# PHOSPHORUS UV

| | |
|---|---|
| PH 0100 CH | 2 x 50 ml |
| PH 0500 CH | 4 x 125 ml |

## INTENDED USE

Reagent for quantitative in vitro determination of phosphorus in biological fluids.

## SUMMARY OF TEST

Phosphorus in the form of inorganic or organic phosphate is an important and widely distributed element in the human body. An adult human has approximately 600 g of phosphate expressed as phosphorus, of which about 85% is in the skeleton and the rest principally in soft tissues. In the soft tissues, most phosphate is cellular.

## PRINCIPLE OF THE METHOD

The phosphate ions react with ammonium molybdate to form a phosphomolybdate complex. The colourless phosphomolybdate complex can be measured directly by ultraviolet (UV) absorption at 340 nm. An acid pH is necessary for the formation of complexes.

## KIT COMPONENTS

**For in vitro diagnostic use only.**
The components of the kit are stable until expiration date on the label.
Keep away from direct light sources.

**PHOS R1    0100:    2 x 50 ml (liquid) blue cap**
**                      0500:    4 x 125 ml (liquid) blue cap**

Composition: ammonium molibdate 0.4 mmol/l, sulphuric acid 0.21 mol/l, surfactant.

**Standard:        inorganic phosphorus 5 mg/dl - 5 ml**

Store all components at 2-8°C.

## MATERIALS REQUIRED BUT NOT SUPPLIED

Current laboratory instrumentation. Spectrophotometer UV/VIS with thermostatic cuvette holder. Automatic micropipettes. Glass or high quality polystyrene cuvettes. Saline solution.

## REAGENT PREPARATION

Use reagent ready to use.
Stability: up to expiration date on labels at 2-8°C.
Stability since first opening of vials: preferably within 60 days at 2-8°C.

## PRECAUTIONS

Reagent may contain some non-reactive and preservative components. It is suggested to handle carefully it, avoiding contact with skin and swallow.
Perform the test according to the general "Good Laboratory Practice" (GLP) guidelines.

## SPECIMEN

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, oxalate, and EDTA interfere with formation of the phosphomolybdate complex and should not be used.
Inorganic phosphate in whole blood specimens may either decrease or increase with time, depending on the type of specimen, temperature, and duration of storage. Levels in plasma or serum are increased by prolonged storage with cells at room temperature or 37 °C; it is important to promptly separate serum or plasma from erythyrocytes. 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 4°C. Glucose phosphate, creatine phosphate, and other organic phosphates may also be hydrolyzed by assay conditions, resulting in overestimation of inorganic phosphate levels.
Phosphate is considered to be stable in serum that has been separated from the clot for days at 4°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 1:20 with purified water before assay.

## TEST PROCEDURE

| | |
|---|---|
| Wavelenght: | 340 nm |
| Lightpath: | 1 cm |
| Temperature: | 25, 30 or 37°C |

| dispense: | blank | standard | sample |
|---|---|---|---|
| reagent | 1 ml | 1 ml | 1 ml |
| water | 10 μl | - | - |
| standard | - | 10 μl | - |
| sample | - | - | 10 μl |

Mix, incubate at 25, 30 or 37°C for 5 minutes.
Read absorbances of standard (As) and samples (Ax) against reagent blank.

## RESULTS CALCULATION

serum/plasma sample:

phosphorus mg/dl = Ax/As x 5 (standard value)

random urine sample:

phosphorus mg/dl = Ax/As x 5 x 20 (standard value and dilution factor)

24 hours urine sample:

$$\text{phosphorus g/24h} = \frac{\text{Ax/As x 5 x 20 x urine volume}}{1000}$$

(standard value, dilution factor and diuresis in decilitres)

## EXPECTED VALUES

serum/plasma (adults):   2.5 - 4.5 mg/dl (0.81 - 1.45 mmol/l)
serum/plasma (children): 4.0 - 7.0 mg/dl (1.29 - 2.26 mmol/l)

urine (nonrestricted diet):   0.4 - 1.3 g/24h (12.9 - 42.2 mmol/24h)

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

## QUALITY CONTROL AND CALIBRATION

It is suggested to perform an internal quality control. For this purpose the  following human based control sera are available:
**QUANTINORM CHEMA**
with normal or close to normal control values
**QUANTIPATH CHEMA**
with pathological control values.
If required, a multiparametric, human based calibrator is available:
**AUTOCAL H**

Please contact Customer Care for further information.

## TEST PERFORMANCE

**Linearity**
the method is linear up to 20 mg/dl.
If the limit value is exceeded, it is suggested to dilute sample 1+9 with distilled water and to repeat the test, multiplying the result by 10.

**Sensitivity/limit of detection (LOD)**
the limit of detection is 0.4 mg/dl.

**Interferences**
no interference was observed by the presence of:
bilirubin                 ≤ 25 mg/dl
hemoglobin             ≤ 100 mg/dl
Hemolysis interferes.
Both positive and negative interferences with lipemic samples has been observed.

**Precision**

| intra-assay (n=10) | mean (mg/dl) | SD (mg/dl) | CV% |
|---|---|---|---|
| sample 1 | 3.50 | 0.05 | 1.50 |
| sample 2 | 5.87 | 0.11 | 1.90 |

| inter-assay (n=20) | mean (mg/dl) | SD (mg/dl) | CV% |
|---|---|---|---|
| sample 1 | 3.41 | 0.08 | 2.40 |
| sample 2 | 5.84 | 0.11 | 1.90 |

**Methods comparison**
a comparison between Chema and a commercially available product gave the following results:

phosphorus  Chema = x
phosphorus competitor = y
n = 102

y = 1.005x - 0.109 mg/dl          $r^2$ = 0.975

## WASTE DISPOSAL

This product is made to be used in professional laboratories.
P501: Dispose of contents according to national/international regulations.

## REFERENCES

Yee H.Y. - Clin. Chem. 14, 898 (1968).
Tietz Textbook of Clinical Chemistry, Second Edition, Burtis-Ashwood (1994).

## MANUFACTURER

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## SYMBOLS

| | | |
|---|---|---|
| | **IVD** | *in vitro* diagnostic medical device |
| | **LOT** | batch code |
| | **REF** | catalogue number |
| | | temperature limit |
| | | use by date |
| | | caution |
| | | consult instructions for use |

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