



PHOSPHORUS U.V.

Determination of inorganic Phosphorus in body fluids

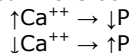


ORDER INFORMATION

REF	Kit size
GA4565 00	5x50 ml
KL4565 00	6x60 ml
BK4565 00	4x60 ml

INDICATION

Most Phosphorus is found in the body in the bone matrix and is excreted in the urine. Levels of Calcium and Phosphorus are closely linked because they are both deposited in the bone together.



Increased values of phosphorus are found in advanced renal insufficiency, pseudo hypoparathyroidism, hypervitaminosis D, and with patients who have hypersecretion of growth hormone. Decreased values are found in hyperparathyroidism, rickets, steatorrhea, and in some renal diseases.

METHOD PRINCIPLE

Inorganic phosphorus reacts, in acid solution, with ammonium molybdate forming a complex which absorbs at ultraviolet field. The intensity of absorption is proportional to inorganic phosphorus concentration present in the sample.

COMPOSITION

REAGENT A (liquid):

Ammonium molybdate	0.5 mmol/l
Nitric acid	200 mmol/l
Surfactant and preservatives.	

STANDARD (liquid):

	1x5 ml
Inorganic phosphorus	4 mg/dl
Verified against NIST reference material.	

Storage and stability

Store at room temperature (15-25 °C). Do not freeze the reagents! The reagents are stable up to the expiry date stated on the label, if contamination and evaporation are avoided, protected from light. The above conditions are valid if the vials are opened just only for the time to take the reagent, closed immediately with their cap and stored at the indicated conservation temperature.

ANCILLARY EQUIPMENT

- Automatic pipettes
- Photometer
- Analysis cuvettes (optical path = 1 cm)
- NaCl solution 9 g/l

SAMPLES

Serum, plasma or urine 24h.

Do not use haemolysed samples. Immediately separate the serum from cells.

Urine 24h: diluted 1:10 with distilled water.

Stable 7 days at 2-8 °C.

Specimen collection / Preanalytical factors

It is recommended that specimen collection should be carried out in accordance with NCCLS Document H11-A3.

INTERNAL QUALITY CONTROL

It is recommended to use commercial Quality Control sera with known Phosphorus concentration. Check that the values obtained are within the reference range provided.

ANALYTICAL PROCEDURE

Allow the reagents to reach working temperature before using.

Pipette into disposable or well clean cuvettes:

	Blank	Standard	Sample
Reagent A	1000 µl	1000 µl	1000 µl
Distilled H ₂ O	10 µl	-	-
Standard	-	10 µl	-
Sample	-	-	10 µl

Mix and incubate for **2 minutes** at **room temperature** (20-25 °C). Read the absorbance (A) of the standard and of the samples against Blank at **340 nm**.
The colour is stable for 30 minutes.

CALCULATION OF RESULTS

Serum-plasma:

$$\text{Phosphorus, mg/dl} = \frac{A \text{ sample}}{A \text{ standard}} \times 4$$

Urine:

$$\text{Phosphorus, g/24h} = \frac{A \text{ sample}}{A \text{ standard}} \times 0.4 \times 1/24\text{h}$$

Conversion factor

Phosphorus [mg/dl] x 0.58 = Phosphorus [mEq/l]

Phosphorus [mg/dl] x 0.32 = Phosphorus [mmol/l]

REFERENCE VALUES

Sample	Subjects	Range	Units
Serum	adults	2.3 ÷ 4.7	mg/dl
	adults	0.74 ÷ 1.52	mmol/l
	children	4.0 ÷ 7.0	mg/dl
	children	1.29 ÷ 2.26	mmol/l
Urine		0.9 ÷ 1.3	g/24h

Each laboratory should establish reference ranges for its own patients population.

ANALYTICAL PERFORMANCES

Precision

Within-run and between-run coefficients of variation have been calculated on replicates of three samples at different Phosphorus concentrations. The obtained results are reported in the following tables:

Sample	Mean (mg/dl)	Within Run		Between Run	
		SD	%CV	SD	%CV
Serum 1	4.02	0.03	0.8	0.13	3.1
Serum 2	5.08	0.03	0.6	0.17	3.4
Serum 3	6.16	0.04	0.6	0.18	2.9

Linearity

The assay is linear up to 25 mg/dl.

Sensitivity

Test sensitivity, in terms of limit of detection, is 0.25 mg/dl.

Correlation

A correlation study comparing the present method and a commercial one gave the following results:

$$y = 0.9189x + 0.1764 \text{ mg/dl} \quad r = 0.9738$$

Interferences

Hemoglobin Interfere, do not use hemolysed sera
Bilirubin > 20 mg/dl
Triglycerides > 1000 mg/dl

PRECAUTIONS IN USE

The reagents contain inactive components such as preservatives (Sodium azide or others), surfactants etc. The total concentration of these components is lower than the limits reported by 67/548/EEC and 88/379/EEC directives about classification, packaging and labelling of dangerous substances. However, the reagents should be handled with caution, avoiding swallowing and contact with skin, eyes and mucous membranes. The use of laboratory reagents according to good laboratory practice is recommended.

Waste Management

Please refer to local legal requirements.

BIBLIOGRAPHY

1. MUNOZ M. A., Clin. Chem. 29/2, 372-374 (1983)
2. DALY JOHN A. , Clin. Chem. 18/3, 1972
3. EU-Dir 1999/11 Commission Directive of 8 March 1999 adapting to technical progress the principles of good laboratory practice as specified in Council Directive 87/18/EEC.
4. NCCLS Document, "Procedures for the collection of arterial blood specimens", Appr. Std., 3rd Ed. (1999).