

**Quantitative determination of Phosphorus in human Serum / Plasma / other body fluids. Only for *In Vitro* Diagnostic use**

#### ORDER INFORMATION

EF	Pack Size
PHOMONO 25	25 X 1 ml
PHOMONO 50	50 X 1 ml
PHO 25	1 X 25 ml
PHO 50	1 X 50 ml
PHO 100	1 X 100 ml
PHO 1000	1 X 1000 ml
PHO 5000	1X5000 ml
PHO 10000	1X10000 ml

#### CLINICAL SIGNIFICANCE

Phosphorus is an essential mineral for tissue bone formation and is required by every cell in the body for normal function. Approximately 85% of the body phosphorus is found in bone and in teeth. Low levels of phosphorus can be caused by hypervitaminosis D, primary hyperparathyroidism, renal tubular disorders, antacids or malabsorption. High levels of phosphorus can be caused by diet, bone metastases, liver disease, alcohol ingestion, diarrhea and vomiting<sup>1,5,6</sup>. Clinical diagnosis should not be made on a single test result; it should integrate clinical and other laboratory data.

#### Method

Photometric UV test with endpoint determination.

#### PRINCIPLE

Inorganic phosphate reacts in acid environment with molybdic acid to form an unreduced phosphomolybdic acid complex, which absorbs light at 340 nm. The absorbance is directly proportional to the phosphorus concentration in the sample.

#### REAGENT

Reagent : Molybdate reagent  
Phosphorus standard : 5.0 mg/dl

#### REAGENT PREPARATION

The Reagent is ready to use.

#### REAGENT STORAGE AND STABILITY

Reagent is stable till expiry when stored at RT  
Store protected from light.

#### WARNING AND PRECAUTIONS

- For in vitro diagnostic use.
- Do not use components beyond the expiration date.
- Do not mix materials from different kit lot numbers.
- Exercise the normal precautions required for handling all laboratory reagents.
- The reagent contains preservative. Do not swallow. Avoid contact with skin and mucous membranes.
- For detailed information refer Material Safety Data Sheet.
- Proceed carefully with this product because due to its nature it can get contaminated easily.
- Most of the detergents and water softening products used in the laboratories contain chelating agents. A defective rinsing will invalidate the procedure.

#### WASTE MANAGEMENT

Please refer to local legal requirements.

#### MATERIALS REQUIRED BUT NOT PROVIDED

- NaCl solution 9 g/L
- General laboratory equipment

#### SAMPLE COLLECTION AND PRESERVATION

##### Serum or plasma (lithium heparin)

Separate from cellular contents immediately after blood collection.  
Stability: at least one year at -20°C in case of immediate freezing.

7 days at 4 – 8°C

Freeze only once! Discard contaminated specimens!

**Urine:** Collect 24-hour urine specimen in Phosphorus free containers. Dilute a sample 1/2 in distilled water. Mix. Multiply results by 2 (dilution factor).

Stability of the sample:

1 week at refrigerator (2-8°C) or frozen (-20°C) temperatures.

#### ASSAY PROCEDURE

##### Operating Instructions

- Check reagent inventories at least daily to ensure that quantities are sufficient for the planned work load.
- Bring all reagents, standard and samples to room temperature 18 - 28°C, prior to analysis.

AUTOMATED PARAMETERS	
Wavelength	340 nm
Cuvette	1 cm light path
Temperature	37 °C
Measurement	Against Reagent Blank
Sample Volume	20 µl
Reagent Volume	1000 µl
Reaction Type	End Point
Incubation	5 mins.
Blank Absorbance Limit	< 0.270
Low Normal	2.5 mg/dl
High Normal	4.5 mg/dl
Linearity	12.0 mg/dl

#### MANUAL ASSAY PROCEDURE

##### Pipette into Test Tubes

	BLANK	STD	SAMPLE
Sample	-	-	20µl
Standard	-	20µl	-
Reagent	1000µl	1000µl	1000µl

Mix well incubates at 37 °C for 5 mins. Measure final absorbance of the sample (Ac) and standard (As) against the reagent blank.

#### SAMPLE DILUTIONS

- This method is linear upto a concentration of 12 mg/dL.
- Dilute samples above this concentration 1:1 with DI Water
- Repeat assay. Multiply the result by 2.

#### CALCULATION

Results are calculated, usually automatically by the instrument, as follows:

Phosphorus Conc. in =  $\frac{\text{Abs Sample} \times \text{Concentration of Standard}}{\text{Serum/Plasma (mg/dL) Abs Standard}}$

Phosphorus Conc. in =  $\frac{\text{Abs Sample} \times \text{Conc. of Standard} \times 2}{\text{Urine (mg/dL) Abs Standard}}$

#### CLIBRATORS AND CONTROLS

For the calibration of automated photometric systems the commercially available suitable multi-calibrator is recommended.

The assigned values of **Phosphorus standard** have been made traceable to a primary phosphorus standard (traceable to the reference material NERL / Weighed purified material).

It is recommended to run a normal and a pathological control serum which is commercially available to verify the performance of the measured procedure. The value of controls should fall within the established limit. Each laboratory should establish corrective action in case of deviations in control recovery.

**PERFORMANCE CHARACTERISTICS  
WITHIN RUN**

Sample	Mean Concentration	SD	CV %
Randox 2	4.14	0.13	3.15%
Randox 3	7.17	0.19	2.16%

**RUN TO RUN**

Sample	Mean Concentration	SD	CV %
Randox 2	4.10	0.09	2.24%
Randox 3	7.36	0.23	3.16%

**LINEARITY**

This method is linear upto a concentration of 12 mg/dL.  
Dilute samples above this concentration 1:1 with DI Water and  
Repeat assay. Multiply the result by 2.

**Limit of detection:** The limit of detection for Phosphorus is 0.2 mg/dL (0.065 mmol/L).

**METHOD COMPARISON**

A comparison of Phosphorus with a commercially available assay (x) using 59 samples gave following results:  $R^2 = 0.9400$

**REFERENCE VALUES**

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: 0.4 – 1.3 g/24 Hr. Urine / 12.9 – 42 mmol/24 Hr.  
The reference values are to be considered as indicative only. Every laboratory should establish its own normal range.

**LIMITATION OF THE PROCEDURE**

- For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.






**INTERFERENCE**

- Bilirubin: No interference found upto Bilirubin 25mg/dl.
- Hemoglobin: No interference found upto 100mg/dl.
- Both positive and negative interferences with lipemic samples has been observed
- These characteristics have been obtained using an automatic analyzer. Results may vary if a different instrument or a manual procedure is used.

**BIBLIOGRAPHY**

- Thomas L. Clinical Laboratory Diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft; 1998. p. 241-7.
- Endres DB, Rude RK. Mineral and bone metabolism. In: Burtis CA, Ashwood ER, editors. Tietz Textbook of Clinical Chemistry. 3rd ed. Philadelphia: W.B Saunders Company; 1999. p. 1395-1457.  
Burtis CA, Ashwood ER, Bruns DE. Tietz Textbook of Clinical Chemistry. 4th ed. Elsevier Saunders; 2006. p. 1908.

**GLOSSARY OF SYMBOL**

	Consult Instruction for Use
	Catalog Number
	Store between
	Manufacturer
	Keep away from sunlight



Paramcare Life Sciences Private Limited, G/F-12/13, Evershine-2,  
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