PHOSPHORUS UV

Ammonium molybdate method

Liquid Reagent ready to use

REF. 0031	**2x100 ml**
REF. 0039	**4x100 ml**
REF. 0041	**4x250 ml**

CE IVD

Azienda certificata DNV

GIESSE DIAGNOSTICS

INTENDED USE

Quantitative determination of phosphorus in serum, plasma, urine.

PRINCIPLE

The phosphate ions react with ammonium molybdate to form a phosphomolybdate complex whose absorbance at 340 nm is proportional to the phosphorus quantity in the sample.

An acid pH is necessary for the formation of complexes.

SAMPLE

Serum is the preferred specimen. Although heparinized plasma is acceptable, levels of inorganic phosphate are about 0.2 to 0.3 mg/dl lower than in serum.

Anticoagulants such as citrate, oxlate, and EDTA interfere with formation of the phosphomolybdate complex and should not be used.

It is important to promptly separate serum or plasma from erythrocytes. Hemolyzed specimens are unacceptable because erythrocytes contain high concentrations of organic phosphate esters, which can be hydrolyzed to inorganic phosphate during storage. Inorganic phosphate increases by 4 to 5 mg/dl per day in hemolyzed specimens stored at 2-8 °C.

Phosphate is considered to be stable in serum that has been separated from the clot for days at 2-8 °C and months when frozen.

Urine samples should be collected in 6 mol/l HCl, 20-30 ml for a 24 hours specimen, to avoid precipitation of phosphate complexes.

Dilute urine samples 1:10 with distilled water before assay.

KIT COMPONENTS

Reagent (A) Volume = 100/250 ml	Ammonium molybdate Sulfuric acid 96 %	0.5 mmol/l 150 mmol/l
Standard Volume = 10 ml	Phosphorus Sodium azide	**5 mg/dl** (1.615 mmol/l) 4 mmol/l

The reagents are stable until the expiration date indicated on the label if stored at 2-8°C and protected from light. Do not freeze. Once opened reagents are stable for 2 months at 2-8°C if contamination is avoided.

Keep bottles closed when not in use.

REAGENT PREPARATION

Liquid Reagent, bring to room temperature (15-25°C) before use.

A yellow color (O.D. > 0.100) indicates that the reagent is contaminated.

PRECAUTIONS AND WARNINGS

Reagent may contain some non-reactive and preservative components. It is suggested to handle carefully it, avoiding contact with skin and swallow.

Use the normal precautions required in the laboratory.

Dispose of waste according to local laws.

PROCEDURE

Wavelength:	340 nm
Lightpath :	1 cm
Temperature:	25, 30, 37°C
Reading:	against blank reagent
Method:	Increasing End Point
Sample/Reagent:	1/100

pipette:	**blank**	**sample**	**standard**
Reagent (A)	1000 µl	1000 µl	1000 µl
water	10 µl		
sample		10 µl	
standard			10 µl

Mix, incubate at 25, 30, 37 °C for 5 minutes, read against blank reagent the absorbance of the sample (Ax) and the standard (As).

Reaction volumes can be proportionally varied.

This method describes the manual procedure to use the kit.

For automated procedure, ask for specific applications.

RESULTS CALCULATION

Serum/plasma:

Phosphorus mg/dl = Ax/As x 5 (standard Value)

24 hours Urine:

Phosphorus mg/24h = Ax/As x 5 x 10 (dilution) x urine Volume (in dl)

Conversion factor : mg/dl x 0.323 = mmol/l

EXPECTED VALUES

Serum/plasma

Adults:	**2.5 – 4.5 mg/dl**	**(0.81 – 1.45 mmol/l)**
Children:	**4.0 – 7.0 mg/dl**	**(1.29 – 2.26 mmol/l)**

Urine

Adults:	**400 – 1300 mg/24h**	**(12.9 – 42.2 mmol/24h)**

Each laboratory should establish appropriate reference intervals related to its population.

QUALITY CONTROL

You must perform the controls at each kit's use and verify that the values obtained are within the reference range reported in the operating instructions. For this purpose we recommend the use of control sera: PRECISENORM (REF.6000) and PRECISEPATH (REF.6001).

PERFORMANCE

Sensitivity: the sensitivity of the method is: 0.3 mg/dl.

Linearity: the method is linear up to 20 mg/dl. For higher values, dilute the sample 1:2 and multiply the result by 2.

Precision intra-assay:

	Level 1	Level 2
Mean (g/dl)	4.08	7.10
DS	0.16	0.13
CV %	3.92	1.83

Precision inter-assay:

	Level 1	Level 2
Mean (g/dl)	4.07	6.81
DS	0.09	0.15
CV %	2.21	2.20

Interferences: triglycerides do not interfere up to 500 mg/dl. Glucose up to 600 mg/dl does not interfere. Albumin does not interfere up to 20 g/dl. Bilirubin at a concentration equal or greater than 12 mg/dl interfere. Hemoglobin from 0.15 g/dl interferes. Hemoglobin and bilirubin in the above concentrations involves an increase of inorganic phosphorus by about 10 %.

Correlation against a reference method: Y = 0.9843x + 0.0742 r = 0.9986

REFERENCES

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