 3C cleavage site **blue:** PIK3CB **boxed:** L1049R mutation

PIK3CB wt <sup>1</sup> Amino Acid Sequence							
1	MCFSFIMPPA	MADILDIWAV	DSQIASD GSI	PVDFLLPTGI	YIQLEVPREA	TISYIKQMLW	60
61	KQVHNYPMFN	LLMDIDSYMF	ACVNQTAVYE	ELEDETRRLC	DVRPFLPVLK	LVTRSCDPGE	120
121	KLDSKIGVLI	GKGLHEFDSL	KDPEVNEFRR	KMRKFSEEKI	LSLVGLSWMD	WLKQTYPPPEH	180
181	EPSIPENLED	KLYGGKLIVA	VHFENCQDVF	SFQVSPNMNP	IKVNELAIQK	RLTIHGKEDE	240
241	VSPYDYVLQV	SGRVEYVFGD	HPLIQFQYIR	NCVMNRALPH	FILVECKIK	KMYEQEMIAI	300
301	EAAINRNSSN	LPLPLPKKT	RIISHVWENN	NPFQIVLVKG	NKLNTEETVK	VHVRAGLFHG	360
361	TELLCKTIVS	SEVSGKNDHI	WNEPLEFDIN	ICDLPRMARL	CFAYAVLDK	VTKKSKTKTI	420
421	NPSKYQTIRK	AGKVHPVAW	VNTMVDFDKG	QLRTGDIILH	SWSSFPDELE	EMLNPMGTVQ	480
481	TNPYTENATA	LHVKFPENKK	QPYYPFDFK	IIEKAAEIAS	SDSANVSSRG	GKKFLPVLKE	540
541	ILDRDPLSQL	CENEMDLIWT	LRQDCREIFP	QSLPKLLLSI	KWNKLEDVAQ	LQALLQIWPK	600
601	LPPREALELL	DFNYPDQYVR	EYAVGCLRQM	SDEELSQYLL	QLVQVLKYEP	FLDCALSREL	660
661	LERALGNRRI	GQFLFWHLRS	EVHIPAVSVQ	FGVILEAYCR	GSVGHMKVLS	KQVEALNKLK	720
721	TLNSLIKLNA	VKLNRAKGE	AMHTCLKQSA	YREALSDLQS	PLNPCVILSE	LYVECKKYMD	780
781	SKMKPLWLVS	NNKVFGEDEV	GVIFKNGDDL	RQDMLTLQML	RIMDLLWKEA	GLDLRMLPYG	840
841	CLATGDRSGL	IEVVSTSETI	ADIQLNSSNV	AAAAAFNKDA	LLNWLKEYNS	GDDLDR AIEE	900
901	FTLSCAGYCV	ASYVLGIGDR	HSDNIMVKKT	GQLFHIDFGH	ILGNFKSKFG	IKRERVPFIL	960
961	TYDFIHVIQQ	GKTGNTTEKFG	RFRQCCEDAY	LILRRHGNLF	ITL FALMLTA	GLPELTSVKD	1020
1021	<b>IQYLKDSLAL</b>	<b>GKSEEEALKQ</b>	<b>FKQKFDEALR</b>	<b>ESWTTKVNWM</b>	<b>AHTVRKDYRS</b>		1080

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

<sup>1</sup>[NCBI/Protein](#) accession number NP\_006210.1

**GST-Kinase Recombinant Fusion Protein Amino Acid Sequence**

1	MEEQKLISEE	DL	PMVMSAEG	YQYRALYDYK	KEREEDIDLH	LGDILTVMKG	SLVALGFSDG	60
61	QEARPEEIGW	LNGYNETTGE	RGDFPGTYVE	YIGRKKISPP	TPKPRPPRPL	PVAPGSSKTE		120
121	ADVEQQALTL	PDLAEQFAPP	DIAPPLLIK	VEAIEKKGLE	CSTLYRTQSS	SNLAELRQLL		180
181	DCDTPSVGLE	MIDVHVLADA	FKRYLLDLPN	PVIPAAYVSE	MISLAPEVQS	SEEIYQLLKK		240
241	LIRSPSIPHQ	YWLTLQYLLK	HFFKLSQTSS	KNLLNARVLS	EIFSPMLFRF	SAASSDNTEN		300
301	LIKVIEILIS	TEWNERQPAP	ALPPKPPKPT	TVANNGMNNN	MSLQDAEWYW	GDISREEVNE		360
361	KLRDTADGTF	LVRDASTKMH	GDYTLTLRKG	GNNKLIKIFH	RDGKYGFSDP	LTFSVVVELI		420
421	NHYRNESLAQ	YNPKLDVKLL	YPVSKYQQDQ	VVKEDNIEAV	GKKLHKYNTQ	FQEKRSREYDR		480
481	LYEEYTRTSQ	EIQMKRTAIE	AFNETIKIFE	EQCQTQERYS	KEYIEKFKRE	GNEKEIQRIM		540
541	HNYDKLKSRI	SEIIDSRRRL	EEDLKKQAAE	YREIDKRMNS	IKPDLIQLRK	TRDQYLMWLT		600
601	QKGVRRQKLN	EYLVNTEDED	QYSLVEDDED	LPHHDEKTNW	VGSSNRNKAE	NLLRKRDRGT		660
661	FLVRESSKQG	CYACSVVDG	EVKHCVINKT	ATGYGFAEYP	NLYSSLKELV	LHYQHTSLVQ		720
721	HNSLNVTLA	YPVYAQQRR						780

Red: MYC-tag blue: PIK3R1 boxed: variation from RefSeq

**PIK3R1 wt<sup>1</sup> Amino Acid Sequence**

1	MSAEGYQYRA	LYDYKKEREE	DIDLHLGDIL	TVNKGSLVAL	GFSQGQEARP	EEIGWLNQYN	60
61	ETTGERGDFP	GTIVEYIGRK	KISPPTPKPR	PPRPLPVAPG	SSKTEADVEQ	QALTLPLDLAE	120
121	QFAPPDIAPP	LLIKLVEAIE	KGLEECSTLY	RTQSSSNLAE	LRQLLDCDTP	SVDLEMIDVH	180
181	VLADAFKRYL	LDLNPVPIPA	AVYSEMISLA	PEVQSSEEIY	QLLKKLIRSP	SIPHQYWLT	240
241	QYLLKHFFKL	SQTSSKNLLN	ARVLSEIFSP	MLFRFSAASS	DNTENLIKVI	EILISTEWNE	300
301	RQPAPALPPK	PPKPTTVANN	GMNNMSLQD	AEWYWGDISR	EEVNEKLRDT	ADGTFVLRDA	360
361	STKMHGDYTL	TLRKGGNKLN	IKIFHRDGKY	GFSQDPLTFSS	VVELINHYRN	ESLAQYNPKL	420
421	DVKLLYPVSK	YQQDQVVKED	NIEAVGKKLH	EYNTQFQEK	REYDRLYEY	TRTSQEIOMK	480
481	RTAIEAFNET	IKIFEEQCQT	QERYKEYIE	KFKREGNEKE	IQRIMHNYDK	LKSRISEIID	540
541	SRRLEEDLK	QAAEYREID	KRMNSIKPDL	IQLRKTRDQY	LMWLTQKQV	QKKLNEWLGN	600
601	ENTEDQYSLV	EDDEDLPHHD	EKTWNVGSN	RNKAENLLRG	KRDGTFVRE	SSKQGCYACS	660
661	VVDGGEVVKH	VINKTATGYG	FAEPYNLYSS	LKELVLHYQH	TSLVQHNSL	NVTLAYPVYA	720
721	QQRR						780

blue: PIK3R1 sequence expressed in recombinant protein Red: variant in recombinant protein

<sup>1</sup>NCBI/Protein accession number NP\_852664.1  
E451K: SNP variation see NCBI/dbSNP ID: rs17852841