IKKβ (IκB Kinase β) Positive Control

(Human, full length, recombinant enzyme expressed in Sf9)

Cat# CY-E1178-2

Lot No.
For 200 Assays
(10 mUnits / µL x 200 µL)

Product Description: Human full length IKKβ, containing a *N*-terminal GST tag and a C-terminal His tag, expressed in *Sf9*. Purified by GSH-agarose chromatography. The IKKβ Positive control is designed to use for IKK α/β assay/Inhibitor Screening Kit [Cat# CY-1178]. The IKKβ Positive Control should be added to the well at 10 m units/well. Unused IKKβ Positive control should be stored at -70°C.

Product Size: Full length IKKβ: 2 units/200 µL

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

Source: Human full length IKKβ, containing *N*-terminal GST tag and a C-terminal His tag, expressed in *Sf9*.

Molecular Weight: IKKβ demonstrates a single 105 kDa band by SDS-PAGE analysis.

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

Substrates: IKKβ phosphorylates IκBα at Ser32 and Ser36.

Inhibitors: BMS-345541, SC-514 and [5-(p-Fluorophenyl)-2-ureido]thiophene-3-carboxamide are known as a specific inhibitor for IKKβ.

Unit Definitions: One unit is defined as the amount of kinase required to incorporate 1 nmol of phosphate into the IκBα-S32/S36 peptide; KKKERLLDDRHDSGLDSMKDEEYE, per minute at 30°C.

Assay Conditions: Assay activity of IKKβ in a 50 µL reaction containing 20 mM Hepes KOH (pH 7.5), 5 mM $MgCl_2$, 1 mM DTT, 100 µM [gamma ^{32}P] ATP (1 µCi), and 4 µg of IκBα-S32/S36 peptide. Start the reaction by adding 10µL 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. The reaction is terminated by "spotting" a sample onto a phosphocellulose P81 filter paper disc, followed by washing extensively with 70 mM phosphoric acid to remove unincorporated radiolabel. Subsequently rinse the filters with ethanol and dry. The incorporated radioactivity on P81 filter is counted 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:
* IKKα and β Assay/Inhibitor Screening Kit: Cat# CY-1178

For Reference Purpose Only!

General References:

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