

Recombinant full-length human PIM 2 kinase containing N-terminal GST tag

Catalog No.	CRP108A CRP108B	Quantity:	5 µg 20 µg
Description:	Recombinant full-length human PIM 2 containing N-terminal GST tag was expressed by baculovirus in Sf 9 insect cells. A 334-amino acid sequence of the clone was deduced, which showed 90% identity with the mouse Pim2 protein. Like mouse Pim2, the human protein appears to be a serine threonine kinase. Northern blot analysis detected 2 PIM2 transcripts in all tissues tested, but most abundantly in hematopoietic tissues, spleen, thymus, and peripheral blood leukocytes, as well as in testis, small intestine, and colon. It was also highly expressed in human leukemic and lymphoma cell lines and a colorectal adenocarcinoma cell line. The results suggested a role for PIM2 in proliferating cells as well as during meiosis. Potential functions were investigated for the pim family of kinases in factor-dependent murine hematopoietic cells and indicate that pim-2 functions similarly to pim-1 as a pro-survival kinase and suggest that BAD is a legitimate PIM-2 substrate. It was concluded that the transcriptional induction of Pim-2 initiated a novel NF-kappaB activation pathway that regulates cell survival.		
Concentration:	0.1 mg/ml		
Gene Accession No:	NM_006875		
Source:	Sf9 insect cells		
Formulation:	Recombinant protein in storage buffer (50 mM Tris-HCl + 150 mM NaCl + 0.25 mM DTT + 0.1 mM EGTA + 0.1 mM EDTA + 0.1 mM PMSF + 25% glycerol; pH 7.5).		
Purity:	2.5 µg of protein was subjected to SDS-PAGE and Coomassie blue staining. The scan of the gel showed >90% purity of the PIM 2 product, and the band was at ~61 kDa (Fig. 2).		
Specific Activity:	305 nmol/min/mg: 305 nmol phosphate incorporated into the S6K substrate peptide per minute per mg protein at 30°C for 15 minutes using a final concentration of 50 µM ATP (0.83 µCi/assay). See QA/QC section for details.		
Storage & Stability:	Store product frozen at or below -70°C. Stable for 1 year at -70°C as undiluted stock. Aliquot to avoid repeated thawing and freezing.		



QA/QC:

Pim2 protein (100 ng/μl concentration) was diluted to 25 ng/μl with assay dilution buffer (4 mM MOPS + 2.5 mM β-glycerophosphate + 1 mM EGTA + 0.4 mM EDTA + 4 mM MgCl₂ + 0.05 mM DTT + 40 ng/μl BSA; pH 7.2), followed by 2-fold serial dilutions, and then the 10μl diluted proteins were used to phosphorylate the S6K substrate peptide (CKRRRLASLR) in the following assay condition:

- 10 μl diluted Pim2 protein
- 10 μl S6K substrate peptide (1 mg/ml stock)
- 5 μl [³²P] ATP mixture (250 μM ATP, 0.16 μCi/μl in 4x assay dilution buffer)

The various reaction components, except [³²P] ATP, were incubated at 30°C and the reaction started by the addition of [³²P] ATP. After 15 minutes, the reaction was terminated by spotting 20 μl of the reaction mixture onto a phosphocellulose P81 paper. The P81 paper was dried and washed several times in 1% phosphoric acid prior to counting in the presence of scintillation fluid in a scintillation counter. The actual counts, using various dilutions of the enzyme in the assay, are shown in Fig. 1.

Fig. 1 Pim2 activity assay

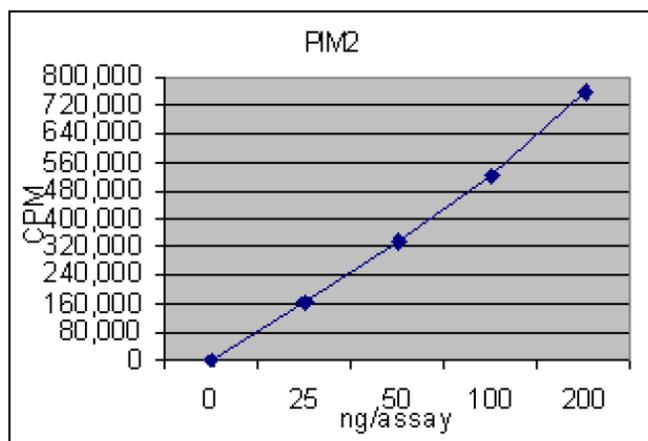
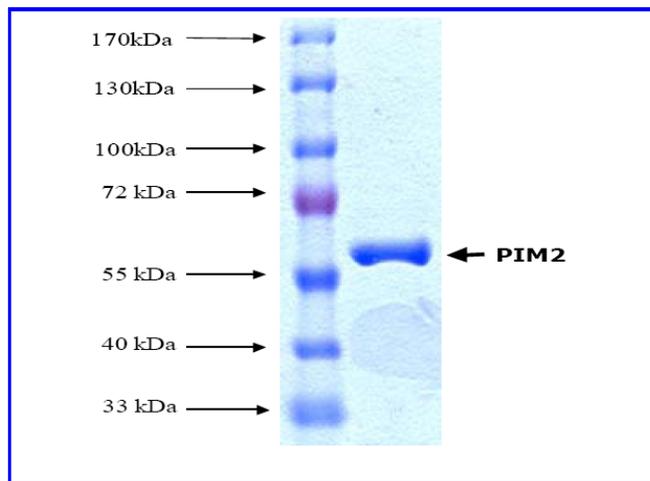


Fig. 2 Pim2 protein gel



Please note: always centrifuge vials before opening.

NOT FOR HUMAN USE. FOR RESEARCH ONLY. NOT FOR DIAGNOSTIC OR THERAPEUTIC USE.

