PHOS2

cobas®

Phosphate (Inorganic) ver.2

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
03183793 122	Phosphate (Inorganic) ver.2 (250 tests)	System-ID 07 6614 3	Roche/Hitachi **cobas c** 311, **cobas c** 501/502

Materials required (but not provided):

10759350 190	Calibrator f.a.s. (12 x 3 mL)	Code 401	
10759350 360	Calibrator f.a.s. (12 x 3 mL, for USA)	Code 401	
12149435 122	Precinorm U plus (10 x 3 mL)	Code 300	
12149435 160	Precinorm U plus (10 x 3 mL, for USA)	Code 300	
12149443 122	Precipath U plus (10 x 3 mL)	Code 301	
12149443 160	Precipath U plus (10 x 3 mL, for USA)	Code 301	
05117003 190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 391	
05947626 190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 391	
05947626 160	PreciControl ClinChem Multi 1 (4 x 5 mL, for USA)	Code 391	
05117216 190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 392	
05947774 190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 392	
05947774 160	PreciControl ClinChem Multi 2 (4 x 5 mL, for USA)	Code 392	
04489357 190	Diluent NaCl 9 % (50 mL)	System-ID 07 6869 3	

English

System information

For **cobas c** 311 analyzer:

PHOS2: ACN 714 (serum/plasma)

SPHO2: ACN 675 (STAT, reaction time: 7: serum/plasma)

PHO2U: ACN 716 (urine)

SPH2U: ACN 656 (STAT, reaction time: 7: urine)

For **cobas c** 501 analyzer:

PHOS2: ACN 714 (serum/plasma/urine)

SPHO2: ACN 675 (STAT, reaction time: 7: serum/plasma/urine)

For **cobas c** 502 analyzer:

PHOS2: ACN 8714 (serum/plasma)

SPHO2: ACN 8675 (STAT, reaction time: 7: serum/plasma)

PHO2U: ACN 8716 (urine)

SPH2U: ACN 8656 (STAT, reaction time: 7: urine)

Intended use

In vitro test for the quantitative determination of phosphorus in human serum, plasma and urine on Roche/Hitachi **cobas c** systems.

Summary[1,2,3,4,5]

88 % of the phosphorus contained in the body is localized in bone in the form of calcium phosphate as the apatite $Ca^{2+}[Ca_3(PO_4)_2]_3^{2-}$. The remainder is involved in intermediary carbohydrate metabolism and in physiologically important substances such as phospholipids, nucleic acids and ATP. Phosphorus occurs in blood in the form of inorganic phosphate and in organically bound phosphoric acid. The small amount of extracellular organic phosphorus is found almost exclusively in the form of phospholipids.

The ratio of phosphate to calcium in the blood is approximately 6:10. An increase in the level of phosphorus causes a decrease in the calcium level. The mechanism is influenced by interactions between parathormone and vitamin D. Hypoparathyroidism, vitamin D intoxication and renal failure with decreased glomerular phosphate filtration give rise to hyperphosphatemia. Hypophosphatemia occurs in rickets, hyperparathyroidism and Fanconi's syndrome.

The preferred method for the determination of inorganic phosphorus is based on the formation of ammonium phosphomolybdate with subsequent reduction to molybdenum blue. Reagent stability problems often occur with this method. The method presented here is based on the reaction of phosphate with ammonium molybdate to form ammonium phosphomolybdate without reduction. The addition of an accelerator gives rise to a more rapid rate of reaction and the application of sample blanking yields more precise results.

Test principle[5]

Molybdate UV.

Inorganic phosphate forms an ammonium phosphomolybdate complex having the formula $(NH_4)_3[PO_4(MoO_3)_{12}]$ with ammonium molybdate in the presence of sulfuric acid.

$$\text{Phosphate} + \text{ammonium molybdate} \xrightarrow{H_2SO_4} \text{ammonium phosphomolybdate}$$

The concentration of phosphomolybdate formed is directly proportional to the inorganic phosphate concentration and is measured photometrically.

Reagents - working solutions

R1 Sulfuric acid: 0.36 mol/L; detergent

R2 Ammonium molybdate: 3.5 mmol/L; sulfuric acid: 0.36 mol/L; sodium chloride: 150 mmol/L

R1 is in position B and R2 is in position C.

Precautions and warnings

For in vitro diagnostic use for health care professionals. Exercise the normal precautions required for handling all laboratory reagents.

Infectious or microbial waste:
Warning: handle waste as potentially biohazardous material. Dispose of waste according to accepted laboratory instructions and procedures.

Environmental hazards:
Apply all relevant local disposal regulations to determine the safe disposal.

Safety data sheet available for professional user on request.

For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

Warning

H290 May be corrosive to metals.

H412 Harmful to aquatic life with long lasting effects.

Prevention:

P234 Keep only in original packaging.

P273	Avoid release to the environment.

Response:

P390	Absorb spillage to prevent material damage.

Disposal:

P501	Dispose of contents/container to an approved waste disposal plant.

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590, USA: 1-800-428-2336

Reagent handling

Ready for use

Storage and stability

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Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	12 weeks

Diluent NaCl 9 %

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	12 weeks

Specimen collection and preparation

For specimen collection and preparation only use suitable tubes or collection containers.

Only the specimens listed below were tested and found acceptable.
Serum
Plasma: Li-heparin and K_2-EDTA plasma

The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.

Urine

Collect in detergent-free containers. Acidify with hydrochloric acid after collection (pH < 3).[6,7]

Stability in serum/plasma:[8]	24 hours at 15-25 °C
	4 days at 2-8 °C
	1 year at (-15)-(-25) °C
Stability in urine:[6,7]	6 months at 2-8 °C (when acidified)
24-hour urine:	Store cooled during collection.

Centrifuge samples containing precipitates before performing the assay.

See the limitations and interferences section for details about possible sample interferences.

Sample stability claims were established by experimental data by the manufacturer or based on reference literature and only for the temperatures/time frames as stated in the method sheet. It is the responsibility of the individual laboratory to use all available references and/or its own studies to determine specific stability criteria for its laboratory.

Materials provided

See "Reagents – working solutions" section for reagents.

Materials required (but not provided)

- See "Order information" section
- General laboratory equipment

Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator's manual for analyzer-specific assay instructions.

The performance of applications not validated by Roche is not warranted and must be defined by the user.

Application for serum and plasma

cobas c 311 **test definition**

Assay type	2-Point End	
Reaction time / Assay points	10 / 6-32 (STAT 7 / 6-32)	
Wavelength (sub/main)	700/340 nm	
Reaction direction	Increase	
Units	mmol/L (mg/dL, mg/L)	
Reagent pipetting		Diluent (H_2O)
R1	90 µL	28 µL
R2	38 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.5 µL	–	–
Decreased	12.5 µL	15 µL	135 µL
Increased	2.5 µL	–	–

cobas c 501 **test definition**

Assay type	2-Point End	
Reaction time / Assay points	10 / 10-47 (STAT 7 / 10-47)	
Wavelength (sub/main)	700/340 nm	
Reaction direction	Increase	
Units	mmol/L (mg/dL, mg/L)	
Reagent pipetting		Diluent (H_2O)
R1	90 µL	28 µL
R2	38 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.5 µL	–	–
Decreased	12.5 µL	15 µL	135 µL
Increased	2.5 µL	–	–

cobas c 502 **test definition**

Assay type	2-Point End	
Reaction time / Assay points	10 / 10-47 (STAT 7 / 10-47)	
Wavelength (sub/main)	700/340 nm	
Reaction direction	Increase	
Units	mmol/L (mg/dL, mg/L)	
Reagent pipetting		Diluent (H_2O)
R1	90 µL	28 µL
R2	38 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.5 µL	–	–

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Decreased	12.5 µL	15 µL	135 µL
Increased	5 µL	–	–

Application for urine

cobas c 311 **test definition**

Assay type	2-Point End
Reaction time / Assay points	10 / 6-32 (STAT 7 / 6-32)
Wavelength (sub/main)	700/340 nm
Reaction direction	Increase
Units	mmol/L (mg/dL, mg/L)
Reagent pipetting	Diluent (H_2O)
R1	90 µL 28 µL
R2	38 µL –

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.5 µL	15 µL	150 µL
Decreased	2.5 µL	8 µL	168 µL
Increased	2.5 µL	15 µL	150 µL

cobas c 501 **test definition**

Assay type	2-Point End
Reaction time / Assay points	10 / 10-47 (STAT 7 / 10-47)
Wavelength (sub/main)	700/340 nm
Reaction direction	Increase
Units	mmol/L (mg/dL, mg/L)
Reagent pipetting	Diluent (H_2O)
R1	90 µL 28 µL
R2	38 µL –

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.5 µL	15 µL	150 µL
Decreased	2.5 µL	8 µL	168 µL
Increased	2.5 µL	15 µL	150 µL

cobas c 502 **test definition**

Assay type	2-Point End
Reaction time / Assay points	10 / 10-47 (STAT 7 / 10-47)
Wavelength (sub/main)	700/340 nm
Reaction direction	Increase
Units	mmol/L (mg/dL, mg/L)
Reagent pipetting	Diluent (H_2O)
R1	90 µL 28 µL
R2	38 µL –

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.5 µL	15 µL	150 µL
Decreased	2.5 µL	8 µL	168 µL
Increased	5 µL	15 µL	150 µL

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	2-point calibration • after reagent lot change • as required following quality control procedures

Calibration interval may be extended based on acceptable verification of calibration by the laboratory.

Traceability: This method has been standardized against NERL primary reference material.
For USA: This method has been standardized against NIST traceable primary reference material.

Quality control

Serum/plasma

For quality control, use control materials as listed in the "Order information" section.

In addition, other suitable control material can be used.

Urine

Quantitative urine controls are recommended for routine quality control.

The control intervals and limits should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

Calculation

Roche/Hitachi **cobas c** systems automatically calculate the analyte concentration of each sample.

Conversion factors:	mmol/L x 3.10 = mg/dL
	mmol/L x 31 = mg/L
	mg/L x 0.0323 = mmol/L

Limitations - interference[6]

Criterion: Recovery within ± 10 % of initial value at a phosphate concentration of 0.87 mmol/L (2.7 mg/dL).

Serum/plasma

Icterus:[9] No significant interference up to an I index of 40 for conjugated and 60 for unconjugated bilirubin (approximate conjugated bilirubin concentration: 684 µmol/L or 40 mg/dL and approximate unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[9] No significant interference up to an H index of 300 (approximate hemoglobin concentration: 186 µmol/L or 300 mg/dL).

Note: This interference results from inorganic phosphates produced by the action of phosphatases on organic phosphates, both of which are released from the red cells upon hemolysis.

Lipemia (Intralipid):[9] No significant interference up to an L index of 1250. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[10,11]

Exception: Phospholipids contained in liposomal drug formulations (eg AmBisome) may be hydrolyzed in the test due to the acidic reaction pH and thus lead to elevated phosphate results.[12]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[13]

Urine

Drugs: No interference was found at therapeutic concentrations using common drug panels.[11]

Criterion: Recovery within ± 10 % of initial value at a phosphate concentration of 13 mmol/L (40.3 mg/dL).

Urea: No significant interference from urea up to a concentration of 1500 mmol/L (9009 mg/dL).

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

ACTION REQUIRED

Special Wash Programming: The use of special wash steps is mandatory when certain test combinations are run together on Roche/Hitachi **cobas c** systems. The latest version of the carry-over evasion list can be found with the NaOHD-SMS-SmpCln1+2-SCCS Method Sheets. For further instructions refer to the operator's manual. **cobas c** 502 analyzer: All special wash programming necessary for avoiding carry-over is available via the **cobas** link, manual input is required in certain cases.

Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.

Limits and ranges

Measuring range

Serum/plasma

0.10-6.46 mmol/L (0.31-20.0 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:2 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 2.

Urine

1.1-92 mmol/L (3.4-285 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:2 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 2.

Lower limits of measurement

Lower detection limit of the test

Serum/plasma

0.10 mmol/L (0.31 mg/dL)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Urine

1.1 mmol/L (3.4 mg/dL)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Expected values

Serum/plasma

Adults:[14]

0.81-1.45 mmol/L (2.5-4.5 mg/dL)

Children:[15]

Age	*Male*	*Female*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*
1–30 d	1.25–2.25 (3.9–6.9)	1.40–2.50 (4.3–7.7)
1–12 m	1.15–2.15 (3.5–6.6)	1.20–2.10 (3.7–6.5)
1–3 y	1.00–1.95 (3.1–6.0)	1.10–1.95 (3.4–6.0)
4–6 y	1.05–1.80 (3.3–5.6)	1.05–1.80 (3.2–5.5)
7–9 y	0.95–1.75 (3.0–5.4)	1.00–1.80 (3.1–5.5)
10–12 y	1.05–1.85 (3.2–5.7)	1.05–1.70 (3.3–5.3)
13–15 y	0.95–1.65 (2.9–5.1)	0.90–1.55 (2.8–4.8)
16–18 y	0.85–1.60 (2.7–4.9)	0.80–1.55 (2.5–4.8)

Roche has not evaluated reference ranges in a pediatric population.

Urine

1st morning urine[16]	13-44 mmol/L (40-136 mg/dL)
24-hour urine[6]	13-42 mmol/d (0.4-1.3 g/d)

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Specific performance data

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Precision was determined using human samples and controls in an internal protocol.

Serum/plasma:

Repeatability (n = 21), intermediate precision (3 aliquots per run, 1 run per day, 21 days);

Urine:

Repeatability (n = 21), intermediate precision (3 aliquots per run, 1 run per day, 10 days).

The following results were obtained:

Serum/plasma

Repeatability	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
Precinorm U	1.24 (3.84)	0.01 (0.03)	0.7
Precipath U	2.05 (6.36)	0.01 (0.03)	0.6
Human serum 1	2.68 (8.31)	0.02 (0.06)	0.6
Human serum 2	1.56 (4.84)	0.01 (0.03)	0.7
Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
Precinorm U	1.23 (3.81)	0.02 (0.06)	1.4
Precipath U	2.04 (6.32)	0.02 (0.06)	1.2
Human serum 3	2.67 (8.28)	0.04 (0.12)	1.4
Human serum 4	1.55 (4.81)	0.02 (0.06)	1.4

Urine

Repeatability	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
Control Level 1	10.2 (31.6)	0.1 (0.3)	1.4
Control Level 2	19.9 (61.7)	0.2 (0.6)	1.2
Human urine 1	40.9 (127)	0.4 (1)	1.0
Human urine 2	6.25 (19.4)	0.08 (0.2)	1.2
Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L (mg/dL)	*mmol/L (mg/dL)*	*%*
Control Level 1	10.0 (31.0)	0.2 (0.6)	1.6
Control Level 2	19.6 (60.8)	0.3 (0.9)	1.7
Human urine 3	40.4 (125)	0.5 (2)	1.3
Human urine 4	6.23 (19.3)	0.12 (0.4)	2.0

The data obtained on **cobas c** 501 analyzer(s) are representative for **cobas c** 311 analyzer(s).

Method comparison

Inorganic phosphate values for human serum, plasma and urine samples obtained on a Roche/Hitachi **cobas c** 501 analyzer (y) were compared with those determined using the corresponding reagent on a Roche/Hitachi 917 analyzer (x).

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Serum/plasma

Sample size (n) = 150

Passing/Bablok[17]	Linear regression
y = 1.022x + 0.000 mmol/L	y = 1.023x - 0.002 mmol/L
τ = 0.978	r = 1.000

The sample concentrations were between 0.62 and 5.54 mmol/L (1.92 and 17.2 mg/dL).

Urine

Sample size (n) = 145

Passing/Bablok[17]	Linear regression
y = 0.976x - 0.053 mmol/L	y = 0.974x - 0.047 mmol/L
τ = 0.967	r = 0.999

The sample concentrations were between 1.61 and 91.5 mmol/L (4.99 and 284 mg/dL).

The data obtained on **cobas c** 501 analyzer(s) are representative for **cobas c** 311 analyzer(s).

References

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A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the Member State in which the user and/or the patient is established.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see dialog.roche.com for definition of symbols used):

CONTENT	Contents of kit
→	Volume after reconstitution or mixing
GTIN	Global Trade Item Number

FOR US CUSTOMERS ONLY: LIMITED WARRANTY

Roche Diagnostics warrants that this product will meet the specifications stated in the labeling when used in accordance with such labeling and will be free from defects in material and workmanship until the expiration date printed on the label. THIS LIMITED WARRANTY IS IN LIEU OF ANY OTHER WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR PARTICULAR PURPOSE. IN NO EVENT SHALL ROCHE DIAGNOSTICS BE LIABLE FOR INCIDENTAL, INDIRECT, SPECIAL OR CONSEQUENTIAL DAMAGES.

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