

## Certificate of Analysis

### PI3 Kinase (p110 $\alpha$ /p85 $\alpha$ ), active (Recombinant enzyme expressed in Sf21 insect cells)

Item# 14-602, 14-602-K, 14-602M

Parent Lot # WAB0319

The data presented in this document apply to the parent lot shown above and to all pack sizes derived from subsequent vialling runs of this parent lot. An alphabetical suffix after the parent lot number is used to denote each vialling run

**Product Description:** Complex of *N*-terminal 6His-tagged recombinant full-length human p110 $\alpha$  and untagged, recombinant, full length, human p85 $\alpha$ . Coexpressed by baculovirus in Sf21 insect cells. Purified using Ni<sup>2+</sup>/NTA-agarose. Purity 82% by SDS-PAGE and Coomassie blue staining. P110 $\alpha$  MW = 125.3kDa, p85 $\alpha$  MW = 83.7kDa.

**Specific Activity (Parent lot# WAB0319):** 199U/mg, where one unit of PI 3-Kinase alpha (p110 $\alpha$ /p85 $\alpha$ ) (h), active activity is defined as 1nmol phosphatidylinositol 3,4,5-trisphosphate formed per minute at 22°C with a final ATP concentration of 100 $\mu$ M.

**Formulation:** 0.82mg/ml of enzyme in 50mM Tris/HCl pH7.5, 300mM NaCl, 0.1mM EGTA, 0.03% Brij-35, 270mM sucrose, 0.2mM PMSF, 1mM benzamidine, 0.1% 2-mercaptoethanol. Frozen solution.

**Storage and Stability:** On receipt of material store at -70°C. Unopened reagent is stable for a minimum of 1 year from date of shipment when stored at recommended storage temperature. Avoid repeat freeze/thaw cycles. For maximum recovery of product, centrifuge original vial prior to removing the cap.

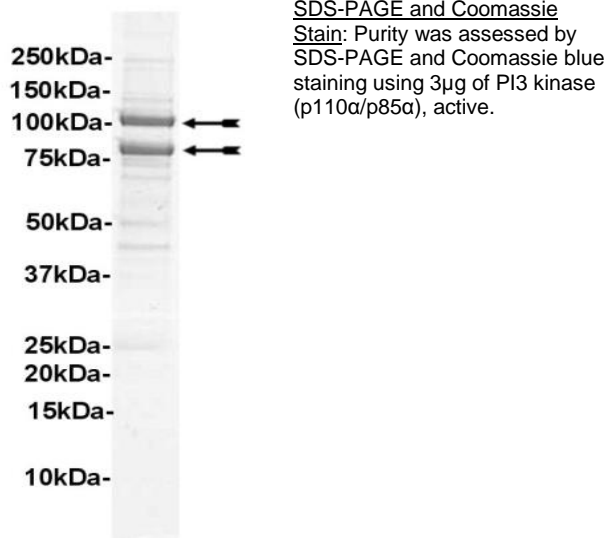
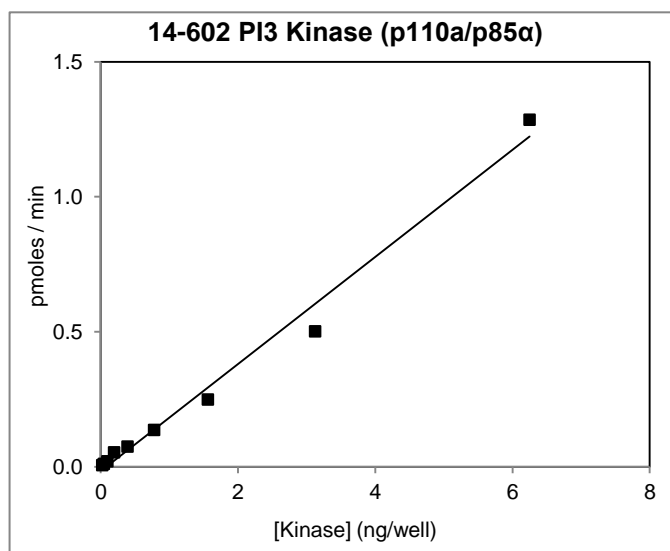
**Handling Recommendations:** Rapidly thaw the vial under cold water and immediately place on ice. Aliquot unused material into pre-chilled micro-centrifuge tubes and immediately snap-freeze the vials in liquid nitrogen prior to re-storage at -70°C.

**FOR IN VITRO RESEARCH USE ONLY  
NOT FOR USE IN HUMANS OR ANIMALS**

### Quality Control Testing

**Kinase Assay:** 0.05–6.25ng of this enzyme phosphorylated 10 $\mu$ M phosphatidylinositol 4, 5-bisphosphate in the assay referenced on page two.

**MS Tryptic Fingerprint:** Confirmed product identity as PI3 Kinase (p110 $\alpha$ /p85 $\alpha$ ) with the translated sequence listed on pages three and five.



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### Eurofins PI3 Kinase Homogeneous Time-resolved Fluorescence (HTRF) Class I Reagent Kits

The following Eurofins kits are suitable for use with this enzyme:

Cat. No	Kit Description
33-016	PI3 Kinase 4-Step Assay Reagent 1-Plate Kit
33-017	PI3 Kinase 4-Step Assay Reagent 5-Plate Kit
33-036	PI3 Kinase 4-Step Assay Reagent Kit (10000 wells)
33-037	PI3 Kinase 4-Step Assay Reagent Kit (50000 wells)
33-040	PI3 Kinase 3-Step Assay Reagent Kit (384 wells)
33-041	PI3 Kinase 3-Step Assay Reagent Kit (1920 wells)
33-047	PI3 Kinase 3-Step Assay Reagent Kit (10000 wells)

Kits 33-016, 33-017, 33-036 and 33-037 provide reagents and assay details for the Eurofins standard 4-step HTRF assay. This assay format is suitable for the majority of small and medium throughput screening work. The 3-step HTRF assay (kits 33-040, 33-041, 33-047) was introduced to reduce the number of assay steps to aid high throughput screening. Items 33-040 and 33-041 are intended as introductory kits for 3-step procedure work up. Please contact us for any further information regarding different kit formats ([discoveryservices@eurofins.com](mailto:discoveryservices@eurofins.com)).

## Certificate of Analysis

### p110 $\alpha$ Sequence Information

<b><u>Protein</u></b>	Human p110 $\alpha$
<b><u>Tags</u></b>	N-terminal 6His
<b><u>Native sequence</u></b>	M8 of the recombinant protein is equivalent to M1 of human p110 $\alpha$
<b><u>Accession number</u></b>	GenBank U79143

#### Recombinant p110 $\alpha$ amino acid sequence:

```

1  MHHHHHMPP  RPSSGELWGI  HLMPPRILVE  CLLPNGMIVT  LECLREATLI  TIKHELFKEA
61  RKYPLHQLLQ  DESSYIFVSV  TQEAEREFF  DETRRLCDLR  LFPFLKVEI  PVGNREEKIL
121 NREIGFAIGM  PVCEFDMVKD  PEVQDFRRNI  LNVCKEAVDL  RDLNSPHSRA  MYVYPPNVES
181 SPELPKHIYN  KLDKGQIIVV  IWVIVSPNND  KQKYTLKINH  DCVPEQVIAE  AIRKKTRSML
241 LSSEQLKLCV  LEYQKGYILK  VCGCDEYFLE  KYPLSQYKYI  RSCIMLGRMP  NLMLMAKESL
301 YSQLPMDCF  MPSYSRRIST  ATPYMNGETS  TKSLWVINS  A  LRIKILCATY  VNVNIRDIDK
361 IYVRTGIYHG  GEPLCDNVNT  QRVPCSNPRW  NEWLNYDIYI  PDLPRAARLC  LSICSVKGRK
421 GAKEEHCPLA  WGINLFDYT  DTLVSGKMAL  NLWPVPHGLE  DLLNPIGVTG  SNPNETPCL
481 ELEFDWFSSV  VKFPDMSVIE  EHANWSVSRE  AGFSYSHAGL  SNRLARDNEL  RENDKEQLKA
541 ISTRDPLSEI  TEQEKDFLWS  HRHYCVTIPE  ILPKLLLSVK  WNSRDEVAQM  YCLVKDWPPI
601 KPEQAMELLD  CNYPDMVRG  FAVRCLEKYL  TDDKLSQYLI  QLVQVLKYEQ  YLDNLLVRF
661 LKKALTNQRI  GHFFFHHLKS  EMHNKTVSQR  FGLLLESYCR  ACGMYLKHLN  RQVEAMEKLI
721 NLTDLKQEK  KDEQKQVQMK  FLVEQMRRPD  FMDALQGF  LS  PLNPAHQLGN  LRLEECRIMS
781 SAKRPLWLNW  ENPDIMSELL  FQNEIIFKN  GDDLQDMLT  LQIIRIMENI  WQNQGLDLRM
841 LPYGCLSIGD  CVGLIEVVRN  SHTIMQIQCK  GGLKGALQFN  SHTLHQWLKD  KNKGEIYDAA
901 IDLFTRSCAG  YCVATFILGI  GDRHNSNIMV  KDDGQLFHID  FGHFLDHKKK  KFGYKRERVP
961 FVLTQDFLIV  ISKGAQECTK  TREFERFQEM  CYKAYLAIRQ  HANLFINLFS  MMLGSGMPEL
1021 QSFDDIAYIR  KTLALDKTEQ  EALEYFMKQM  NDAHGGWTT  KMDWIFHTIK  QHALN

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#### Recombinant p110 $\alpha$ nucleotide sequence:

```

1  atgcatcacc  atccacatca  catgcctcca  agaccatcat  caggtgaact  gtggggcacc
61  cacttgatgc  cccaagaat  cctagtagaa  tgtttactac  caaatggaat  gatagtgact
121  ttagaatgcc  tccgtgaggc  tacattaata  accataaagc  atgaactatt  taaagaagca
181  agaaaatacc  cctccatca  acttctcaaa  gatgaatctt  cttacatttt  cgtaagtgtt
241  actcaagaag  cagaaagga  agaatttttt  gatgaaacaa  gacgactttg  tgaccttcgg
301  ctttttcaac  cttttttaa  agtaattgaa  ccagtaggca  accgtgaaga  aaagatcctc
361  aatcgagaaa  ttggttttgc  tatcggcatg  ccagtgtgtg  aatttgatat  ggttaaagat
421  ccagaagtac  aggacttccg  aagaaatatt  ctgaacgttt  gtaaagaagc  tgtggatcct
481  aggacacctca  attcacctca  tagtagagca  atgtatgtct  atcctccaaa  tgtagaatct
541  tcaccagaat  tgccaaagca  catatataat  aaattagata  aagggcaaat  aatagtggtg
601  atctgggtaa  tagtttctc  aaataatgac  aagcagaagt  atactctgaa  aatcaacct
661  gactgtgtac  cagaacaagt  aattgctgaa  gcaatcagga  aaaaaactcg  aagtatgttg
721  ctatcctctg  aacaactaaa  actctgtgtt  ttagaataatc  agggcaagta  tattttaaaa
781  gtgtgtggat  gtgatgaata  cttcctagaa  aaatattctc  tgagtcagta  taagtatata
841  agaagctgta  taatgcttgg  gaggatgcc  aatttgatgt  tgatggctaa  agaaagcctt
901  tattctcaac  tgccaatgga  ctgttttaca  atgccatctt  attccagacg  catttcaca
961  gctacaccat  atatgaatgg  agaaacatct  acaaaatccc  tttgggttat  aaatagtgca
1021  ctcaagaata  aaattctttg  tgcaacctac  gtgaatgtaa  atattcgaga  cattgataag
1081  atctatgttc  gaacaggat  ctaccatgga  ggagaaccct  tatgtgacaa  tgtgaacact
1141  caaagagtac  cttgttcaa  tcccagggtg  aatgaatggc  tgaattatga  tatatacatt
1201  cctgatcttc  ctcgtctgc  tcgactttgc  ctttccattt  gctctgttaa  aggccgaaag
1261  ggtgctaaag  aggaacactg  tccattggca  tggggaaata  taaacttggt  tgattacaca

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## Certificate of Analysis

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1321 gacactctag tatctggaaa aatggctttg aatctttggc cagtacctca tggattagaa
1381 gatttgctga accctattgg tgttactgga tcaaatccaa ataaagaaac tccatgctta
1441 gagttggagt ttgactgggt cagcagtgtg gtaaagtcc cagatatgtc agtgattgaa
1501 gagcatgcc aattggtctgt atccccgaga gcaggattta gctattccca cgcaggactg
1561 agtaacagac tagctagaga caatgaatta agggaaaatg acaaagaaca gctcaaagca
1621 atttctacac gagatcctct ctctgaaatc actgagcagg agaaagattt tctatggagt
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3181 aaaatggatt ggatcttcca cacaattaa cagcatgcat tgaactga
    
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## Certificate of Analysis

### P85α Sequence Information

<b><u>Protein</u></b>	Human p85α
<b><u>Tags</u></b>	Untagged
<b><u>Native sequence</u></b>	M1 of the recombinant protein is equivalent to M1 of human p85α
<b><u>Accession number</u></b>	GenBank XM_043865

#### Recombinant p85α amino acid sequence:

```

1 MSAEGYQYRA LYDYKKEREE DIDLHLGDIL TVNKGSLVAL GFSDGQEARP EEIGWLNQYN
61 ETTGERGDFP GTYVEYIGRK KISPPTPKPR PPRPLPVAPG SSKTEADVEQ QALTLPLDLAE
121 QFAPPDIAPP LLIKLVEAIE KKGLECSTLY RTQSSSNLAE LRQLLDCDTP SVDLEMIDVH
181 VLADAFKRYL LDLPNPVIPA AVYSEMISLA PEVQSSEEIYI QLLKKLIRSP SIPHQYWLT
241 QYLLKHFFKL SQTSSKNLLN ARVLSEIFSP MLFRFSAASS DNTENLIKVI EILISTEWNE
301 RQPAPALPPK PPKPTTVANN GMNNSLQD AEWYWGDISR EEVNEKLRDT ADGTFLLVRDA
361 STKMHGDYTL TLRKGGNNKL IKIFHRDGKY GFSDDLTFSS VVELINHYRN ESLAQYNPKL
421 DVKLLYPVSK YQQDQVVKED NIEAVGKLLH EYNTQFQEK SREYDRLYEEY TRTSQEIQMK
481 RTAIEAFNET IKIFEEQCQT QERYSKEYIE KFKREGNEKE IQRIMHNYDK LKSRISEIID
541 SRRRLEEDLK KQAAEYREID KRMNSIKPDL IQLRKTRDQY LMWLTQKGVR QKKLNEWLGN
601 ENTEDQYSLV EDDELPHHD EKTWNVGSSN RNKAENLLRG KRDTGFLVRE SSKQGCYACS
661 VVVDGEVKHC VINKTATGYG FAEPNLYSS LKELVLHYQH TSLVQHNSL NVTLAYPVYA
721 QRRR

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#### Recombinant p85α nucleotide sequence:

```

1 atgagtgctg aggggtacca gtacagagcg ctgtatgatt ataaaaagga aagagaagaa
61 gatattgact tgcacttggg tgacatattg actgtgaata aagggtcctt agtagctctt
121 ggattcagtg atggacagga agccaggcct gaagaaattg gctggttaaa tggctataat
181 gaaaccacag gggaaagggg ggactttccg ggaacttacg tagaatatat tggaaagaaa
241 aaaatctcgc ctccacacc aaagccccgg ccacctcggc ctcttcctgt tgcaccaggt
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781 gcaagagtac tctctgaaat tttcagccct atgcttttca gattctcagc agccagctct
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901 cgacagcctg caccagcact gcctcctaaa ccacaaaac ctactactgt agccaacaac
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1261 gatgtgaaat tactttatcc agtatccaaa taccacagg atcaagttgt caaagaagat
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1381 cgagaatatg atagattata tgaagaatat acccgacat cccaggaaat ccaaatgaaa
1441 aggacagcta ttgaagcatt taatgaaacc ataaaaatat ttgaagaaca gtgccagacc
1501 caagagcggg acagcaaaga atacatagaa aagtttaaac gtgaaggcaa tgagaaagaa
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## Certificate of Analysis

```
1621 agtagaagaa gattggaaga agacttgaag aagcaggcag ctgagtatcg agaaattgac
1681 aaacgtatga acagcattaa accagacctt atccagctga gaaagacgag agaccaatac
1741 ttgatgtggt tgactcaaaa aggtgttcgg caaaagaagt tgaacgagtg gttgggcaat
1801 gaaaacactg aagaccaata ttcactgggtg gaagatgatg aagatttgcc ccatcatgat
1861 gagaagacat ggaatgttgg aagcagcaac cgaaacaaag ctgaaaacct gttgagaggg
1921 aagcgagatg gcacttttct tgtccgggag agcagtaaac agggctgcta tgcctgctct
1981 gtagtgggtg acggcgaagt aaagcattgt gtcataaaca aaacagcaac tggctatggc
2041 tttgccgagc cctataactt gtacagctct ctgaaagaac tgggtgctaca ttaccaacac
2101 acctcccttg tgcagcacia cgactccctc aatgtcacac tagcctaccc agtatatgca
2161 cagcagaggc gatga
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Reviewed and approved by site quality representative.

Unless otherwise stated in our catalogue or other company documentation accompanying the product(s), our products are intended for research use only and are not to be used for any other purpose, which includes but is not limited to, unauthorized commercial uses, in vitro diagnostic uses, ex vivo or in vivo therapeutic uses or any type of consumption or application to humans or animals.

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