PTP (2-315)/PTPN2. E.coli Active Enzyme

Protein Tyrosine Phosphatase, Non-receptor Type 2; T-cell Protein Tyrosine Phosphatase

Background

T-cell protein tyrosine phosphatase (TC-PTP), also known as PTPT and PTPN2, is an enzyme that removes phosphate groups covalently attached to tyrosine residues in proteins. This enzyme has two C-terminal end splice variants with distinctly different subcellular localizations. The shorter 45 kilodalton isoform is exclusively nuclear in resting cells, but redistrubutes to the cytosol upon stimulation with growth factors 1 and cellular stress 2 . The longer 48 kilodalton isoform is exclusively found in the endoplasmic reticulum 3 and seems to have distinctly different physiologic substrates from the smaller isoform. 1, 4 Although found in many cell types and tissues, TC-PTP is particularly prominent in hemopoietic cell types. 5, 6 Knockout mice lacking TC-PTP are born viable but die 3 to 5 weeks after birth of erythropoietic and lymphopoietic deficits 7 , indicating a critical role for TC-PTP in bone marrow maturation. TC-PTP will dephosphorylate a wide range of phosphoproteins, such as p52 Shc 6 and receptors for EGF 1 , Insulin 8 and growth hormone. 6 The recombinant protein lacks the C-terminal 100 amino acids that determine intracellular localization but is fully active. 9

Activity

1.5 nmole/min/μg of enzyme; Detemined using pNPP; Reaction conditions: 50 μM pNPP, 10 min incubation at 30ºC, 0.5 μg enzyme.

Purity

>95% pure as determined by SDS-PAGE

Applications

Useful for the study of enzyme kinetics, screening inhibitors, and selectivity profiling.

Ordering Information

Catalog Number
X1660E

Size
20 μg

Customer Storage
Product should be stored at -80ºC. Aliquot to avoid freeze/thaw cycles

Formulation
25 mM Tris-HCl, pH 8.0, 75 mM NaCl, 0.05% Tween-20, 50% glycerol, 2 mM EDTA, 1 mM DTT, 10 mM glutathione

Ship Conditions
Ship on dry ice, freeze upon arrival

Stability
Products are stable for one year from purchase when stored properly

Concentration
See vial for concentration

Source
Recombinant enyzme produced in E. coli

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Assay Methods

Materials

1. Assay Buffer: 50 mM HEPES, pH 7.4, 100 mM NaCl, 2 mM EDTA, 3 mM DTT
2. Stop solution: 2M K_2CO_3
3. 190 mM pNPP
4. Microtiter plate
5. Microtiter plate reader capable of measurements at 405 nm
6. Water bath or incubator at 30ºC

Procedure

1. Prepare reaction mixture:
a. 73 μl assay buffer
b. 26 μl pNPP (Final concentration of pNPP is 50 mM)
c. 1 μl of TCPTP
2. Mix well and start reaction at 30ºC in water bath and incubate for 10 min.
3. Add 100 μl per well of 2 M K_2CO_3 to stop the reaction.
4. Read absorbance at 405 nm using a microtiter plate reader.

References

1. Tiganis, T. et al., 1999, J. Biol. Chem. 274:27768.
2. Lam, M.H. et al., 2001, J. Biol. Chem. 276:37700.
3. Lorenzen, J.A. et al., 1995, J. Cell Biol. 131:631.
4. Tiganis, T. et al., 1998, Mol. Cell. Biol. 18:1622.
5. Cool, D.E. et al., 1989, Proc. Natl. Acad. Sci. USA 86:5257.
6. Pasquali, C. et al., 2003, Mol. Endocrinol. 17:2228.
7. You-Ten, K.E. et al., 1997, J. Exp. Med. 186:683.
8. Galic, S. et al., 2003, Mol. Cell. Biol. 23:2096.
9. Cool, D.E. et al., 1990, Proc. Natl. Acad. Sci. USA 87:7280.

Product Specific References

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