



**PTP1B full length. E.coli Active Enzyme**

**BACKGROUND**

Protein tyrosine phosphatase 1B (PTP1B) is an enzyme that removes phosphate groups covalently attached to tyrosine residues in proteins. This ubiquitously expressed enzyme is anchored in the endoplasmic reticulum by its C-terminal end and has its catalytic regions exposed to the cytosol. PTP1B will dephosphorylate a wide variety of phosphoproteins, such as receptors for the growth factors insulin and epidermal growth factor (EGF), c-Src and beta-catenin. Of particular interest is the observation that PTP1B knock-out mice are resistant to high-caloric intake-induced obesity and have exaggerated insulin responses, suggesting that PTP1B may play an important role in regulating growth factor responsiveness.

**ACTIVITY**

Useful for the study of enzyme kinetics, regulation, to dephosphorylate target substrates and for screening inhibitors. Specific Activity: 10 U/ug. One unit will hydrolyze 1 nmol p-nitrophenyl phosphate per minute at pH 7.4 and 30 °C. Assay buffer: 50 mM HEPES, pH 7.4, 2 mM EDTA, 3mM DTT, 100 mM NaCl, 50 mM pNPP.

**PURITY**

>70% by SDS-PAGE

**APPLICATIONS**

The specific activity of mouse PTP1B was determined using pNPP.

Application: Useful for the study of enzyme kinetics, regulation, to dephosphorylate target substrates and for screening inhibitors

**ORDERING INFORMATION**

**CATALOG NUMBER**

X1765E

**SIZE**

20 µg

**CUSTOMER STORAGE**

Product should be stored at -80°C.  
Aliquot to avoid freeze/thaw cycles

**FORMULATION**

Provided in 25 mM Tris-HCl, 75 mM NaCl, pH 8.0, 0.05% Tween, 2 mM EDTA, 1 mM DTT and 50% glycerol, 10 mM glutathione

**SHIP CONDITIONS**

Ship on dry ice, freeze upon arrival

**STABILITY**

Stable for > 6 months when stored as recommended.

**CONCENTRATION**

Lot specific, see vial

**SOURCE**

Full-Length PTP1B (a.a. 1-436) with N-terminal GST-His tag expressed in E.coli expression system

## **ASSAY METHODS**

### **MATERIALS**

Assay Buffer: 50 mM HEPES, pH 7.4, 100 mM NaCl, 2 mM EDTA, 3 mM DTT

Substrate: 5X pNPP (10 mM) in assay buffer

Enzyme Prep.: Dilute enzyme (0.4 mg/ml starting concentration) 1:80 in assay buffer.

### **PROCEDURE**

1. Prepare reaction mixtures in 96-well plate (keep on ice) in the following order:
  - 60  $\mu$ l of assay buffer
  - 20  $\mu$ l of 10 mM pNPP in assay buffer (final conc. 2 mM)
  - 20  $\mu$ l of 5 ng/ $\mu$ l PTP1B enzyme in assay buffer (final conc. 25 nM).
  - Mix well and start reaction at 30°C water bath and incubate for 10 min.
2. Add 100  $\mu$ l per well of 2 M K<sub>2</sub>CO<sub>2</sub> to stop reaction.
3. Read absorbance at 405 nm using a plate reader.

### **REFERENCES**

1. Frangioni, J.V., et al. (1992). The nontransmembrane tyrosine phosphatase PTP-1B localizes to the endoplasmic reticulum via its 35 amino acid C-terminal sequence. *Cell* 68(3);545-560
2. Elchebly, M., et al (1999). Increased insulin sensitivity and obesity resistance in mice lacking the protein tyrosine phosphatase-1B gene. *Science* 283(5407);1544-1548

### **PRODUCT SPECIFIC REFERENCES**

Last Modified  
5/30/2017

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