



CaM kinase II Positive Control

Product Data Sheet

For Research Use Only, Not for use in diagnostic procedures



CaM kinase II Positive Control

(Human, a.a.1-328, recombinant protein expressed in Sf9 cells)
Cat# CY-E1173

Lot No.
For 200 Assays
3 units (30 m units/ μ L)

Supplied with 100X Calmodulin (25 μ g/mL x 0.2 mL).

Product Description: Human Ca²⁺/calmodulin-dependent protein kinase II (CaM kinase II), containing an N-terminal GST tag and a C-terminal His tag, is expressed in recombinant baculovirus infected Sf9 cells and purified by sequentially using GSH agarose and Ni-NTA agarose chromatography. The CaM kinase II Positive Control is designed to use for CycLex CaM kinase II kinase Assay Kit (Cat# CY-1173). The CaM kinase II Positive Control should be added to the well at 15 m units/well. Unused CaM kinase II Positive Control should be stored at -70°C.

Product Size: 3 units/100 μ 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 Ca²⁺/calmodulin-dependent protein kinase II, 1-328, containing an N-terminal GST tag and a C-terminal His tag, expressed in Sf9 cells.

Molecular Weight: 62 kDa doublet bands by SDS-PAGE analysis.

Purity: > 90 % pure as determined by SDS-PAGE analysis.

Substrates: CaM kinase II phosphorylates a number of substrates, including synthetic peptides.

Inhibitors: Selective CaM kinase II inhibitor has not been discovered yet.

Unit Definition: One unit is defined as the amount of kinase required to incorporate 1nmol of phosphate into the syntide-2 per minute at 30°C.

Assay Conditions: Assay activity of CaM kinase II in a 50 μ L reaction containing 20 mM Hepes KOH (pH 7.5), 0.1 mM EGTA, 5 mM MgCl₂, 200 ng calmodulin, 2 mM CaCl₂, 1 mM DTT, 100 μ M [γ -³²P] ATP (1 μ Ci), and 4 μ g of syntide-2. Start the reaction by adding 10 μ L of the enzyme, diluted 5-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 spotting to phosphocellulose P81 paper, and then wash 4 times with 75 mM H₃PO₄ for 15 min and rinse P81 paper 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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