



Plk1 Positive Control

Product Data Sheet

For Research Use Only, Not for use in diagnostic procedures



## Plk1 Positive Control

(Human, full length, recombinant enzyme expressed in *E. coli*)  
Cat# CY-E1163

Lot No.  
For 200 Assays  
20 m units (0.2 m units/ $\mu$ L)

**Product Description:** Constitutive active form of human Plk1, in which there is one mutation T210D in an active loop of kinase domain, containing an N-terminal GST tag, is expressed in *E. coli*. and purified by GSH agarose chromatography. The Plk1 Positive Control is designed to use for Plk1 Assay/Inhibitor Screening Kit (Cat# CY-1163). The Plk1 Positive Control should be added to the well at 0.1 m units/well. Unused Plk1 Positive Control should be stored at -70°C.

**Product Size:** 20 m units/100  $\mu$ L

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

**Source:** Human Plk1, full length, T210D, containing an N-terminal GST tag, expressed in *E. coli*.

**Molecular Weight:** 92 kDa band by SDS-PAGE analysis.

**Purity:** > 85 % pure as determined by SDS-PAGE analysis.

**Substrates:** Plk1 phosphorylates a number of substrates, including Cyclin B, MBP.

**Inhibitors:** Specific Plk1 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 alpha-casein per minute at 30°C.

**Assay Conditions:** Assay activity of Plk1 in a 50  $\mu$ L reaction containing 20 mM Hepes KOH (pH 7.5), 5 mM MgCl<sub>2</sub>, 1 mM DTT, 100  $\mu$ M [ $\gamma$ -<sup>32</sup>P] ATP (1  $\mu$ Ci), and 4  $\mu$ g of alpha-casein. Start the reaction by adding 10 $\mu$ L of the enzyme, diluted 50-fold in a buffer containing 20 mM Hepes KOH (pH 7.5), 1 mM DTT, 0.03 % Brij35. Incubate for 30 minutes at 30°C. Terminate the reaction by adding 600  $\mu$ 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.

**Storage and Stability:** Stable for 12 months at -70°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.



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**References:**

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