BLK (Human), Active

Full-length recombinant protein expressed in Sf9 cells

Cat# CY-SPB02

Lot No. B242-2
5 µg 0.1 µg/µl

Background:

BLK, also known as B lymphoid kinase, is a 55 kd tyrosine kinase with a SH3, SH2 and catalytic domain that contain consensus sequences of the src protein tyrosine kinase family. BLK is expressed specifically in the B cell lineage and plays a role in signal transduction pathway that is restricted to B lymphoid cells (1). Stimulation of resting B-lymphocytes with antibodies to surface immunoglobulin (sIgD or sIgM) induces activation of BLK (2)

Product Description:

Recombinant full-length human BLK was expressed by baculovirus in Sf9 insect cells using an N-terminal GST tag. The gene accession number is BC007371.

Gene Aliases:

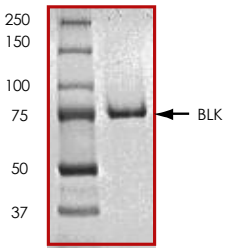
MGC10442

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 84kDa.



Storage:

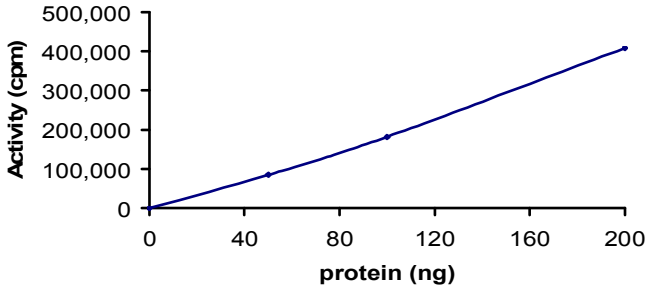
Store product at –70 °C. For optimal storage, aliquot target into smaller quantities after centrifugation and store at recommended temperature. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles.

Stability:

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

Specific Activity:

The specific activity was determined to be 112 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:

Poly (Glu:Tyr, 4:1) peptide was diluted in distilled H_2O to a final concentration of 1 mg/ml.

5 X Kinase Assay Buffer:

25mM MOPS, 12.5mM β-glycerol-phosphate, 20mM MgC12, 25mM $MnC1_2$, 5mM EGTA, 2mM EDTA. Add 0.25mM DTT to Kinase Assay Buffer prior to use

References:

1.Dymecki, SM. et al: Specific expression of a tyrosine kinase gene, blk, in B lymphoid cells. Science. 1990 Jan 19;247(4940):332-6.

2.Burkhardt, AL. et al: Anti-immunoglobulin stimulation of B lymphocytes activates src-related protein-tyrosine kinases. Proc Natl Acad Sci U S A. 1991 Aug 15;88(16):7410-4.

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