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Product Information

Polynucleotide phosphorylase from *Escherichia coli,* histidine tagged recombinant, expressed in *Escherichia coli*

Catalog Number **N9664**
Storage Temperature –70 °C

EC 2.7.7.8
Synonyms: Polyribonucleotide nucleotidyltransferase, PNPase

Product Description
Polynucleotide phosphorylase (PNPase) is a bifunctional enzyme with a 3′-terminal oligonucleotide polymerase activity and a phosphorolytic 3′ to 5′ exoribonuclease activity.[1] It is also involved in mRNA processing and degradation in bacteria, plants, and humans.[2] The subunit molecular mass is 84–86 kDa.[3] The native protein is present as a trimeric homopolymer with a molecular mass of 230 kDa.[3,4]

Polynucleotide phosphorylase (PNPase) is present in the chloroplasts and mitochondria of some eukaryotic cells. The enzyme is a functional part of the "degradosome", a multienzyme complex (molecular mass ~500 kDa).[5,6] PNPase was shown to protect *E. coli* against oxidative stress by specifically binding to RNA molecules that were oxidatively damaged.[7] The *E. coli* PNPase enzyme was shown to polymerize all ribonucleotides in the presence of divalent cations such as Mg^{2+} or Mn^{2+} for GDP.[7,8] The affinity of *E. coli* PNPase to poly(G) sequences is very low and thus, this polynucleotide can be used as an effective barrier to exonuclease activity in yeast and chloroplasts.[5]

It is important to mention the enzyme activity on different nucleotides may require different assay conditions, e.g., maximal polymerization activity was observed with ADP or IDP at 55 °C and pH 9.1; whereas, CDP and UDP were maximal at 37 °C. GDP polymerization is very sensitive to pH changes and gave the lowest activity of any of the nucleotides tested at 37 °C.

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 $MgCl_2$, 200 mM KCl, and 21.4% (w/v) glycerol.

Purity: ≥90% (SDS-PAGE)

Specific activity: ≥250 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 37 °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 may be kept for up to 2 months at 4 °C or refrozen in aliquots at –70 °C.

References
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6. Carpousis, A.J., The *Escherichia coli* RNA degradosome: structure, function and relationship in other ribonucleolytic multienzyme complexes. *Biochem. Soc. Trans.*, **30**, 150-155 (2002).
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