ORDER INFORMATION

CODE: DL0211 - R1 - 2 X 20 ML + R2 - 2 x 5 ML DL0212 - R1 - 4 X 20 ML + R2 - 4 x 5 ML

DELTA Alkaline Phosphatase

(PNPP/AMP)

SAFETY PRECAUTIONS AND WARNINGS:

This reagent is for In vitro diagnostic use only.

INTENDED USE:

This reagent kit is intended for "in vitro" quantitative determination of Alkaline phosphatase activity in serum.

CLINICAL SIGNIFICANCE:

Alkaline phosphatase is a membrane-bound enzyme which is present in most tissues. It has three different isoenzymes derived from small intestine - placenta - bone / liver / kidney. It is a dimer molecule containing Zn⁺⁺ ions, which play a role in the maintenance of structure and catalysis. The enzyme found in human serum is derived from bone, liver and small intestine. During pregnancy the enzyme from the placenta dominates (it is heat stable at 65°C). In the past the iso-enzymes were separated using various inhibitors and heat. The role of electrophoresis is growing in determining the concentrations. The increase in enzyme activity is prevalent in various hepatic and bone decrease states. The level is also increased in certain diseases of the thyroid gland, intestinal tract and in several bacterial infections.

PRINCIPLE:

p-Nitrophenyl phosphate is converted to p-nitrophenol and phosphate by alkaline phosphatase. The increase of absorption at 405 nm is proportional to the alkaline phosphatase concentration in the sample.

REAGENT COMPOSITION:

Reagent 1: AMP Buffer Reagent.

Reagent 2: Substrate (p-Nitrophenyl Phosphate)

MATERIALS REQUIRED BUT NOT PROVIDED:

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

SAMPLES:

Serum free of hemolysis.

WORKING REAGENT PREPARATION & STABILITY:

Mix 4 volumes of Reagent 1, with 1 Volume of reagent 2. Working reagent is stable for 30 days at 2°-8°C.

GENERAL SYSTEM PARAMETERS:

Reaction type Kinetic Reaction (Increasing)

Wave length 405 nm Light Path 1Cm Reaction Temperature 37°C

Blank / Zero Setting With Distilled Water

Reagent Volume 1_ml Sample Volume 20 µl Lag / Delay Time 60 Sec. Read Time 90 Sec. Interval Time 30 Sec. Factor 2720 Linearity 1600 U/I

ASSAY PROCEDURE:

Working Reagent	1000 μ Ι
Sample	20 μl

Mix and after 60 second incubation, measure the change in absorbance every 30 seconds for 90 seconds at 37°C.

Determine the $\Delta A/min$.

CALCULATION:

Alkaline Phosphatase Activity (U/I) = $\Delta A/min. \times 2720$

LINEARITY:

Reagent is Linear up to 1600 U/I

Dilute the sample appropriately and re-assay if Alkaline Phosphatase Activity exceeds 1600 U/I or Δ Abs / min Exceeds 0.588. Multiply result with dilution factor.

REFERENCE NORMAL VALUE:

Children: 104-390 U/I Adults : 25 - 140 U/I

The reference values are only indicative in nature. Every laboratory should establish its own normal ranges.

QUALITY CONTROL:

For accuracy it is necessary to run known controls with every assay.

LIMITATION & PRECAUTIONS:

- 1. Storage conditions as mentioned on the kit to be adhered.
- Do not freeze or expose the reagents to higher temperature as it may affect the performance of the kit.
- 3. Before the assay bring all the reagents to room temperature.
- 4. Avoid contamination of the reagent during assay process.
- Use clean glassware free from dust or debris.
- Reagent to sample ratio as mentioned here above must be strictly observed as any change in to it will effect the factor.

BIBLIOGRAPHY:

Fundamental of Clinical Chemistry, Young D.S, Tietz, N. Fundamentals of Clinical Chemistry 602/609, Kaplan, M.M.New England.