

CUSTOMBIOTECH

Alkaline Phosphatase

Test principle: Enzymatic colorimetric

4-Nitrophenyl phosphate (colorless) \xrightarrow{ALP} 4-Nitrophenol (yellow)

The assay method was first described by King and Armstrong, modified by Ohmori, Bessey, Lowry, and Brock, and later improved by Hausamen *et al.* In 1983, the International Federation of Clinical Chemistry (IFCC) recommended a standardized method for the determination of alkaline phosphatase using an optimized substrate concentration and 2-amino-2-methyl-1-propanol as buffer, plus the cations magnesium and zinc. The assay described below meets the recommendations of the IFCC. Colorimetric assay in accordance with a standardized method. In the presence of magnesium and zinc ions, p-nitrophenyl phosphate is cleaved by phosphatases into phosphate and p-nitrophenol.

The p-nitrophenol released is directly proportional to the catalytic ALP activity. It is determined by measuring the increase in absorbance at 409 nm.

Proposed reagent composition

approximately 5+1 formulation

Reagent 1

| Composition | Concentration |
|---|---------------|
| 2-Amino-2-methyl-1-propanol (pH 10.44) | 1.724 mmol/l |
| Magnesium acetate | 3.830 mmol/l |
| Zinc sulfate | 0.766 mmol/l |
| N-(2-hydroxyethyl)-ethylenediamine triacetic acid | 3.830 mmol/l |

Reagent 2

| Composition | Concentration | Catalog Number |
|-------------------------|---------------|----------------|
| p-Nitrophenyl phosphate | 132.8 mmol/l | 10 270 857 103 |
| Preservative | | |

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