ANA BIO CLR PHOSPHORUS

(UV - End Point Method)

Intended Use

Phosphorus is a reagent kit used for the quantitative determination of Inorganic phosphorus in serum based on UV - End Point method using Ammonium molybdate. The reagents are for *in-vitro* diagnostic use.

Principle

Inorganic phosphorus reacts with Ammonium molybdate in strong acidic medium to form Phospho-Inorganic Molybdate complex.

The absorbance of this complex is directly proportional to the phosphorus concentration.

Phosphorus + Ammonium molybdate $\xrightarrow{\text{Acid pH}}$ Phospho-Inorganic Molybdate complex

Reagents provided

1. Molybdate Reagent.
2. Standard - Phosphorus (5 mg/dL).

Reagent storage and stability

Molybdate reagent and standard are stable till the expiry date stated on the container label.

Specimen collection and preservation

Blood should be collected in clean dry container. Neatly separated serum should be used. Plasma is not recommended as anticoagulants may cause false low results.

Phosphorus is stable for 7 days in neatly separated serum. If the estimation is not possible within 7 days then the specimen should be preserved at -10°C and should be used within 3 weeks.

Assay guidelines for Analyzers

Reaction type	UV - End point
Reaction time	5 minutes at 37°C
Wavelength	340 nm
Zero setting with	Reagent Blank
Blank absorbance limit	< 0.300 Abs.
Sample volume	10 µl (0.01 ml)
Reagent volume	1000 µl (1.0 ml)
Standard concentration	5 mg/dL
Linearity	20 mg/dL

Assay guidelines for Manual procedure

Bring the reagent and standard to room temperature before performing the assay.

Reagents	Blank	Standard	Sample
Molybdate Reagent	1000 µl (1.0 ml)	1000 µl (1.0 ml)	1000 µl (1.0 ml)
Standard	-	10 µl (0.01 ml)	-
Sample	-	-	10 µl (0.01 ml)

1. Mix thoroughly and incubate at 37°C for 5 minutes.
2. Read the absorbance of specimen and standard against blank at 340 nm.
3. The final colour is stable for two hours if not exposed to direct light.

Calculation

$$\text{Inorganic phosphorus in mg/dL} = \frac{\text{Absorbance of Sample}}{\text{Absorbance of Standard}} \times 5$$

Note: The specimen to working reagent ratio can be altered proportionately without affecting the results.

Normal Range

Adults	:	2.5 - 5.0 mg/dL
Children	:	4.0 - 7.0 mg/dL

Limitation

- Discard the working reagent if the absorbance of the same is more than 0.300 against distilled water at 340 nm.
- If the phosphorus value exceeds linearity limit then dilute the specimen suitably with normal saline and repeat the assay. In such case the assay value should be multiplied by the dilution factor to obtain correct phosphorus value of the specimen.
- Strong lipemic and hemolytic sera should not be used.
- Contaminated glassware is the greatest source of error. Disposable plastic tubes and clean glasswares are recommended for the test.
- The reagent contains sulphuric acid. Avoid contact with skin and mucous membrane. If you come in contact with the reagent wash thoroughly with water.

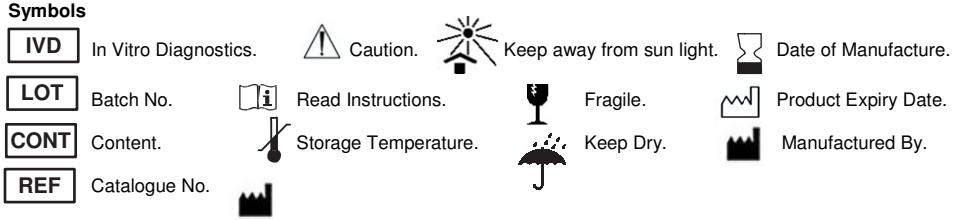
Quality Control

To ensure adequate quality control, it is recommended that each assay should include a normal and an abnormal commercial reference control serum. It should be realized that the use of quality control material checks both instrument and reagent functions together. Factors which might effect the performance of this test include proper instrument function, temperature control, cleanliness of glassware and accuracy of pipetting.

References

1. Daly, J.A., Clin. Chem., 18: 263, 1972.
2. Gamst, O. and Try, K., Scand. J. Clin. Lab. Invest. 40 1980.
3. Amador, E. and Urban, J; Clin. Chem. 18, 60, 1977.

Symbols



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