

Product Information

Polynucleotide phosphorylase human, histidine-tagged recombinant, expressed in *Escherichia coli*

Catalog Number **N1290**

Storage Temperature –70 °C

EC 2.7.7.8

Synonyms: Polyribonucleotide nucleotidyltransferase, hPNPase

Product Description

Polynucleotide phosphorylase (PNPase) is a bifunctional enzyme with a phosphorolytic 3' to 5' exoribonuclease activity and a 3'-terminal oligonucleotide polymerase activity.¹ It is also involved in mRNA processing and degradation in bacteria, plants, and humans.²

The human PNPase (hPNPase) was identified in an overlapping pathway screen to discover genes displaying coordinated expression as a consequence of terminal differentiation and senescence of melanoma cells.³ Recent studies show, unlike chloroplast and plant mitochondria in which the PNPase is located in the stroma and matrix, the human PNPase is mostly or exclusively located in the mitochondrial intermembrane space (IMS).⁴⁻⁵ The human PNPase exhibits a very similar structure to the bacterial enzyme. The subunit molecular mass of the human enzyme is ~90 kDa and the native protein is present as a multimeric complex.⁶

hPNPase was co-purified with the RNA helicase hSUV3 suggesting participation of both proteins in mtRNA metabolism. hPNPase was also purified as an interacting protein with TCL1, an oncoprotein promoting B and T cell malignancies and apoptotic stimuli.^{4,7,8} Given the above, hPNPase has a pleotropic biological effect on mitochondrial homeostasis, cellular senescence, mtRNA poly(A) tail length alteration, and cell stress.⁹⁻¹⁰ Currently, efforts are directed towards understanding the regulation of hPNPase expression, its subcellular localization, and the mechanism(s) by which it performs its numerous cellular functions. In comparison with bacteria and chloroplast PNPase, hPNPase responds to much lower concentrations of P_i, changing its mode of activity between degradation and polymerization.

The specificity of the enzyme for polymerization is high for ADP, like that of the *E. coli* PNPase, with lower activity when incubated with other NDPs.

The product is supplied as a solution in 20 mM HEPES buffer, pH 7.9, with 0.1 mM EDTA, 2 mM DTT, 12.5 mM MgCl₂, ~130 mM KCl, and 20% (w/v) glycerol.

Purity: ≥90% (SDS-PAGE)

Specific activity: ≥20 units/mg-protein

Unit definition: One unit will polymerize 1.0 μmole of ADP, releasing 1.0 μmole of inorganic phosphate in 15 minutes at pH 9.1 at 42 °C.

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

Store the product at –70 °C. The product is stable for at least 2 years as supplied. Avoid repeated freezing and thawing. After initial thawing, the enzyme should be refrozen at –70 °C in aliquots.

References

1. Yehudai-Resheff, S., et al., Polynucleotide phosphorylase functions as both an exonuclease and a Poly (A) polymerase in spinach chloroplasts. *Mol. Cell. Biol.*, **21**, 5408-5416 (2001).
2. Liou, G.G., et al., RNA degradosomes exist *in vivo* in *Escherichia coli* as multicomponent complexes associated with the cytoplasmic membrane via the N-terminal region of ribonuclease E. *Proc. Natl. Acad. Sci. USA*, **98**, 63-68 (2001).
3. Leszczyniecka, M., et al., Identification and cloning of human polynucleotide phosphorylase, hPNPase old-35, in the context of terminal differentiation and cellular senescence. *Proc. Natl. Acad. Sci. USA*, **99**, 16636-16641 (2002).
4. Chen, H.W., et al., Mammalian polynucleotide phosphorylase is an intermembrane space RNase that maintains mitochondrial homeostasis. *Mol. Cell. Biol.*, **26**, 8475-8487 (2006).

5. Rainey, R.N., et al., A new function in translocation for the mitochondrial i-AAA protease Yme1: import of polynucleotide phosphorylase into the intermembrane space. *Mol. Cell. Biol.*, **26**, 8488–8497 (2006).
6. Portnoy, V., et al., Analysis of the human polynucleotide phosphorylase (PNPase) reveals differences in RNA binding and response to phosphate compared to its bacterial and chloroplast counterparts. *RNA*, **14**, 297–309 (2008).
7. Szczesny, R.J., Human mitochondrial RNA turnover caught in flagranti: involvement of hSuv3p helicase in RNA surveillance. *Nucl. Acids Res.*, **38**, 279–298 (2010).
8. French, S.W., et al., The TCL1 oncoprotein binds the RNase PH domains of the PNPase exoribonuclease without affecting its RNA degrading activity. *Cancer Lett.* **248**, 198–210 (2007).
9. Borowski, L.S., et al., RNA turnover in human mitochondria: More questions than answers? *Biochim. Biophys. Acta.*, [Epub ahead of print] doi:10.1016/j.bbabi.2010.01.028.
10. Chen, H.W., Human polynucleotide phosphorylase: location matters. *Trends Cell Biol.*, **17**, 600-608 (2007).

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