ROCK2, GST-tag Recombinant

Catalog: 40086
Lot: 240315

Product Information

Description:	Recombinant human ROCK2 (Rho-associated protein kinase 2), encompassing amino acids 5-554. The constructs contain an N-terminal GST-tag. The protein was affinity purified and is active.
Species:	Human
Construct:	ROCK2 (GST-5-554)
Concentration:	0.10 mg/ml
Expression System:	Sf9
Purity:	≥90%
Format:	Aqueous buffer solution.
Formulated In:	50 mM Tris-HCl, pH 7.5, 150 mM NaCl, 10 mM Glutathione, 0.1 mM EDTA, 0.25 mM DTT, 0.1 mM PMSF, and 25% glycerol
MW:	88 kDa
Genbank Accession:	NM_004580
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:	370 pmol/min/µg
Assay Conditions:	ROCK2 activity was measured by using S6K Substrate (KRRRLASLR), 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 ROCK2 with 1250 pmoles of [33P]-ATP in 5 mM MOPS, pH 7.2, 2.5 mM β-glycerol-phosphate, 5 mM $MgCl_2$, 1 mM EGTA, 0.4 mM EDTA, 50 ng/µl BSA, 50 µM DTT and substrate at a final concentration of 200 µg/ml. The reaction was initiated by addition of 5 µl of [33P]-ATP Assay Cocktail (50 µM of [33P]-ATP with 50 µM ATP) to 20 µl of Reaction Mix (10 µl of diluted ROCK2 + 5 µl of Substrate Solution + 5 µl of distilled water), 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 µg 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.

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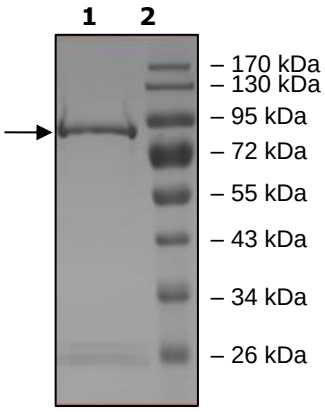
Quality Control Data

4-20% SDS-PAGE Coomassie Staining



Specific Activity