Alkaline Phosphatase

Modified AMP-PNPP Method

CLINICAL SIGNIFICANCE

An alkaline Phosphatase level test (ALP) measure the alkaline phosphatise enzyme in your bloodstream. The test requires a simple blood draw and often a routine part of blood test. Abnormal levels of ALP in your blood most often indicate a problem with your liver, gallbladder, or bones. However, they may also indicate malnutrition, kidney cancer tumors, intestinal issues, a pancreas problem, or a serious infection. Y=the normal range of Alp varies from person to person and depends on your age, blood type, gender and whether your pregnant.

PRINCIPLE

Alkaline Phosphatase (ALP) at an alkaline pH hydrolyses pnitrophenyl phosphate into p-nitrophenol & inorganic phosphates. The rate of formation of p-nitrophenol is measured as an increase in absorbance which is directly proportional to the ALP activity in the sample.

$$\text{p - Nitrophenyl phosphate} + H_2O \xrightarrow{\text{ALP}} \text{p - Nitrophenol} + \text{Inorganic phosphate}$$

ALP: Alkaline Phosphatase

KIT CONTENTS

	25 X 01 ml	2 X 25 ml
Reagent 1: ALP reagent	25 X 01 ml	2 X 25 ml

STORAGE INSTRUCTIONS AND REAGENT STABILITY

Reagents are stable at 2^0-8^0C upto the expiry date mentioned on the label.

REAGENTS PREPARATION

All reagents are ready to use.

SAMPLE MATERIAL

Serum or Plasma. (Free from haemolysis.)

The stability in serum/plasma: 2 days at 2^0-8^0C.

ASSAY PROCEDURE

Wavelength / Filter : 405 nm

Temperature : 37^{0}C

Cuvette : 1 cm light path.

Reagent	Volume
R_1-ALP reagent	1000ul
Serum	20ul

Mix well and aspirate & read the initial absorbance Ao & repeat the absorbance readings after every 1 & 2 minutes for total 2 minutes time. Calculate the mean absorbance change per minute i.e.(Δ A / min).

CALCULATION:

$$\text{ALP in U/L} = \frac{\text{(Abs.T) - (Abs.B)}}{\text{(Abs.S) - (Abs.B)}} \times 3100$$

REFERENCE VALUES:

Serum (Adults): 25 - 140 U/L at 37° C

(Children up to 12 yrs): 104 - 390 U/L at 37° C

"Each Laboratory should check if references ranges are transferable to its own patient's population and determine own references ranges if necessary."

PERFORMANCE CHARACTERISTICS

Sensitivity / Limit of Quantitation: 28 U/L

Linearity: up to 1000 U/L

Specificity /Interferences: Haemoglobin up to 2.50 g/dl, ascorbate up to 62 mg/l, bilirubin up to 20 mg/dl and triglycerides up to 500 mg/dl do not interfere with the test.

WARNINGS AND PRECAUTIONS

1. Keep out reach of children. In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
2. Take off immediately all contaminated clothing.
3. Wear suitable gloves and eyes/face protection.
4. Always use safety pipettes to pull the reagents into a pipette.
5. Reagents may contain some non-reactive and preservative components. It is suggested to handle carefully, avoid direct contact with skin and do not swallow.
6. Perform the test according to the "current Good Laboratory Practice" (cGLP) guidelines.

SYSTEM PARAMETERS

Parameters	FOR SEMI-AUTO	FOR FULLY- AUTO
Mode of reaction	Kinetic method	Kinetic method
Wavelength / Filter	405 nm	405 nm
Slope of reaction	Increasing	Increasing
Temperature	37^{O}C	37^{O}C
Blank Reagent	Distilled water	Distilled water
Factor	3100	3100
Delay Time	60 sec.	
Read Time	120 sec	
Interval Time	60 sec	
Sample volume	20ul	06ul
Reagent R_1 volume	1000ul	300ul
Linearity	1000 U/L	1000 U/L

REFERENCES:

1. Bessey O.A., Lowry O.H. And Brock M.J.: Biol. Chem., 164,321,1946.
2. Bowers G.N.Jr. and McComb R.B., Clin. Chem. 12, 70, 1966.
3. McComb R.B. and Bowers G.N.Jr.: Clin. Chem. 18, 97, 1972.
4. Z. Klin. Chem. Klin. Biochem. 8, 658 (1970); 9, 464 (1971); 10, 182 (1972).
5. Kubler W.: Symp. D. Deutschen Ges. fur Lab. Med. Mainz (1973).
6. Thomas L.: Labor. u. Diag. 1 Aufl., p. 64. Die Med. Verlag, Marburg (1978).
7. Rick W.: Klinische Chemie und Mikroskopie, p. 294, 6th edition, Springer Verlag, Berlin (1990).

IVD

Marketed By:
SPHERIX DIAGNOSTICS
OFFICE NO 131, S.NO.17/1A/2
PALLADIUM GRAND, PH 2,
PUNE CITY, DHANORI
PUNE – 411015, MAHARASHTRA