

LYPHOZYME

# ALKALINE PHOSPHATASE

(AMP OPTIMIZED IFCC METHOD)

Code	Product Name	Pack Size
LP001A	Lyphozyme Alkaline phosphatase	30 x 1.2 ml

## INTENDED USE:

This reagent kit is intended for "In Vitro" quantitative determination of Alkaline Phosphatase activity in serum/plasma.

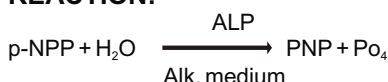
## CLINICAL SIGNIFICANCE:

Elevated levels of Serum Alkaline Phosphatase are generally observed in growing children. In grown up adults the elevated levels are associated with born diseases like, metastasis, rickets, paget's disease, healing fractures, as wells in liver and biliary tract diseases, more over progressive increase in serum Alkaline Phosphatase activity may be the first indication of adverse reaction to certain therapeutic drugs like androgens, anabolic steroids, estrogen, sulphonamides, MAO inhibitors etc. The decrease levels are found in severe anaemia, scurvy etc.

## PRINCIPLE:

Alkaline Phosphatase in serum catalyse the hydrolysis of p. nitrophenyl phosphate to p. nitrophenol and phosphate. p. nitrophenol is yellow coloured compound. As the reaction progresses the rate of absorbance increases which is proportional to the activity of Alkaline Phosphatase in the sample. This reaction takes place in alkaline medium and in presence of magnesium ions. The change in absorbance is measured at 405 nm.

## REACTION:



## REAGENTS:

Reagent 1 : PNPP Substrate

Reagent 2 : AMP buffer

## MATERIALS REQUIRED BUT NOT PROVIDED:-

- Clean & Dry Glassware.
- Laboratory Glass Pipettes or Micropipettes & Tips.
- Colorimeter or Bio-Chemistry Analyzer.

## SAMPLES:

Unhaemolysed Serum, heparinized plasma / EDTA plasma

## PREPARATION OF REAGENT & STABILITY:

Step1: Bring the buffer / kit contents to room temperature

Step 2 : Dissolve the substrate reagent in the buffer provided as per the volume indicated on the vial

Step 3 : Allow the working reagent mixture to stand for 10 minutes at room temperature for equilibration.

The working reagent is ready for the use.

1. The unopened substrate vial is stable till expiry date when store at 2°-8° C.
2. The reconstituted reagent is stable for 7 days at 2°-8°C.

## GENERAL SYSTEM PARAMETERS:

Reaction type	: Kinetic reaction (Increasing)
Wave length	: 405 nm
Temperature	: 37°C
Delay	: 30 Sec.
Interval	: 30 Sec.
No. of readings	: 4
Sample volume	: 20 µl.
Reagent volume	: 1ml
Factor	: 2712
Zero setting	: Deionised water
Light path	: 1 cm



**BEACON**

## PROCEDURE:

Working Assay Table :

Working Reagent	1.0 ml.
Sample	20µl

Mix well. Read absorbance after 30 seconds. Repeat reading after every 30 seconds i.e. Up to 120 seconds at 405 nm wavelength.

Determine the mean absorbance change per minute (ΔA/minute)

## CALCULATION :

Alkaline Phosphatase activity IU/L = ΔA/min X 2712

Where ΔA = change in Absorbance per min. and 2712 is factor

## LINEARITY:

This kit has been standardized to perform accurately upto an enzyme activity of 1500 IU / L.

## NORMAL VALUE:

The expected range of normal value using this procedure.

Children (3-15 yrs): 104 - 390 IU / Ltr

Adults : 25 - 147 IU / Ltr

## QUALITY CONTROL :

For accuracy, it is advised to run known serum controls with each assay.

## LIMITATION & PRECAUTIONS :

1. Storage conditions as mentioned on the kit to be adhered.
2. PNPP is photo sensitive so if exposed to light it would start deteriorating.
3. Discard the reagent if the initial absorbance exceeds 0.4 at 405 nm.
4. Avoid hemolysed serum as it interferes with the result.
5. Reagent : Sample ratio as mentioned here above must be strictly observed as any change in it will adversely effect the factor.

## BIBLIOGRAPHY:

1. Rec. GSCC (DGKC) ; J. Clin. Chem. Clin. Biochem. 1972; 10 : 182.
2. Heerspijck W., Hafkenscheidt J. C. M., Siepelvander Ven Jongekryg J., Djit C. C. M., Enzyme 25, 333 - 341 (1980).



## SYMBOLS USED ON LABELS

REF	Catalogue Number	Manufacturer	See Instruction for Use
LOT	Lot Number	CONT	Content
Expiry Date		IVD	In Vitro Diagnostics
		Storage Temperature	

BEA/24/ALP/LP/IFU-01