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Product Information

Phosphatase, Alkaline from bovine intestinal mucosa

Buffered aqueous glycerol solution

A2356

Product Description

CAS Registry Number: 9001-78-9

Enzyme Commission (EC) Number: 3.1.3.1

Synonym: alkaline phosphomonoesterase, phosphomonoesterase, glycerophosphatase, alkaline phosphohydrolase, alkaline phenyl phosphatase, orthophosphoric-monoester phosphohydrolase (alkaline optimum)

K_M:

- 1.5×10^{-3} M (*p*-nitrophenyl phosphate)
- 19×10^{-3} M (phosphoenolpyruvate)

Molecular mass: ^{1,2} 140–160 kDa

 $E_{278}^{1\%} = 7.6 - 10.5$

Isoelectric point: $^{3-5}$ several isozymes with a pI range of 4.4-5.8

Bovine intestinal alkaline phosphatase is a dimeric, membrane-derived glycoprotein.^{1,2,6} At least three isoforms exist, which typically possess two N-linked and one or more O-linked glycans per monomer.¹ The enzyme requires zinc, and magnesium or calcium divalent ions for activity.^{3,7}

Alkaline phosphatase has a broad specificity for phosphate esters of alcohols, amines, pyrophosphate, and phenols. It is routinely used to dephosphorylate proteins and nucleic acids.⁸⁻¹⁰ Other applications of alkaline phosphatase include conjugation to antibodies and other proteins for ELISA, Western blotting, and histochemical detection.^{11,12}

Alkaline phosphatase may be used to dephosphorylate the 5'-termini of DNA or RNA to prevent self-ligation. DNA or RNA can also be tagged with radiolabeled phosphate (via T4 polynucleotide kinase) after dephosphorylation with alkaline phosphatase.¹³ Alkaline phosphatase has also been used to dephosphorylate casein and other proteins.^{14,15}

pH optimum

- The enzyme is most stable in the pH range 7.5–9.5.²
- The pH optimum for enzymatic activity is pH 8–10.
- The pH optimum will change depending upon substrate, substrate concentration, and ionic concentration.⁴
- The enzyme activity for this product is determined at pH 9.8 [diethanolamine (DEA) buffer enzyme assay].

Inhibitors^{5,11}

- Chelating agents
- Arsenate
- Cysteine
- Iodine
- Inorganic phosphate
- Pyrophosphate
- Diisopropyl phosphate
- Triphenylphosphate
- Diisopropyl fluorophosphate
- L-phenylalanine

Levamisole (such as Cat. No. L9756) is typically used to inhibit endogenous alkaline phosphatase activity, while only slightly inhibiting the intestinal enzyme.^{16,17}

Precautions and Disclaimer

This product is for R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.



Product

This product is supplied as a solution in 40% glycerol containing 6 mM Tris, 6 mM MgCl₂ and 0.12 mM ZnCl₂, at pH \sim 7.6.

Specific Activity: \geq 5,500 units/mg protein (DEA units)

Unit Definition: One DEA unit will hydrolyze 1 μ mole of 4-nitrophenyl phosphate per minute at pH 9.8 at 37 °C.

One glycine unit is approximately equivalent to \sim 3 DEA units.

Storage/Stability

Store the solution, as supplied, at 2-8 °C. The product remains active for at least 1 year.

Preparation Instructions

Dilute solutions of alkaline phosphatase should be prepared in 10 mM Tris HCl (pH 8.0), 1-5 mM MgCl₂, 0.1-0.2 mM ZnCl₂. 50% Glycerol can be included for long term storage at 2-8 $^{\circ}$ C.

References

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