p56Lck Positive Control
(Human, recombinant protein expressed in Sf9)
Cat# CY-E1084

Lot No.
For 100 Assays
(1 unit / µL x 100 µL)

Product Description:
Catalytic domain of human Lck, corresponding to 217-509 a.a. containing a *N*-terminal GST tag and a C-terminal His tag, expressed in recombinant Baculovirus-infected sf9 cells. Purified by sequentially using GSH agarose and Ni-NTA agarose chromatography. The Lck Positive control is designed to use for CycLex Lck Kinase Assay/Inhibitor Screening Kit (Cat# CY-1084). The Lck Positive Control should be added to the well at 1 unit/well. For instance, diluted positive control 1:10, use 10 µL for 1 assay. Unused Lck Positive Control should be stored at -70°C.

Product Size: Recombinant Lck: 100 units/100 µL

Formulation: The Lck Positive Control is supplied frozen in a buffer containing 20mM Hepes-KOH (pH 7.5), 1 % BSA, 1mM EDTA, 2 mM DTT, 50mM NaCl, 0.03 % Brij35 and 50% glycerol.

Source: Human Lck containing N-terminal GST-tag and C-terminal His tag, expressed in sf9 cells.

Molecular Weight: Lck Positive Control demonstrates a single 62 kDa bands by SDS-PAGE analysis.

Purity: Lck Positive Control is greater than 75 % pure as determined by SDS-PAGE analysis.

Substrates: Lck phosphorylates poly[Glu, Tyr] 4:1 as a exogenous substrate.

Inhibitors: BMS-238497 and BMS-243117 are known as selective small molecule Lck inhibitor.

Unit Definition: One unit is defined as the amount of kinase required to incorporate 1 nmol of phosphate into the Lck (autophosphorylation) under oligomerized and activated condition per 60 minute at 30°C.

Assay Conditions: Assay activity of Lck in a 50 µL reaction containing 20 mM Hepes KOH (pH 7.5), 4 mM $MgCl_2$, 2 mM $MnCl_2$, 1 mM DTT, 50 µM [gamma ^{32}P] ATP (1 µCi), and 4 µg of CycLex-"Tyrosine kinase-binding module". Start the reaction by adding 10µL of the enzyme, diluted 10-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 µL of cold 10 % TCA solution containing 0.2 % Sodium pyrophosphate and stand on ice for 15 min. Filtrate acid insoluble material through GFC filters (Whattman 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.

Related Products:
* CycLex Lck Kinase Assay/Inhibitor Screening Kit: Cat# CY-1084
* CycLex Tyrosine Kinase Assay Kit Module-1: Cat# CY-2080

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