

Certificate of Analysis

PIP5K1 α , active

(Recombinant enzyme expressed in Sf21 insect cells)

Item # 14-844, 14-844-K, 14-844M

Parent Lot # 1779292

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: N-terminal 6His-tagged, recombinant human PIP5K1 α full length. Expressed by baculovirus in Sf21 insect cells. Purified using immobilized metal affinity chromatography. Purity 73.6% by SDS-PAGE and Coomassie blue staining. MW = 60kDa.

Specific Activity (Parent lot# 1779292): 11971U/mg, where one unit of PIP5K1 α , active activity is defined as 1nmol phosphatidylinositol 4,5-bisphosphate (PIP₂) formed per minute at room temperature with a final ATP concentration of 200 μ M.

Formulation: 0.466mg/ml of enzyme in 50mM Tris/HCl pH8.0, 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.

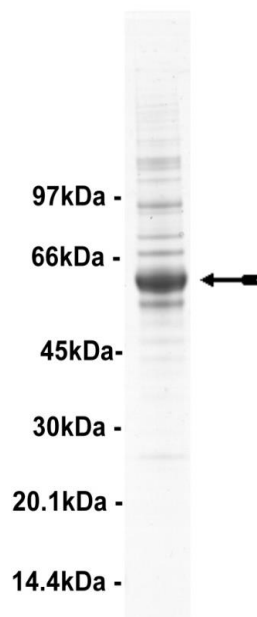
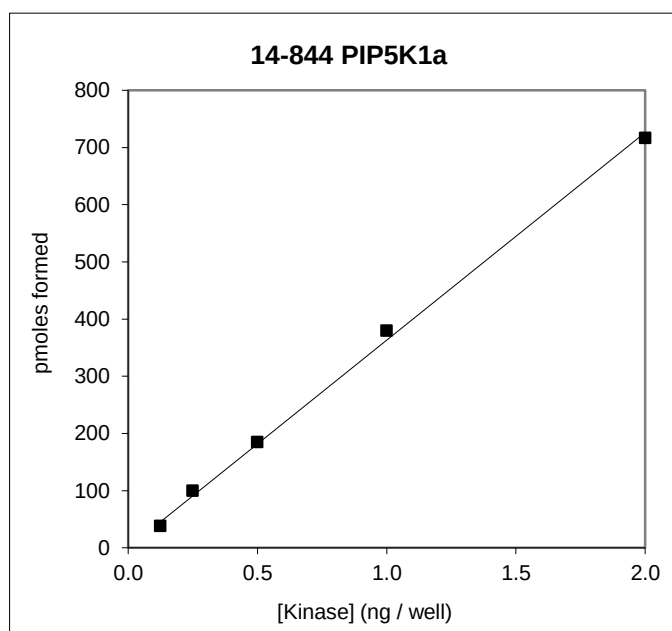
Handling Recommendations: Rapidly thaw the vial under cold water and immediately place on ice. Aliquot unused material into pre-chilled micro-centrifuge tubes and immediately snap-freeze the vials in liquid nitrogen prior to re-storage at -70°C.

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

Quality Control Testing

Kinase Assay: 0.13–2.0ng of this enzyme phosphorylated 25 μ M phosphatidylinositol 5-phosphate (PI5-P) in the assay referenced on page two.

MS Tryptic Fingerprint: Confirmed product identity as PIP5K1 α with the translated sequence listed on page three.



SDS-PAGE and Coomassie Stain: Purity was assessed by SDS-PAGE and Coomassie blue staining using 3 μ g of active PIP5K1 α , active.

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Eurofins PI4 Kinase Homogeneous Time-resolved Fluorescence (HTRF) PIP4-K Reagent Kits

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

Cat. No	Kit Description
33-054	PIP4-Kinase HTRF Assay (384 wells)
33-055	PIP4-Kinase HTRF Assay (1920 wells)

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PIP5K1 α Sequence Information

Protein	Human PIP5K1 α
Tags	N-terminal 6His
Native sequence	M34 of the recombinant protein is equivalent to M1 of PIP5K1 α .
Accession number	GenBank U78577

Recombinant PIP5K1 α amino acid sequence:

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1  MSYYHHHHH  DYDIPTTENL  YFQGAMDFEF  AAKMASASSG  PSSSVGFSSF  DPAVPSC TLS
61  SASGIKR PMA  SEVPYASGMP  IKKIGHRSVD  SSGETTYKKT  TSSALKGAIQ  LGITHTVGSL
121  STKPERD VLM  QDFYVVESIF  FPSEGSNLTP  AHYNDFRFK  TYAPVAFRYF  RELFGIRPDD
181  YLYSLCSEPL  IELCSSGASG  SLFYVSSDDE  FIIKTVQHKE  AEFLQKLLPG  YVMNLNQNP R
241  TLLPKFYGLY  CVQAGGKNIR  IVVMNLLPR  SVKMHIKYDL  KGSTYKRRAS  QKEREKPLPT
301  FKDLDFLQDI  PDGLFLDADM  YNALCKTLQR  DCLVLQSFKI  MDYSLLSIHI  NIDHAQREPL
361  SSETQYSVD T  RRPAPQKALY  STAMESIQGE  ARRGGTMETD  DHMGGIPARN  SKGERLLLYI
421  GIIDILQSYR  FVKKLEHSWK  ALVHDGDTV S  VHRPGFYAER  FQRFMCNTVF  KKIPCVHLGR
481  PDVLPQTPL  EEISEGSPIP  DPSFSPLVGE  TLQMLTTSTT  LEKLEVAESE  FTH

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Recombinant PIP5K1 α nucleotide sequence:

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1  atgtcgtact  accatcacca  tcaccatcac  gattacgata  tcccaacgac  cgaaaacctg
61  tattttcagg  gcgccatgga  tccggaattc  gctgctaaga  tggcgctcggc  ctctccggg
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1561  ttggaaaagc  ttgaagttgc  agagtcagag  ttcaccatt  aa

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Reviewed and approved by site quality representative.

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