p38 alpha (Human), Active

Full-length recombinant protein expressed in Sf9 cells

Cat# CY-SPM39

Lot No. K193-1
5 µg 0.1 µg/µl

Background:

p38 α (p38-alpha, SAPK2A) is a member of the p38 MAPK family which are activated by various environmental stresses and proinflammatory cytokines (1). The activation of p38 requires its phosphorylation by MAP kinase kinases (MKKs), or its autophosphorylation triggered by the interaction of MAP3K7IP1/TAB1 protein with this kinase (2). The substrates of p38 include transcription regulator ATF2, MEF2C, and MAX, cell cycle regulator CDC25B, and tumor suppressor p53, which suggest the roles of this kinase in stress related transcription and cell cycle regulation, as well as in genotoxic stress response (5).

Product Description:

Recombinant full-length human p38 α was expressed by baculovirus in Sf9 insect cells using a N-terminal GST tag. The gene accession number is NM_139012.

Gene Aliases:

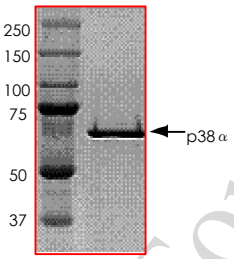
CSBP1;CSBP2;CSPB1;PRKM14;PRKM15;SAPK2A;MAPK14

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 >90% by densitometry. Approx. MW 67kDa.



Storage:

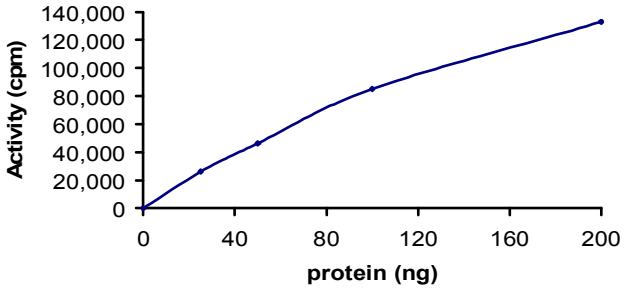
Store product at –70 °C. For optimal storage, aliquot target into smaller quantities after centrifugation and store at recommended temperature. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles.

Stability:

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

Specific Activity:

The specific activity was determined to be 77 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 0.5 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:

N-terminal GST tagged recombinant human ATF2 (1-254) prepared in buffer (50mM Tris-HCl, pH 7. 2, 50mM $NaC1_2$, 5mM EDTA and 0.25mM DTT) to a final concentration of 0.5 mg/ml.

5 X Kinase Assay Buffer:

25mM MOPS, 12.5mM β-glycerol-phosphate, 25mM $MgC1_2$, 5mM EGTA, 2mM EDTA. Add 0.25mM DTT to Kinase Assay Buffer prior to use.

References:

1.Han, J. et al: A MAP kinase targeted by endotoxin and hyperosmolarity in mammalian cells. Science 265: 808-811, 1994.

2.Ge, B. et al: MAPKK-independent activation of p38-alpha mediated by TAB1-dependent autophosphorylation of p38-alpha. Science 295: 1291-1294, 2002.

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