EphA2 Positive Control
(Human, recombinant protein expressed in Sf9)
Cat# CY-E1092

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

Product Description:
Catalytic domain of human EphA2, corresponding to 613-871 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 EphA2 Positive control is designed to use for CycLex EphA2 Kinase Assay/Inhibitor Screening Kit (Cat# CY-1092). The EphA2 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 EphA2 Positive Control should be stored at -70°C.

Product Size: Recombinant EphA2: 100 units/100 µL

Formulation: The EphA2 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 EphA2 containing N-terminal GST-tag and C-terminal His tag, expressed in sf9 cells.

Molecular Weight: EphA2 Positive Control demonstrates a single 62 kDa bands by SDS-PAGE analysis.

Purity: EphA2 Positive Control is greater than 85 % pure as determined by SDS-PAGE analysis.


Inhibitors: Specific EphA2 inhibitor has not been discovered yet.

Unit Definition: One unit is defined as the amount of kinase required to incorporate 1 nmol of phosphate into the EphA2 (autophosphorylation) under oligomerized/activated condition per 60 minute at 30°C.

Assay Conditions: Assay activity of EphA2 in a 50 µL reaction containing 20 mM Hepes KOH (pH 7.5), 4 mM MgCl₂, 2 mM MnCl₂, 1 mM DTT, 50 µM [gamma ³²P] 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 60 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. Filter acid insoluble material through GF/C 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.
Related Products:
* CycLex EphA2 Kinase Assay/Inhibitor Screening Kit: Cat# CY-1092

References:

PRODUCED BY

CycLex Co., Ltd.
1063-103 Terasawaoaka
Ina, Nagano 396-0002
Japan
Fax: +81-265-76-7618
e-mail: info@cyclex.co.jp
URL: http://www.cyclex.co.jp

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