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Product Information

PTPN2, active, GST tagged, human recombinant, expressed in *E. coli* cells

Catalog Number **SRP5075**
Storage Temperature –70 °C

Synonyms: TC-PTP, PTPT, TCELLPTP, TCPTP

Product Description
Protein tyrosine phosphatase, non-receptor type 2 (PTPN2), is one of the most abundant mammalian tyrosine phosphatase. The protein encoded by this gene is a member of the protein tyrosine phosphatase (PTP) family.[1] By virtue of protein tyrosine phosphatase activity, PTPN2 is known to be a signaling molecule that regulates a variety of cellular processes including cell growth, differentiation, mitotic cycle, and oncogenic transformation involved in cell communication and signal transduction.[2]

Recombinant full length human PTPN2 was expressed in *E .coli* cells using an N-terminal GST tag. The gene accession number is NM_080422. Recombinant protein stored in 20 mM MOPS, pH 7.5, 50 mM NaCl, 10 mM glutathione, 0.25 mM DTT, 0.1 mM PMSF, and 30% glycerol.

Molecular mass: ~69 kDa

Purity: 70–95% (SDS-PAGE, see Figure 1)

Specific Activity: 6,928–9,372 nmole/min/mg
(see Figure 2)

Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability
The product ships on dry ice and storage at –70 °C is recommended. After opening, aliquot into smaller quantities and store at –70 °C. Avoid repeated handling and multiple freeze/thaw cycles.

Figure 1.
SDS-PAGE Gel of Typical Lot
70–95% (densitometry)

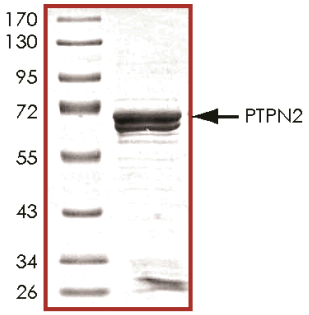
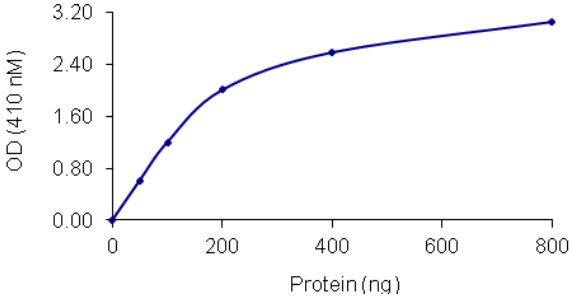


Figure 2.
Specific Activity of Typical Lot
6,928–9,372 nmole/min/mg



Procedure

Preparation Instructions

Phosphatase Assay Buffer – 125 mM HEPES, pH7.2, 250 mM NaCl, and 12.5 mM EDTA.

Phosphatase Dilution Buffer – Dilute the Phosphatase Assay Buffer 5-fold with a 5 mM DTT and 65 ng/µl BSA solution.

Phosphatase Solution – Dilute the active PTPN2 (0.1 µg/µl) with Phosphatase Dilution Buffer to the desired concentration.
Note: The lot-specific specific activity plot may be used as a guideline (see Figure 2). It is recommended the researcher perform a serial dilution of active PTPN2 for optimal results.

Stopping Solution – 2 M NaOH

Substrate Stock Solution – Prepare 500 mM *p*-nitrophenyl phosphate (pNPP) Substrate Stock Solution by dissolving 131.5 mg of pNPP in 1 ml of Phosphatase Dilution Buffer. Store at –20 °C. Avoid direct light exposure.

Substrate Assay Solution – Prepare 50 mM pNPP Substrate Assay Solution by diluting the Substrate Stock Solution 10-fold with Phosphatase Dilution Buffer. Prepare fresh before assay.

Phosphatase Assay

1. Prepare sufficient Substrate Assay Solution.
2. Thaw the active PTPN2 and Phosphate Dilution Buffer on ice.
3. In a pre-cooled microcentrifuge tube, add the following reaction components:
   10 µl of Phosphatase Solution
   20 µl of 50 mM pNPP Substrate Assay solution
   170 µl of Phosphatase Dilution Buffer
4. Set up a blank control as outlined in step 3, substituting 10µl of Phosphatase Dilution Buffer for the Phosphatase Solution.
5. Initiate each reaction by incubating the mixture in a water bath at 37 °C for 20 minutes.
6. After the 20 minute incubation, stop the reaction by the addition of 50 µl of 2 M NaOH Stopping Solution.
7. Measure the absorbance of the reaction solution in a spectrophotometer at 405 nm.
8. Determine the Phosphatase specific activity.

Calculations:

1. Specific Phosphatase Activity (SA) (nmole/min/mg)

$$\text{nmole/min/mg} = \frac{Pv \times OD_{405nm}}{\varepsilon \times d \times T \times Pm}$$

Pv - Phosphatase volume (µl)
ε - extinction coefficient (17.8 µl/nmole/cm)
d - pathlength of light (cm)
T - incubation time (min)
Pm - Phosphatase amount (mg)

References

1. Gupta, S. et al., A nuclear protein tyrosine phosphatase activates p53 and induces caspase-1-dependent apoptosis. FEBS Lett., **532**, 61-66 (2002).
2. Yamamoto, T. et al., The nuclear isoform of protein-tyrosine phosphatase TC-PTP regulates interleukin-6-mediated signaling pathway through STAT3 dephosphorylation. Biochem. Biophys. Res. Commun., **297**, 811-817 (2002).

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