



Pim-1 Positive Control

Product Data Sheet

For Research Use Only, Not for use in diagnostic procedures

## Pim-1 Positive Control

(Human, full length, recombinant enzyme expressed in *E. coli*.)

Cat# CY-E1167

Lot No.  
For 200 Assays  
(0.02 units /  $\mu\text{L}$  x 100  $\mu\text{L}$ )

**10X BSA (100  $\mu\text{g}/\text{mL}$  x 0.25 mL) is supplied to make an enzyme dilution buffer**

**Product Description:** Human full length Pim-1, containing a *N*-terminal GST tag, expressed in *E. coli*. Purified by GSH agarose chromatography. The Pim-1 Positive control is designed to use for Pim-1 Assay/Inhibitor Screening Kit [Cat# CY-1167]. The Pim-1 Positive Control should be added to the well at 10 m units/well. For instance, diluted positive control 1:20, use 10  $\mu\text{L}$  for 1 assay. Unused Pim-1 Positive control should be stored at  $-70^{\circ}\text{C}$ .

**Product Size:** Full length Pim-1: 2 units/100  $\mu\text{L}$

**Formulation:** The Pim-1 Positive Control is supplied frozen in a buffer containing 20mM Hepes-KOH (pH 7.5), 1 % BSA, 1mM EDTA, 1 mM DTT, 50mM NaCl, 0.03 % Brij35 and 50% glycerol.

**Dilution:** The Pim-1 Positive control should be diluted with an enzyme dilution buffer to avoid inactivating the enzyme activity in low protein concentration condition. Enzyme dilution buffer: Mix 9-parts of Kinase buffer and 1-part of 10X BSA (100  $\mu\text{g}/\text{mL}$ )

**Source:** Human full length Pim-1, containing *N*-terminal GST tag, expressed in *E. coli*.

**Molecular Weight:** Pim-1 demonstrates a single 60 kDa band by SDS-PAGE analysis.

**Purity:** Pim-1 is greater than 90 % pure as determined by SDS-PAGE analysis.

**Substrates:** Pim-1 phosphorylates a number of substrates, including p21waf1, NuMA, HP1beta and C-TAK1.

**Inhibitors:** Specific Pim-1 inhibitor has not been discovered yet.

**Unit Definitions:** One unit is defined as the amount of kinase required to incorporate 1 nmol of phosphate into MBP per minute at  $30^{\circ}\text{C}$ .

**Assay Conditions:** Assay activity of Pim-1 in a 50  $\mu\text{L}$  reaction containing 20 mM Hepes KOH (pH 6.5), 5 mM  $\text{MgCl}_2$ , 1 mM DTT, 100  $\mu\text{M}$  [ $\gamma$ - $^{32}\text{P}$ ] ATP (1  $\mu\text{Ci}$ ), and 4  $\mu\text{g}$  of GST-p21waf1. Start the reaction by adding 10 $\mu\text{L}$  of the enzyme, diluted 50-fold in a buffer containing 20 mM Hepes KOH (pH 6.5), 1 mM DTT, 0.03 % Brij35. Incubate for 30 minutes at  $30^{\circ}\text{C}$ . Terminate the reaction by adding 600  $\mu\text{L}$  of cold 10 % TCA solution containing 0.2 % sodium pyrophosphate and stand on ice for 15 min. Filtrate acid insoluble material through GFC filters (Whatman Inc.), wash 4 times with 1 % TCA and rinse filters with ethanol. Dry filters and count in a liquid scintillation counter.



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**Storage and Stability:** Stable for 12 months at  $-70^{\circ}\text{C}$  from date of shipment. For maximum recovery of product, centrifuge the original vial after thawing and prior to removing the cap. Aliquot enzyme to avoid repeated freezing and thawing.

#### **Related Products:**

\* Pim-1 Kinase Assay/Inhibitor Screening Kit: Cat# CY-1167

#### **General References:**

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