



**User's Manual**

# **PI(4)P 5-Kinase (PIP5KI) Activity Assay Kit**



**DEIA-XYZ15**



**96T**



This product is for research use only and is not intended for diagnostic use.

For illustrative purposes only. To perform the assay the instructions for use provided with the kit have to be used.

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## PRODUCT INFORMATION

### Intended Use

PI(4)P 5-Kinase (PIP5KI) Activity Assay Kit is an ATP depletion assay which quantifies the remaining ATP levels in solution following the kinase reaction. The assay can be used to quantify PI(4)P 5-Kinase in vitro or in cell lysate.

### General Description

Type I PI(4)P 5-Kinase (PIP5KI) catalyzes the phosphorylation of phosphatidylinositol-4-phosphate [PI(4)P] to phosphatidylinositol-4,5-bisphosphate [PI(4,5)P<sub>2</sub>]. PIP5KI has been shown to play a role in several cellular functions including gene transcription, actin cytoskeleton reorganization, secretion and endocytosis. It is thought that PIP5KI activity is responsible for the majority of the PI(4,5)P<sub>2</sub> found in the cell which is an important second messenger for many cellular processes. The assay reaction is prepared by the addition of PIP5KI (enzyme), PI(4)P (substrate) in reaction buffer and initiated by the addition of ATP. The reaction is then stopped by adding the ATP detector after the chosen reaction time. The luminescent signal is inversely correlated with the kinase activity. PI(4)P 5-Kinase (PIP5KI) Activity Assay ELISA Kit is tested using SignalChem's PIP5KIA (P16-10AG) kinase.

### Reagents And Materials Provided

1. PI(4)P Substrate (522 µmol): 1 vial
2. Kinase Reaction Buffer (10 mL): 1 bottle
3. Diluent (150 µL): 1 vial
4. Kinase Cofactor (100 µL): 1 vial
5. ATP Detector (5 mL): 1 bottle
6. 10 mM ATP (50 µL): 1 vial
7. 50 µmol dried dithiothreitol (DTT) powder: 1 vial
8. 96-well round bottom white plate: 1 each
9. Microtiter Plate Seal: 1 each

### Materials Required But Not Supplied

1. Source of purified PI(4)P 5-Kinase as positive control (optional)
2. Plate reader capable of reading luminescence in 96-well microtiter plates
3. Microcentrifuge tubes (0.5 mL or 1.5 mL) or reservoir for standard dilution or sample loading Multichannel pipettes or automatic
4. pipetting station

### Storage

2-8°C and -20°C