

## 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

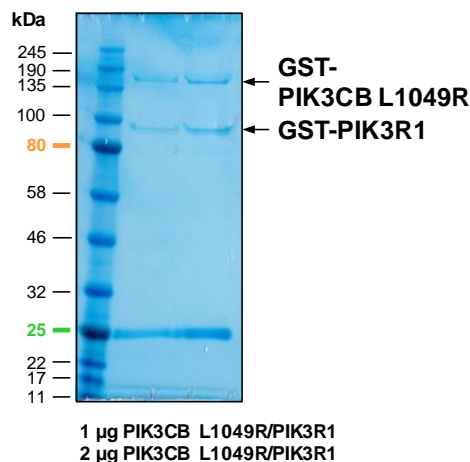
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

**Biochemical Parameters:**

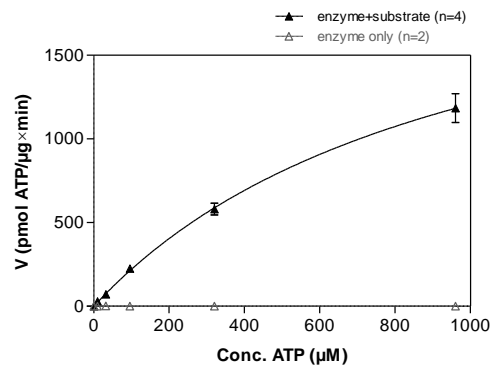
Specific kinase activity (P<sub>i</sub> transfer): 2397 pmol/µg × min

ATP-K<sub>M</sub>: 985 µM

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



**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-Glycerol-3-(Phosphoinositol-4,5-Bisphosphate))
  - PS: 1-Palmitoyl-2-Oleoyl-sn-Glycerol-3-(Phospho-L-Serine)
  - Kinase: 4 µg/ml
- ADP-Glo™ assay (Promega)

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# PIK3CB L1049R/PIK3R1

Product No.: 1585-1165-1

| GST-PIK3CB L1049R Recombinant Fusion Protein Amino Acid Sequence |                    |                   |                   |                         |                   |                    |      |
|--|--------------------|-------------------|-------------------|-------------------------|-------------------|--------------------|------|
| 1  | MSPILGWYKI         | KGLVQPTRL         | LEYLEEKYEE        | HLYERDEGDK              | WRNKKFELGL        | EFPNLPYYID         | 60   |
| 61   | GDVKLTQSM          | IIRYIADKHN        | MLGGCPKERA        | EISMLEGAVL              | DIRYGVSRIA        | YSKDFETLKV         | 120  |
| 121  | DFLSKLP            | KMFKDRLCHK        | TYLNGDHVTH        | PDFMLYDALD              | VVLYMDPMCL        | DAFPKLVCFK         | 180  |
| 181  | KRIEAI             | QID 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>CDPGEKLSK</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>QDVFSFQVS</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>PPKKTRISH</b>  | <b>VWENNNPFQI</b>       | <b>VLVKGNKLN</b>  | <b>EETVKVHVRA</b>  | 600  |
| 601  | <b>GLFHGTSELLC</b> | <b>KTIVSSEVSG</b> | <b>KNDHIWNEPL</b> | <b>EFDINICDLP</b>       | <b>RMARLCFAVY</b> | <b>AVLDKVKTKK</b>  | 660  |
| 661  | <b>STKTINPSKY</b>  | <b>QTIRKAGKVH</b> | <b>YPVAWNTMV</b>  | <b>FDFKQLRTG</b>        | <b>DIILHSSWF</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>MKVLKQVEA</b>   | 960  |
| 961  | <b>LNKLKTLSNL</b>  | <b>IKLNAVKLN</b>  | <b>AKGKEAMHTC</b> | <b>LKQSAAREAL</b>       | <b>SDLQSPNLC</b>  | <b>VILSELYVEK</b>  | 1020 |
| 1021   | <b>CKYMSKMKP</b>   | <b>LWLNVNKKVF</b> | <b>GEDSVGVIFC</b> | <b>NGDDLQDML</b>        | <b>TLQMLRLMDL</b> | <b>LWKEAGLDR</b>   | 1080 |
| 1081   | <b>MLPYGCLATG</b>  | <b>DRSGLIEVVS</b> | <b>TSETIADIQL</b> | <b>NSSNVAASAA</b>       | <b>FNKDALLNL</b>  | <b>KEYNSGDDL</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>TEKFRFRQC</b>  | <b>CEDAYLILRR</b>       | <b>HGNLFITLFA</b> | <b>LMLTAGLPEL</b>  | 1260 |
| 1261   | <b>TSVKDIQYLK</b>  | <b>DSLALGKSEE</b> | <b>EALKQFKQKF</b> | <b>DEA<b>RRES</b>WT</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 | DSQIASDGS  | PVDFLLPTGI  | YIQLEVPREA | TISYIKQMLW  | 60   |
| 61   | KQVHNYPMF  | LLMDIDSYM  | ACVNQTAVY  | ELEDETRRLC  | DVRPFLPVLK | LVTRSCDPGE  | 120  |
| 121  | KLDSKIGVLI | GKGLHEFDSL | KDPEVNEFRR | KMRKFSEEKI  | LSLVGLSWMD | WLKQTYPPEH  | 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  | CFAYAVLKD  | VTKKSKTI    | 420  |
| 421  | NPSKYQTIRK | AGKVHPVAW  | VNTMVDFDK  | QLRTGDIILH  | SWSSFPDELE | EMLNPMGTVQ  | 480  |
| 481  | TNPYTENATA | LHVKFPENKK | QPYYPFDK   | IIEKAAEIAS  | SDSANVSSRG | GKKFLPVLKE  | 540  |
| 541  | ILDRDPLSQL | CENEMDIWT  | LRQDCREIFP | QSLPKLLLSI  | KWNKLEDVAQ | LQALLQIWPK  | 600  |
| 601  | LPPREALELL | DFNYPDQYVR | EYAVGCLRQM | SDEELSQYLL  | QLVQVLKYE  | FLDCALSREL  | 660  |
| 661  | LERALGNRRI | GQFLFWHLRS | EVHIPAVSVQ | FGVILEAYCR  | GSVGHMKVLS | KQVEALNKLK  | 720  |
| 721  | TLNSLIKLNA | VKLNRAKGE  | AMHTCLKQSA | YREALSDLQ   | PLNCPVILSE | LYVECKYMD   | 780  |
| 781  | SKMKPLWL   | VYNNKVFGE  | DSVGVIFKNG | DDLQMLTLQML | RIMDLLWKEA | GLDLRMLPYG  | 840  |
| 841  | CLATGDRSGL | IEVSTSETI  | ADIQLNSSNV | AAAAAFNKDA  | LLNWLKEYNS | GDDLDRATIEE | 900  |
| 901  | FTLSCAGYCV | ASYVLGIGDR | HSDNIMVKK  | TGQLFHIDFGH | ILGNFKSKFG | IKRERVPFIL  | 960  |
| 961  | TYDFIHVIQQ | GKTGNTEKFG | RFRQCCE    | DAYLILRRHGN | LFIITLFA   | LMLTAGLPEL  | 1020 |
| 1021                                       | IQYLKDSLAL | GKSEEEALKQ | FKQKFDEALR | ESWTTKVN    | NWMAHTVR   | KDYRS       | 1080 |

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

<sup>1</sup>[NCBI/Protein](https://www.ncbi.nlm.nih.gov/protein/NP_006210.1) accession number NP\_006210.1

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**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 | PVIPAAYVE  | MISLAPEVQS | SEEYIQLLKK  |            | 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 | EQCQTQERYE | KEYIEKFKRE | GNEKEIQRIM  |            | 540 |
| 541 | HNYDKLKSRI | SEIIDSRRRL | EEDLKKQAAE | YREIDKRMNS | IKPDLIQLRK | TRDQYLMWLT  |            | 600 |
| 601 | QKGVRRQKLE | EWLGNATED  | QYSLVEDDED | LPHHDEKTNW | VGSSNRNKAE | NLLRKRDRGT  |            | 660 |
| 661 | FLVRESSKQG | CYACSVVDG  | EVKHCVINKT | ATGYGFAEPE | NLYSSLKELV | LHYQHTSLVQ  |            | 720 |
| 721 | HNDLNLVTLA | 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  | LDLNPVIPA  | AVYSEMISLA | PEVQSSEEYI | QLLKKLIRSP | SIPHQYWLT   | 240 |
| 241 | QYLLKHFFKL  | SQTSSKNLLN | ARVLSEIFSP | MLFRFSAASS | DNTENLIKVI | EILISTEWNE  | 300 |
| 301 | RQPAPALPPK  | PPKPTTVANN | GMNNMSLQD  | AEWYWGDISR | EEVNEKLRDT | ADGTFVLRDA  | 360 |
| 361 | STKMHGDYTL  | TLRKGGNK   | IKIFHRDGKY | GFSDDLTFSS | VVELINHYRN | ESLAQYNPKL  | 420 |
| 421 | DVKLLYPVSK  | YQQDQVVKED | NIEAVGKKLH | EYNTQFQEK  | REYDRLYEY  | TRTSQEIOMK  | 480 |
| 481 | RTAIEAFNET  | IKIFEEQCQT | QERYKEYIE  | KFKREGNEKE | IQRIMHNYDK | LKSRISEIID  | 540 |
| 541 | SRRLEEDLK   | QAAEYREID  | KRMNSIKPDL | IQLRKTRDQY | LMWLTQKQVR | QKKLNEWLGN  | 600 |
| 601 | ENTEDQYSLV  | EDDEDLPHHD | EKTWNVGSN  | RNKAENLLRG | KRDGTFVRE  | SSKQGCYACS  | 660 |
| 661 | VVDGGEVVKHC | VINKTATGYG | FAEPYNYLSS | 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