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ProductInformation

Nuclease P_1 from Penicillium citrinum

Product Number **N 8630**
Storage Temperature 2-8 °C

Product Description
Enzyme Commission (EC) Number: 3.1.30.1
CAS Number: 54576-84-0
Molecular Weight: 42-50 kDa[1,2]

This enzyme hydrolyzes both 3'-5' phosphodiester bonds in RNA and heat-denatured DNA, and 3'-phosphomonoester bonds in mono and oligo nucleotides terminated with a 3'-phosphate group without base specificity. The enzyme does not actually attack double-stranded nucleic acids, especially in the presence of more than 400 mM of sodium chloride at pH 6.0. The rate of hydrolysis of 2'-AMP is 3,000-fold less than that of 3'-AMP. The enzyme does not attack 5'-nucleotides, p-nitrophenylphosphate, bis-p-nitrophenylphosphate, nor 3', 5'-cyclic AMP.

The enzyme has an optimal temperature of approximately 70 ºC, but for a long incubation, a temperature below 60 ºC is more suitable. It is stable in the pH range of 5 - 8.[3]

Approximately 50% inactivation is observed after treatment in 29 mM veronal-acetate buffer, pH 5.3, at 67 ºC for 15 minutes.

Optimal conditions for complete digestion of RNA or heat-denatured DNA are pH 5.3 and 50 ºC. Under such conditions, one mg of the enzyme should completely hydrolyze 2 g of RNA or 0.2 g of heat-denatured DNA into 5'-mononucleotides in one hour. Addition of 0.1 mM $ZnCl_2$ to the reaction mixture is effective for stabilization of the enzyme during incubation.

Precautions and Disclaimer
For Laboratory Use Only. Not for drug, household or other uses.

Storage/Stability
It was reported that when an enzyme solution containing 50 µg of the enzyme in 1 ml of 0.1 M ammonium acetate, pH 4.5, was allowed to stand at 4 ºC for 3 days, enzyme activity toward 3'-AMP, RNA, and heat-denatured DNA was reduced to 26%, 15% and 19%, respectively. The lower the pH of the enzyme solution, the higher the rate of inactivation. Two ml of diluted amino acids (a mixture containing 50 µM of each amino acid except proline at 0.1 mM, pH 5.3) and 0.1 ml of 0.1 M zinc chloride were added to the partially inactivated enzyme solution and heated at 60 ºC for 15 minutes. After this procedure, enzyme activity towards 3'-AMP, RNA, and heat-denatured DNA was increased to 78%, 62% and 64%, respectively. It was noted that the addition of 1% human serum albumin and 2 mM zinc chloride stabilized an enzyme solution (80 µg/ml, pH 5.3) through an incubation at 70 ºC for 30 minutes. With either HSA or Zn^{2+}, activity towards RNA or 3'-AMP was completely lost. With both, the remaining activities were 77% and 79%, respectively.[3]

References
1. Martin, S. A., et al., A Comparative Study of Nucleases Exhibiting Preference for Single-stranded Nucleic Acid. Biochim. Biophys. Acta., **867**, 76-80 (1986).
2. Shishido, K., and Ando, T., Cold Spring Harbor Monograph Series, **14**, 155-185 (1982).
3. Fujimoto, M., et al., Identity of Phosphodiesterase and Phosphomonoesterase Activities with Nuclease P1. Agr. Biol. Chem., **38**, 785-790 (1974).

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