

CSK (Human), Active, Tag-free

Full-length recombinant protein expressed in Sf9 cells

Cat# CY-SPC63-U

Lot No. K137-1
5 µg 0.1 µg/µl

Background:

CSK is a cytoplasmic tyrosine kinase that has been shown to downregulate the tyrosine kinase activity of the c-src through tyrosine phosphorylation of the c-src carboxy terminus (1). A yeast 2-hybrid system has been used to identify proteins associated with CSK. The Src homology-3 (SH3) domain of CSK associates with a proline-rich region of PEP, a protein-tyrosine phosphatase expressed in hemopoietic cells (2). This association is highly specific and it is speculated that PEP may be an effector and/or regulator of CSK in T cells and other hemopoietic cells.

Product Description:

Recombinant full-length tag-free human CSK was expressed by baculovirus in Sf9 insect cells. The gene accession number is NM_004383.

Gene Aliases:

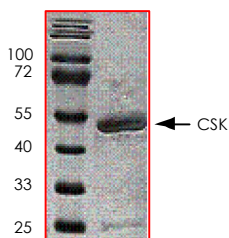
None

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 78kDa.



Storage:

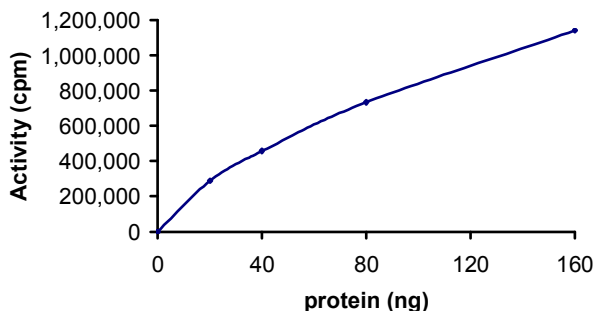
Store product at -70°C . For optimal storage, aliquot target into smaller quantities after centrifugation and store at recommended temperature. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles.

Stability:

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

**Specific Activity:**

The specific activity was determined to be 425 nmol/min/mg as per Activity Assay Protocol.

**Activity Assay Protocol:**

Assay activity of the kinase in a 25 μ L reaction consisting of 5 μ L of 5 X Kinase Assay Buffer, 10 μ L of 1 mg/ml the Substrate Solution, 5 μ L of diluted kinase and 5 μ L of 250 μ M ATP solution containing [γ - 32 P] ATP (0.167 μ Ci/ μ L). Start the reaction by adding the ATP solution. Incubate for 15 minutes at 30°C. Terminate the reaction by spotting 20 μ L 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:

Poly (Glu:Tyr, 4:1) synthetic peptide substrate diluted in distilled H₂O to a final concentration of 1 mg/ml.

5 X Kinase Assay Buffer:

25mM MOPS, 12.5mM β -glycerol-phosphate, 20mM MgCl₂, 25mM MnCl₂, 5mM EGTA, 2mM EDTA. Add 0.25mM DTT to Kinase Assay Buffer prior to use.

References:

1. Partanen, J. et al: Cyl encodes a putative cytoplasmic tyrosine kinase lacking the conserved tyrosine autophosphorylation site (Y416-src). *Oncogene* 6: 2013-2018, 1991.
2. Cloutier, J.-F. et al: Association of inhibitory tyrosine protein kinase p50(csk) with protein tyrosine phosphatase PEP in T cells and other hemopoietic cells. *EMBO J.* 15: 4909-4918, 1996.

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