

## Product Information

**PTPN7, active, GST tagged, human recombinant, expressed in *E. coli* cells**

Catalog Number **SRP5077**  
Storage Temperature  $-70^{\circ}\text{C}$

Synonyms: LC-PTP, LPTP, HEPTP, PTPNI, BPTP-4

### Product Description

PTPN7 gene is preferentially expressed in a variety of hematopoietic cells and is an early response gene in lymphokine stimulated cells.<sup>1</sup> The noncatalytic N-terminus of this PTP can interact with MAP kinases and negatively regulates ERK2 and p38 MAP-kinases activity.<sup>2</sup> PTPN7 was shown to be involved in the regulation of T cell antigen receptor (TCR) signaling, which was thought to function through dephosphorylating the molecules related to MAP kinase pathway.<sup>3</sup>

Recombinant full length human PTPN7 was expressed in *E. coli* cells using an N-terminal GST tag. The gene accession number is NM\_002832. 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: ~67 kDa

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

Specific Activity: 820–1,110 nmole/min/mg  
(see Figure 2)

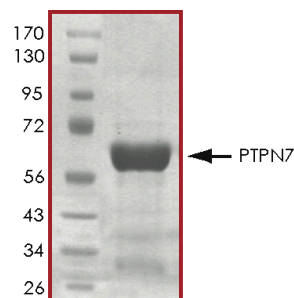
### Precautions and Disclaimer

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.

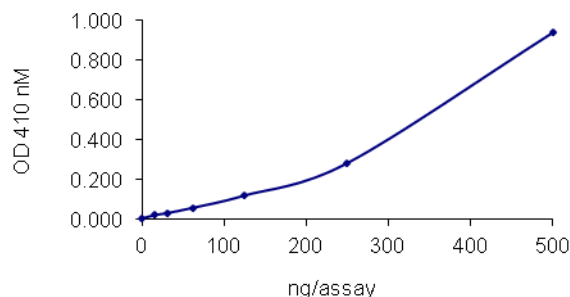
### Storage/Stability

The product ships on dry ice and storage at  $-70^{\circ}\text{C}$  is recommended. After opening, aliquot into smaller quantities and store at  $-70^{\circ}\text{C}$ . 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  
820–1,110 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 PTPN7 (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 PTPN7 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 PTPN7 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)

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

- 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. Adachi, M. et al., Protein-tyrosine phosphatase expression in pre-B cell NALM-6. *Cancer Res.*, **52**, 737-740 (1992).
2. Pettiford, S.M. et al., The MAP-kinase ERK2 is a specific substrate of the protein tyrosine phosphatase HePTP. *Oncogene*, **19**, 858-69 (2000).
3. Oh-hora, M. et al., Direct suppression of TCR-mediated activation of extracellular signal-regulated kinase by leukocyte protein tyrosine phosphatase, a tyrosine-specific phosphatase. *J. Immunol.*, **163**, 1282-1288 (1999).

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