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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<sub>M</sub>:

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

Molecular mass: <sup>1,2</sup> 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.<sup>1,2,6</sup> At least three isoforms exist, which typically possess two N-linked and one or more O-linked glycans per monomer.<sup>1</sup> The enzyme requires zinc, and magnesium or calcium divalent ions for activity.<sup>3,7</sup>

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.<sup>8-10</sup> Other applications of alkaline phosphatase include conjugation to antibodies and other proteins for ELISA, Western blotting, and histochemical detection.<sup>11,12</sup>

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.<sup>13</sup> Alkaline phosphatase has also been used to dephosphorylate casein and other proteins.<sup>14,15</sup>

#### pH optimum

- The enzyme is most stable in the pH range 7.5–9.5.<sup>2</sup>
- The pH optimum for enzymatic activity is pH 8–10.
- The pH optimum will change depending upon substrate, substrate concentration, and ionic concentration.<sup>4</sup>
- The enzyme activity for this product is determined at pH 9.8 [diethanolamine (DEA) buffer enzyme assay].

#### Inhibitors<sup>5,11</sup>

- 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.<sup>16,17</sup>

## 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 MgCl<sub>2</sub> and 0.12 mM ZnCl<sub>2</sub>, at pH  $\sim$ 7.6.

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

Unit Definition: One DEA unit will hydrolyze 1  $\mu$ 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 MgCl<sub>2</sub>, 0.1-0.2 mM ZnCl<sub>2</sub>. 50% Glycerol can be included for long term storage at 2-8  $^{\circ}$ C.

#### References

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