MST1 (Human), Active

Full-length recombinant protein expressed in Sf9 cells

Cat# CY-SPS25

Lot No. P048-1
5 µg 0.1 µg/µl

Background:

MST1 belongs to a family of proteins that share similarity with a budding yeast serine/threonine kinase, sterile-20 (Ste20). Endogenous full-length MST1 is activated by a variety of stressful stimuli, accompanied by the secondary appearance of a 36 kDa Thr183-phosphorylated, caspase-cleaved catalytic fragment (1). Recombinant MST1 undergoes a robust autoactivation in vitro, mediated by an intramolecular autophosphorylation on the activation loop of an MST dimer. MST1 can initiate apoptosis when transiently overexpressed in mammalian cells. Interference with the ability of endogenous MST1 to associate with the putative tumor suppressor proteins Nore1/RASSF can inhibit Ras-induced apoptosis (2).

Product Description:

Recombinant full-length human MST1 was expressed by baculovirus in Sf9 cells using an N-terminal GST tag. The gene accession number is NM_006282.

Gene Aliases:

STK4, KRS2, YSK3, DKFZp686A2068

Formulation:

Recombinant protein stored in 50mM Tris-HCl, pH 7.5, 150mM NaCl, 0.25mM DTT, 0.1mM EGTA, 0.1mM EDTA, 0.1mM PMSF, 25% glycerol.

Purity & Molecular Weight:

The purity was determined to be >85% by densitometry. Approx. MW 83kDa.

Storage:

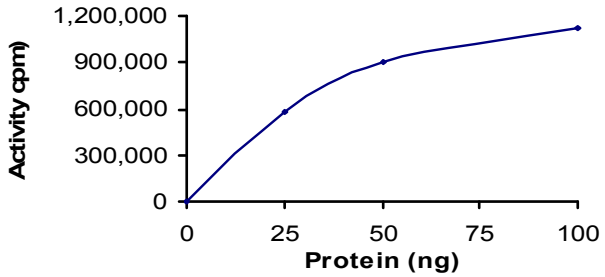
Store product at –70 °C. For optimal storage, aliquot target into smaller quantities after centrifugation and store at recommended temperature. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles.

Stability:

1 year at –70 °C from date of shipment.

Specific Activity:

The specific activity was determined to be 1120 nmol /min/mg as per Activity Assay Protocol.



Activity Assay Protocol:

Assay activity of the kinase in a 25 µL reaction consisting of 5 µL of 5 X Kinase Assay Buffer, 10 µL of 1 mg/ml the Substrate Solution, 5 µL of diluted kinase and 5 µL of 250 µM ATP solution containing [gamma ^{32}P] ATP (0.167 µCi/µL). Start the reaction by adding the ATP solution. Incubate for 15 minutes at 30°C. Terminate the reaction by spotting 20 µL of the reaction mixture onto phosphocellulose P81 paper. Air-dry the P81 paper and sequentially wash 4 times for approximately 10 minutes each in 1% phosphoric acid with constant gentle stirring. Count the P81 paper in a liquid scintillation counter.

Substrate Solution:

Axltide (KKSRGDYMTMQIG) diluted in distilled H_2O to a final concentration of 1mg/ml.

5 X Kinase Assay Buffer:

25mM MOPS pH 7.2, 12.5mM β-glycerol-phosphate, 25mM $MgCl_2$, 5mM EGTA, 2mM EDTA. Add 0.25mM DTT to Kinase Assay Buffer prior to use.

References:

1.De Souza, P M. et al: Mammalian Sterile20-like kinase 1 and the regulation of apoptosis. Biochem Soc Trans. 2004 Jun;32(Pt3):485-8.

2.Avruch, J. et al: Nore1 and RASSF1 Regulation of Cell Proliferation and of the MST1/2 Kinases. Methods Enzymol. 2005;407:290-310.

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