

Product Information

Description:	Recombinant human ALK (F1174S) (anaplastic lymphoma kinase), encompassing amino acids 1060-end with a F1174S mutation. This construct contains an N-terminal GST-tag. The recombinant protein was affinity purified and is active.
Background:	ALK (anaplastic lymphoma kinase), also known as ALK tyrosine kinase receptor or CD246 (cluster of differentiation 246), is a receptor tyrosine kinase involved in signal transduction. In the presence of a ligand ALK dimerizes and a conformational change result in autoactivation of the kinase domain. Activated ALK will phosphorylate other ALK receptors and activate downstream signaling pathways. ALK is present in the nervous system during development, where it participates in retinal axon growth and targeting, synapse development, sleep, learning and long-term memory. Interestingly, dysfunction of ALK in one of three possible ways can lead to cancer: fusion with another gene, gene duplication or gene mutations. ALK, as its name indicates, has been linked to anaplastic large-cell lymphoma, but also non-small-cell lung cancer (NSCLC), neuroblastoma, breast cancer, renal carcinoma and others. Inhibitors of ALK show great therapeutical potential, two of them being already commercially available for the treatment of late-stage lung cancer and NSCLC. Further studies into ALK will deepen our understanding of its functions, find new inhibitors and new therapeutic avenues for patients with ALK-linked cancer.
Species:	Human
Construct:	ALK (F1174S) (GST-1060-end)
Mutation:	F1174S
Concentration:	0.10 mg/ml
Expression System:	Sf9
Purity:	70% (Purity calculation does not include co-purifying Glutathione-binding proteins.)
Format:	Aqueous buffer solution.
Formulated In:	50 mM Sodium Phosphate, pH 7.0, 300 mM NaCl, 150 mM Imidazole, 0.1 mM PMSF, 0.25 mM DTT, 25% glycerol
MW:	90 kDa
Genbank Accession:	NM_004304
Stability:	At least 6 months at -80°C.
Storage:	-80°C
Instructions for Use:	Thaw on ice and gently mix prior to use. DO NOT VORTEX. Perform a quick spin before opening. Aliquot into small volumes and flash freeze for long term storage. Avoid multiple freeze/thaw cycles.
Specific Activity:	50 pmol/min/μg
Assay Conditions:	ALK (F1174S) activity was measured by using the IGF1Rtide synthetic peptide (KKKSPGEYVNIEFG) diluted in distilled water to a working concentration of 1 mg/ml, in a [33P]-ATP based assay. Reaction was initiated by mixing increasing amounts of ALK (F1174S) with 1250 pmoles of [33P]-ATP in 5 mM MOPS, pH 7.2, 2.5 mM β-glycerol-phosphate, 5 mM MgCl ₂ , 2.5 mM MnCl ₂ , 1 mM EGTA, 0.4 mM EDTA, 50 ng/μl BSA prepared with 50 μM DTT, 50 μM ATP and substrate at a final concentration of 200 μg/ml. The reaction was initiated by addition of [33P]-ATP Assay Cocktail, followed by a 15-minute incubation at 30°C. The reaction was terminated by spotting the reaction mixture on phosphocellulose P81 paper, air-dry and three 10-minute washes with 1% phosphoric acid solution. Radioactivity was measured in a scintillation counter. The corrected activity (RLU) was calculated by removing the blank value for each sample. The Kinase Specific Activity was calculated as follows: $RLU / [(specific\ activity\ of\ [33P]-ATP\ in\ cpm/pmol) * (Reaction\ time\ in\ min) * (Enzyme\ amount\ in\ \mu g\ or\ mg)] * [(Reaction$

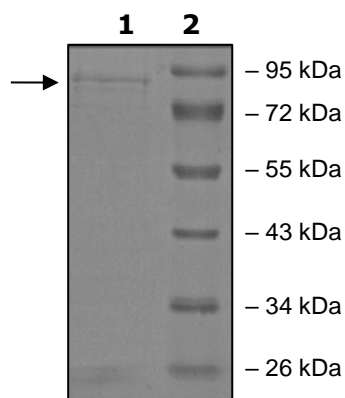
Volume) / (Spot Volume)]. The blank was determined from a “no substrate” sample by replacing the substrate solution with an equal volume of distilled water.

Applications:

Useful for the study of enzyme kinetics, screening inhibitors, and selectivity profiling.

Quality Control Data

4-20% SDS-PAGE Coomassie Staining



Specific Activity

