

Certificate of Analysis

PIP4K2 α , active

(Recombinant enzyme expressed in Sf21 insect cells)

Item # 14-901, 14-901-K, 14-901M

Parent Lot # D8KN057N

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 PIP4K2 α full length. Expressed by baculovirus in Sf21 insect cells. Purified using Ni²⁺/NTA agarose.

Purity 96% by SDS-PAGE and Coomassie blue staining. MW = 51kDa.

Specific Activity (Parent lot# D8KN057N): 1354U/mg, where one unit of PIP4K2 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: 2.13mg/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.

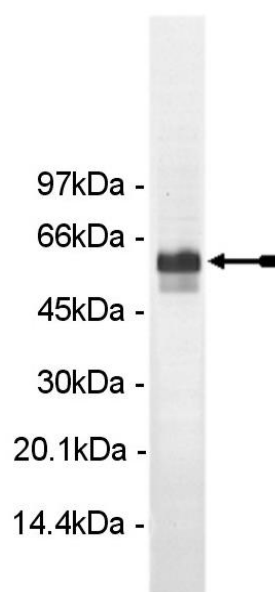
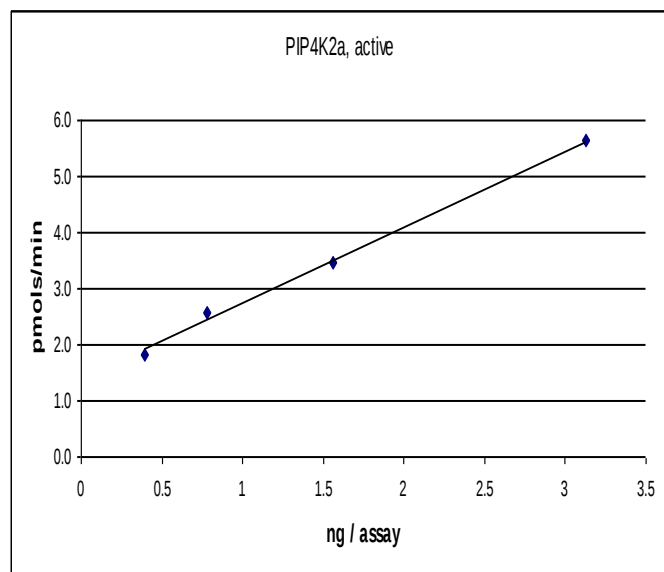
Handling Recommendations: Rapidly thaw the vial under cold water and immediately place on ice. Aliquot unused material into pre-chilled micro-centrifuge tubes and immediately snap-freeze the vials in liquid nitrogen prior to re-storage at -70°C.

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

Quality Control Testing

Kinase Assay: 0.4–3.2ng of this enzyme phosphorylated 25 μ M phosphatidylinositol 5-phosphate (PI5-P) in the assay referenced on page two.

MS Tryptic Fingerprint: Confirmed identity as PIP4K2 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 PIP4K2 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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PIP4K2 α Sequence Information

<u>Protein</u>	Human PIP4K2 α
<u>Tags</u>	N-terminal 6His
<u>Native sequence</u>	M41 of the recombinant protein is equivalent to M1 of PIP4K2 α
<u>Accession number</u>	GenBank NM_005028

Recombinant PIP4K2 α amino acid sequence:

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1 MSYYHHHHHH DYDIPTTENL YFQGAMDPEF KGLRRRAQLV MATPGNLGSS VLASKTKTKK
61 KHFVAQKVKL FRASDPLLSV LMWGVNHSIN ELSHVQIPVM LMPDDFKAYS KIKVDNHLFN
121 KENMPSHFKF KEYCPMVFRN LRERFGIDDQ DFQNSLTRSA PLPND SQARS GARFHTSYDK
181 RYIIKITITSE DVAEMHNILK KYHQYIVECH GITLLPQFLG MYRLNVDGVE IYVIVTRNVF
241 SHRLSVYRKY DLKGSTVARE ASDKEKAKEL PTLKDNDFIN EGQKIYIDDN NKKVFLEKLE
301 KDVEFLAQLK LMDYSLLVGI HDVERAEQEE VECEENDGEE EGESDGTHPV GTPPDSPGNT
361 LNSSPPLAPG EFDPNIDVYG IKCHENS PRK EVYFMAIID LTHYDAKKKA AHAAKTVKHG
421 AGAEISTVNP EQYSKRFLDF IGHILT

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Recombinant PIP4K2 α nucleotide sequence:

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1 atgtcgtact accatcacca tcaccatcac gattacgata tcccaacgac cgaaaacctg
61 tattttcagg gcgccatgga tccggaattc aaaggcctac gtcgacgagc tcaactagtg
121 atggcgaccc ccggcaacct agggctcctc gtcctggcga gcaagaccaa gaccaagaag
181 aagcacttcg tagcgagaaa agtgaagctg tttcgggcca gcgacccgct gctcagcgct
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1321 attggccaca tcttgacgta a

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Reviewed and approved by site quality representative.

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