08057427500V8.0

CA2

Calcium Gen.2

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057427190*	08057427500	Calcium Gen.2 (1500 tests)	System-ID 2034 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057427214*	08057427500	Calcium Gen.2 (1500 tests)	System-ID 2034 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

10759350190	Calibrator f.a.s. (12 x 3 mL)	Code 20401	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

CA2: ACN 20340 (Serum/plasma)

CA2U: ACN 20341 (Urine)

Intended use

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

Summary

Measurements of calcium, measured with this device, in human serum, plasma and urine, are used in the diagnosis of hypercalcemia/hypercalciuria (such as observed in hyperparathyroidism and cancer, endocrine disorders, inherited hypercalcemia, excessive vitamin D intake, chronic kidney disease) and of hypocalcemia/hypocalciuria (such as observed in hypoparathyroidism, vitamin D or magnesium deficiency, calcium homeostasis bone disease).[1]

Calcium is the most abundant mineral element in the body with about 99 % in the bones primarily as hydroxyapatite. The remaining calcium is distributed between the various tissues and the extracellular fluids where it performs a vital role for many life sustaining processes. Among the extra skeletal functions of calcium are involvement in blood coagulation, neuromuscular conduction, excitability of skeletal and cardiac muscle, enzyme activation, and the preservation of cell membrane integrity and permeability. Urinary calcium results from glomerular filtration of albumin-free plasma calcium and intense calcium reabsorption along the different tubular segments.[2]

Serum calcium levels and hence the body content are controlled by parathyroid hormone (PTH), calcitonin, and vitamin D. An imbalance in any of these modulators leads to alterations of the body and serum calcium levels. Increases in serum PTH or vitamin D are usually associated with hypercalcemia. Increased serum and urine calcium levels may also be observed in multiple myeloma and other neoplastic diseases. Hypocalcemia may be observed e.g. in hypoparathyroidism, nephrosis, and pancreatitis.[1]

Test principle

Calcium ions react with 5-nitro-5'-methyl-BAPTA (NM-BAPTA) under alkaline conditions to form a complex. This complex reacts in the second step with EDTA.

$$Ca^{2+} + \text{NM-BAPTA} \xrightarrow[\text{pH}]{\text{alkaline}} \text{calcium-NM-BAPTA complex}$$

$$\text{calcium-NM-BAPTA complex} + \text{EDTA} \longrightarrow \text{NM-BAPTA} + \text{calcium EDTA complex}$$

The change in absorbance is directly proportional to the calcium concentration and is measured photometrically.

Reagents - working solutions

R1 CAPSO:[a)] 557 mmol/L; NM-BAPTA: 2 mmol/L; pH 10.0; non-reactive surfactant; preservative

R3 EDTA: 7.5 mmol/L; pH 7.3; non-reactive surfactant, preservative

a) 3-[cyclohexylamino]-2-hydroxy-1-propanesulfonic acid

R1 is in position B and R3 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.

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

Danger

H318 Causes serious eye damage.

Prevention:

P280 Wear eye protection/ face protection.

Response:

P305 + P351 + P338 + P310 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/ doctor.

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590

Reagent handling

Ready for use

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	26 weeks

CA2

Calcium Gen.2

cobas®

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: Fresh serum collected in the fasting state is the preferred specimen.
Plasma: Li-heparin plasma.

Serum or plasma should be separated from blood cells as soon as possible, because prolonged contact with the clot may cause lower calcium values.[3] Sera from patients receiving EDTA (treatment of hypercalcemia) are unsuitable for analysis, since EDTA will chelate the calcium and render it unavailable for reaction with NM-BAPTA. Co-precipitation of calcium with fibrin (i.e. heparin plasma), lipids, or denatured protein has been reported with storage or freezing.[1,4]

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

Urine specimens should be collected in acid-washed bottles. 24-hour specimens should be collected in containers containing 20-30 mL of 6 mol/L HCl to prevent calcium salt precipitation. Precipitated calcium salts may not be completely dissolved by the addition of HCl following urine collection.[5]

If stabilizers are added to the sample, the sample index feature must not be used.

Stability in *serum/plasma*:[6]	7 days at 15-25 °C
	3 weeks at 2-8 °C
	8 months at -20 °C (± 5 °C)

Freeze only once.

Stability in *urine*:[6]	2 days at 15-25 °C
	4 days at 2-8 °C
	3 weeks at -20 °C (± 5 °C)

Freeze only once.

Stored serum or urine specimens must be mixed well prior to analysis.

Centrifuge samples containing precipitates before performing the assay.

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

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

Test definition

Reporting time	10 min	
Wavelength (sub/main)	376/340 nm	
Reagent pipetting		Diluent (H_2O)
R1	15 µL	120 µL
R3	15 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.3 µL	–	–
Decreased	2.3 µL	–	–
Increased	2.3 µL	–	–

Application for urine

Test definition

Reporting time	10 min	
Wavelength (sub/main)	376/340 nm	
Reagent pipetting		Diluent (H_2O)
R1	15 µL	120 µL
R3	15 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.5 µL	–	–
Decreased	1.5 µL	15 µL	60 µL
Increased	1.5 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Application for serum/plasma (ACN 20340)

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	Automatic full calibration - after reagent lot change
	Full calibration - every 8 weeks - as required following quality control procedures

Application for urine (ACN 20341)
Transfer of calibration from serum/plasma application (ACN 20340)

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

Traceability: This method has been standardized against the SRM 956 c Level 2 reference material.

Quality control

For quality control, use control materials as listed in the “Order information” section. In addition, other suitable control material can be used.

Serum/plasma:	PreciControl ClinChem Multi 1, PreciControl ClinChem Multi 2
Urine:	Quantitative urine controls are recommended for routine quality control.

The control intervals and limits should be adapted to each laboratory’s individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 26 weeks.

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

cobas c systems automatically calculate the analyte concentration of each sample in the unit mmol/L (mg/dL, mg/L).

Conversion factors: mmol/L x 4.01 = mg/dL
mmol/L x 40.1 = mg/L

In studies with 24-hour urine, multiply the value obtained by the 24-hour volume in order to obtain a measurement in mg/24 h or mmol/24 h.

Limitations - interference

Criterion: Recovery within ± 10 % of initial value at a calcium concentration of 2.2 mmol/L.

Serum/plasma

Icterus:[7] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[7] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[7] No significant interference up to an L index of 1000. There is a poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Magnesium: No significant interference from magnesium up to a concentration of 15 mmol/L (36.5 mg/dL).

Drugs: No interference was found at therapeutic concentrations using common drug panels.[8,9]

The interference of intravenously administered gadolinium containing MRI (magnetic resonance imaging) contrast media was tested (Omniscan®, Optimark®) but no interference was found at the therapeutic concentration. Interferences at higher concentrations were observed.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[10]

Urine

Icterus: No significant interference up to a conjugated bilirubin concentration of 1026 µmol/L or 60 mg/dL.

Hemolysis: No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Magnesium: No significant interference from magnesium up to a concentration of 60 mmol/L (145.8 mg/dL).

Urea: No significant interference from urea up to a concentration of 1600 mmol/L (9610 mg/dL).

Drugs: No interference was found at therapeutic concentrations using common drug panels.[9]

The interference of intravenously administered gadolinium containing MRI (magnetic resonance imaging) contrast media was tested (Omniscan®, Optimark®). For Omniscan® no interference was observed at the therapeutic concentration, but there was interference at higher concentrations. For Optimark® interference was observed at therapeutic and higher concentrations.

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Limits and ranges

Measuring range

Serum/plasma

0.20-5.0 mmol/L (0.8-20.1 mg/dL)

Urine

0.20-7.5 mmol/L (0.8-30.1 mg/dL)

Determine urine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:5 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 5.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Serum/plasma and urine

Limit of Blank	= 0.10 mmol/L (0.4 mg/dL)
Limit of Detection	= 0.20 mmol/L (0.8 mg/dL)
Limit of Quantitation	= 0.20 mmol/L (0.8 mg/dL)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 30 %. It has been determined using low concentration calcium samples.

Expected values[11]

mmol/L

Serum/plasma

Children (0-10 days):	1.90-2.60 mmol/L
Children (10 days-2 years):	2.25-2.75 mmol/L
Children (2-12 years):	2.20-2.70 mmol/L
Children (12-18 years):	2.10-2.55 mmol/L
Adults (18-60 years):	2.15-2.50 mmol/L
Adults (60-90 years):	2.20-2.55 mmol/L
Adults (> 90 years):	2.05-2.40 mmol/L

Urine

2.5-7.5 mmol/24 h with normal food intake.

mg/dL

Serum/plasma

Children (0-10 days):	7.6-10.4 mg/dL
Children (10 days-2 years):	9.0-11.0 mg/dL
Children (2-12 years):	8.8-10.8 mg/dL
Children (12-18 years):	8.4-10.2 mg/dL
Adults (18-60 years):	8.6-10.0 mg/dL
Adults (60-90 years):	8.8-10.2 mg/dL
Adults (> 90 years):	8.2-9.6 mg/dL

Urine

100-300 mg/24 h with normal food intake.

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision

(2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Serum/plasma

Repeatability	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PCCC1[b)]	2.22	0.0103	0.5
PCCC2[c)]	3.41	0.0136	0.4
Human serum 1	0.398	0.00739	1.9
Human serum 2	1.43	0.00982	0.7
Human serum 3	2.12	0.0112	0.5
Human serum 4	2.42	0.0152	0.6
Human serum 5	4.13	0.0155	0.4

Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PCCC1[b)]	2.21	0.0140	0.6
PCCC2[c)]	3.41	0.0272	0.8
Human serum 1	0.398	0.00924	2.3
Human serum 2	1.43	0.0109	0.8
Human serum 3	2.13	0.0134	0.6
Human serum 4	2.42	0.0212	0.9
Human serum 5	4.13	0.0185	0.4

b) PreciControl ClinChem Multi 1

c) PreciControl ClinChem Multi 2

Urine

Repeatability	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
Control 1[d)]	1.73	0.0138	0.8
Control 2[d)]	2.39	0.0150	0.6
Human urine 1	0.374	0.0104	2.8
Human urine 2	1.44	0.0127	0.9
Human urine 3	2.25	0.0161	0.7
Human urine 4	3.56	0.0217	0.6
Human urine 5	6.25	0.0294	0.5

Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
Control 1[d)]	1.73	0.0150	0.9
Control 2[d)]	2.39	0.0180	0.8
Human urine 1	0.374	0.0170	4.5
Human urine 2	1.43	0.0159	1.1
Human urine 3	2.25	0.0236	1.0
Human urine 4	3.56	0.0282	0.8
Human urine 5	6.22	0.0425	0.7

d) commercially available control material

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

Calcium values for human serum, plasma and urine samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Serum/plasma

Sample size (n) = 71

Passing/Bablok[12]	Linear regression
y = 0.988x + 0.0216 mmol/L	y = 0.985x + 0.0295 mmol/L
τ = 0.964	r = 0.999

The sample concentrations were between 0.260 and 4.84 mmol/L.

Urine

Sample size (n) = 68

Passing/Bablok[12]	Linear regression
y = 0.964x + 0.0148 mmol/L	y = 0.967x + 0.0055 mmol/L
τ = 0.987	r = 1.000

The sample concentrations were between 0.270 and 7.12 mmol/L.

Calcium values for human serum, plasma and urine samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Serum/plasma

Sample size (n) = 73

Passing/Bablok[12]	Linear regression
y = 1.024x - 0.0379 mmol/L	y = 1.024x - 0.0304 mmol/L
τ = 0.977	r = 0.999

The sample concentrations were between 0.250 and 4.63 mmol/L.

Urine

Sample size (n) = 71

Passing/Bablok[12]	Linear regression
y = 0.996x - 0.00631 mmol/L	y = 0.990x + 0.00625 mmol/L
τ = 0.989	r = 0.999

The sample concentrations were between 0.300 and 7.42 mmol/L.

Calcium values for human serum, plasma and urine samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Serum/plasma

Sample size (n) = 73

Passing/Bablok[12]	Linear regression
y = 1.021x - 0.0334 mmol/L	y = 1.011x - 0.0110 mmol/L
τ = 0.955	r = 0.999

The sample concentrations were between 0.464 and 4.85 mmol/L.

Urine

Sample size (n) = 72

Passing/Bablok[12]	Linear regression
y = 1.017x + 0.00977 mmol/L	y = 1.016x + 0.0110 mmol/L
τ = 0.992	r = 1.000

The sample concentrations were between 0.250 and 7.13 mmol/L.

References

1 Endres DB, Rude RK. Mineral and Bone Metabolism. In: Burtis CA, Ashwood ER, Bruns ED, eds. Tietz Textbook of Clinical Chemistry and Molecular Diagnostics, 4th ed. St. Louis (MO): Saunders Elsevier 2006:1891-1965.

2 Rathod A, Bonny O, Guessous I, et al. Association of urinary calcium excretion with serum calcium and vitamin D levels. Clin J Am Soc Nephrol. 2015 Mar 6;10(3):452-62.

3 Heins M, Heil W, Withold W. Storage of Serum or Whole Blood Samples? Effect of Time and Temperature on 22 Serum Analytes. Eur J Clin Chem Clin Biochem 1995;33:231-238.

4 Wilding P, Zilva JF, Wilde CE. Transport of specimens for clinical chemistry analysis. Ann Clin Biochem 1977;14:301-306.

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7 Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-475.

8 Breuer J. Report on the Symposium "Drug effects in Clinical Chemistry Methods". Eur J Clin Chem Clin Biochem 1996;34:385-386.

9 Sonntag O, Scholer A. Drug interference in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.

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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 navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

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www.roche.com
+800 5505 6606

Calibrator for automated systems

cobas®

REF 10759350190 10759350500 → 12 x 3 mL Calibrator

English

System information

For use on **cobas c** and COBAS INTEGRA analyzer systems, refer to the corresponding method sheet of the assay for the identification on the systems.

Intended use

Calibrator for automated systems (C.f.a.s.) is for use in the calibration of quantitative Roche methods on Roche clinical chemistry analyzers as specified in the value sheets.

Summary

C.f.a.s. is a lyophilized calibrator based on human serum.

The concentrations and activities of the calibrator components have been adjusted to ensure optimal calibration of the appropriate Roche methods on clinical chemistry analyzers.

Some methods specified in the relevant value sheet may not be available in all countries.

Reagents – working solutions

Reactive components in the lyophilizate:

Human serum with chemical additives and material of biological origin as specified.

The origin of the biological additives is as follows:

Analyte	Origin
ALT (GPT)	porcine heart
AST (GOT)	human, recombinant
Acid phosphatase	human prostate/potato
Albumin	bovine plasma
Aldolase	rabbit muscle
Alkaline phosphatase	human placenta (recombinant)
Amylase, total	porcine pancreas
Amylase, pancreatic	porcine pancreas
Cholesterol	bovine plasma
Cholinesterase	human serum
Creatine kinase	rabbit muscle
γ-GT	human, recombinant
GLDH	bacterial, recombinant
LD (LDH)	porcine heart
Lipase	human pancreas (recombinant)
Triglycerides	chicken egg yolk

Non-reactive components:

Stabilizers

The concentrations and activities of the calibrator components are lot-specific. The exact calibrator values are given in the electronically available or enclosed value sheets.

The values are also encoded in the enclosed calibrator barcode sheets for COBAS INTEGRA and **cobas c** 111 analyzers.

For the **cobas c** analyzers (except for the **cobas c** 111 analyzer) the values are encoded in electronic files sent via the **cobas** link to the analyzers.

Calibrator values

The calibrator values were determined using the method stated in the electronically available or enclosed value sheets. Determinations were performed under strictly standardized conditions on Roche analyzers using Roche system reagents and the Roche master calibrator.

The calibrator values were obtained via single determinations performed in different laboratories, in several separate runs. The calibrator value specified is the mean of all values obtained.

Traceability information is given in the respective instructions for use of the system reagents.

Precautions and warnings

For in vitro diagnostic use for laboratory 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.

All human material should be considered potentially infectious. All products derived from human blood are prepared exclusively from the blood of donors tested individually and shown to be free from HBsAg and antibodies to HCV and HIV. The testing methods use assays that have been approved or cleared by the FDA or that are in compliance with the legal rules of the European Union (IVDR 2017/746/EU, IVDD 98/79/EC, Annex II, List A). However, as no testing method can rule out the potential risk of infection with absolute certainty, the material should be handled with the same level of care as a patient specimen. In the event of exposure, the directives of the responsible health authorities should be followed.[1,2]

Handling

Carefully open one bottle avoiding the loss of lyophilizate, and pipette in exactly 3.0 mL of distilled/deionized water. Carefully close the bottle and dissolve the contents completely by occasional gentle swirling within 30 minutes. Avoid the formation of foam.

The enclosed barcoded labels are intended exclusively for **cobas c** systems (except for the **cobas c** 513 analyzer) to identify the calibrator. Attach the barcoded labels to the tubes carrying the sample cups containing the calibrator material.

Storage and stability

Store at 2-8 °C.

Criterion for the stability data stated by Roche:

Recovery within ± 5 % of initial value.

Stability of the lyophilized calibrator at 2-8 °C:

Up to the stated expiration date.

Stability of the components in the reconstituted calibrator*:

at 15-25 °C	8 hours
at 2-8 °C	2 days
at -20 °C (± 5 °C)	28 days (when frozen once)

*Exceptions: see below

Stability of acid phosphatase, non-prostatic acid phosphatase and prostatic acid phosphatase in the reconstituted calibrator (criterion: ± 10 % of initial value):

at 15-25 °C	2 hours
at 2-8 °C	1 day
at -20 °C (± 5 °C)	14 days (when frozen once)

Stability of total bilirubin in the reconstituted calibrator (when stored protected from light):

at 15-25 °C	6 hours
at 2-8 °C	1 day
at -20 °C (± 5 °C)	14 days (when frozen once)

Stability of direct bilirubin in the reconstituted calibrator (when stored protected from light):

at 15-25 °C	3 hours
at 2-8 °C	8 hours
at -20 °C (± 5 °C)	14 days (when frozen once)

Store calibrator tightly capped (and protected from light) when not in use.

Materials provided

- See “Reagents – working solutions” section

Calibrator for automated systems

cobas®

- Barcoded labels

Materials required (but not provided)

- Roche system reagents and clinical chemistry analyzers
- General laboratory equipment

Assay

Use C.f.a.s. as specified in the relevant Method Sheet for the system reagents.

References

1 Occupational Safety and Health Standards: Bloodborne pathogens. (29 CFR Part 1910.1030). Fed. Register.

2 Directive 2000/54/EC of the European Parliament and Council of 18 September 2000 on the protection of workers from risks related to exposure to biological agents at work.

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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard:

CONTENT	Contents of kit
CALIBRATOR	Calibrator
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH
Sandhofer Strasse 116
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www.roche.com
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Roche

CHOL2

cobas®

Cholesterol Gen.2

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057443190*	08057443500	Cholesterol Gen.2 (2600 tests)	System-ID 2041 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057443214*	08057443500	Cholesterol Gen.2 (2600 tests)	System-ID 2041 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

10759350190	Calibrator f.a.s. (12 x 3 mL)	Code 20401	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

CHOL2-A: ACN 20410: Abell/Kendall Standardization

CHOL2-I: ACN 20411: ID/MS Standardization

Intended use

In vitro test for the quantitative determination of cholesterol in human serum and plasma on **cobas c** systems.

Summary

Measurements of cholesterol, performed with this assay, in human serum and plasma, are used in screening an individual's risk of developing atherosclerotic disease and as an aid in diagnosis, therapy guidance and monitoring of disorders involving elevated cholesterol levels as well as lipid and lipoprotein metabolic disorders.

Cholesterol is a steroid with a secondary hydroxyl group in the C3 position. It is synthesized in many types of tissue, but particularly in the liver and intestinal wall. Approximately three quarters of cholesterol is newly synthesized and a quarter originates from dietary intake. Cholesterol assays are used for screening for atherosclerotic risk and in the diagnosis and treatment of disorders involving elevated cholesterol levels as well as lipid and lipoprotein metabolic disorders.[1,2,3]

Cholesterol analysis was first reported by Liebermann in 1885 followed by Burchard in 1889.[4,5] In the Liebermann-Burchard reaction, cholesterol forms a blue-green dye from polymeric unsaturated carbohydrates in an acetic acid/acetic anhydride/concentrated sulfuric acid medium. The Abell and Kendall method is specific for cholesterol, but is technically complex and requires the use of corrosive reagents.[6] In 1974, Roeschlau and Allain described the first fully enzymatic method.[7,8] This method is based on the determination of Δ4-cholestenone after enzymatic cleavage of the cholesterol ester by cholesterol esterase, conversion of cholesterol by cholesterol oxidase, and subsequent measurement by the Trinder reaction of the hydrogen peroxide formed.[9] Optimization of ester cleavage (> 99.5 %) allows standardization using primary and secondary standards and a direct comparison with the CDC and NIST reference methods.[10,11]

Nonfasting sample results may be slightly lower than fasting results.[12,13,14]

The Roche cholesterol assay meets the 1992 National Institutes of Health (NIH) goal of less than or equal to 3 % for both precision and bias.[14]

The assay is optionally standardized against Abell/Kendall and isotope dilution/mass spectrometry.

Test principle

Enzymatic, colorimetric method.

Cholesterol esters are cleaved by the action of cholesterol esterase to yield free cholesterol and fatty acids. Cholesterol oxidase then catalyzes the oxidation of cholesterol to cholest-4-en-3-one and hydrogen peroxide. In the presence of peroxidase, the hydrogen peroxide formed effects the oxidative coupling of phenol and 4-aminoantipyrine (4-AAP) to form a red quinone-imine dye.

CE

$$\text{Cholesterol esters} + H_2O \longrightarrow \text{cholesterol} + \text{RCOOH}$$

$$\text{Cholesterol} + O_2 \xrightarrow{\text{CHOD}} \text{cholest-4-en-3-one} + H_2O_2$$

$$2\,H_2O_2 + \text{4-AAP} + \text{phenol} \xrightarrow{\text{POD}} \text{quinone-imine dye} + 4\,H_2O$$

The color intensity of the dye formed is directly proportional to the cholesterol concentration. It is determined by measuring the increase in absorbance.

Reagents – working solutions

R1 PIPES buffer: 225 mmol/L, pH 6.8; Mg^{2+}: 10 mmol/L; sodium cholate: 0.6 mmol/L; 4-aminoantipyrine: ≥ 0.45 mmol/L; phenol: ≥ 12.6 mmol/L; fatty alcohol polyglycol ether: 3 %; cholesterol esterase (Pseudomonas spec.): ≥ 25 µkat/L (≥ 1.5 U/mL); cholesterol oxidase (E. coli): ≥ 7.5 µkat/L (≥ 0.45 U/mL); peroxidase (horseradish): ≥ 12.5 µkat/L (≥ 0.75 U/mL); stabilizers; preservative

R1 is in position B.

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.

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

Warning

H319 Causes serious eye irritation.

Prevention:

P264 Wash skin thoroughly after handling.

P280 Wear eye protection/ face protection.

Response:

CHOL2

Cholesterol Gen.2

cobas®

P305 + P351 + P338	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P337 + P313	If eye irritation persists: Get medical advice/attention.

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590

Reagent handling

Ready for use

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	26 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

Do not use citrate, oxalate or fluoride.[15]

Fasting and nonfasting samples can be used.[13]

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.

Centrifuge samples containing precipitates before performing the assay.

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

Stability:[1,16]	7 days at 15-25 °C
	7 days at 2-8 °C
	3 months at -20 °C (± 5 °C)

Freeze only once.

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

Test definition

Reporting time	10 min		
Wavelength (sub/main)	700/505 nm		
Reagent pipetting		Diluent (H_2O)	
R1	26 µL	51 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.1 µL	–	–
Decreased	1.1 µL	10.0 µL	90 µL
Increased	1.1 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	Blank calibration - every 7 days on-board - every 7 days during shelf life
	Full 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 according to Abell/Kendall[14] and also by isotope dilution/mass spectrometry.[17]

Quality control

For quality control, use control materials as listed in the “Order information” section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory’s individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 26 weeks.

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

cobas c systems automatically calculate the analyte concentration of each sample in the unit mmol/L (mg/dL, g/L).

Conversion factors:	mmol/L x 38.66 = mg/dL
	mmol/L x 0.3866 = g/L

Limitations – interference

Criterion: Recovery within ± 10 % of initial value at a cholesterol concentration of 5.2 mmol/L.

Icterus:[18] No significant interference up to an I index of 16 for conjugated bilirubin and 14 for unconjugated bilirubin (approximate conjugated bilirubin concentration 274 µmol/L or 16 mg/dL; approximate unconjugated bilirubin concentration 239 µmol/L or 14 mg/dL).

Hemolysis:[18] No significant interference up to an H index of 700 (approximate hemoglobin concentration: 435 µmol/L or 700 mg/dL).

Lipemia (Intralipid):[18] No significant interference up to an L index of 2000. 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.[19,20]

Acetaminophen intoxications are frequently treated with N-acetylcysteine. N-Acetylcysteine at the therapeutic concentration when used as an antidote and the acetaminophen metabolite N-acetyl-p-benzoquinone imine (NAPQI) independently may cause falsely low results.

Venipuncture should be performed prior to the administration of metamizole. Venipuncture immediately after or during the administration of metamizole may lead to falsely low results.

In very rare cases, gammopathy, in particular type IgM (Waldenström’s macroglobulinemia), may cause unreliable results.[21]

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

CHOL2

Cholesterol Gen.2

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

Limits and ranges

Measuring range

0.1-20.7 mmol/L (3.86-800 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:10 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 10.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 0.1 mmol/L (3.86 mg/dL)
Limit of Detection	= 0.1 mmol/L (3.86 mg/dL)
Limit of Quantitation	= 0.1 mmol/L (3.86 mg/dL)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 20 %. It has been determined using low concentration cholesterol samples.

Expected values

mmol/L

Clinical interpretation according to the recommendations of the European Atherosclerosis Society:[2]

	mmol/L	*Lipid metabolic disorder*
Cholesterol	< 5.2	No
Triglycerides	< 2.3	No
Cholesterol	5.2-7.8	Yes, if HDL-cholesterol < 0.9 mmol/L
Cholesterol	> 7.8	Yes
Triglycerides	> 2.3	Yes

Recommendations of the NCEP Adult Treatment Panel for the following risk-cutoff thresholds for the US American population:[3]

Desirable cholesterol level	< 5.17 mmol/L
Borderline high cholesterol	5.17-6.18 mmol/L
High cholesterol	≥ 6.21 mmol/L

mg/dL

Clinical interpretation according to the recommendations of the European Atherosclerosis Society:[2]

	mg/dL	*Lipid metabolic disorder*
Cholesterol	< 200	No
Triglycerides	< 200	No
Cholesterol	200-300	Yes, if HDL-cholesterol < 35 mg/dL
Cholesterol	> 300	Yes
Triglycerides	> 200	Yes

Recommendations of the NCEP Adult Treatment Panel for the following risk-cutoff thresholds for the US American population:[3]

Desirable cholesterol level	< 200 mg/dL
Borderline high cholesterol	200-239 mg/dL
High cholesterol	≥ 240 mg/dL

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PCCC1[a)]	2.36	0.00970	0.4
PCCC2[b)]	5.15	0.0184	0.4
Human serum 1	0.226	0.00478	2.1
Human serum 2	5.02	0.0167	0.3
Human serum 3	6.02	0.0214	0.4
Human serum 4	9.55	0.0314	0.3
Human serum 5	17.9	0.0845	0.5

Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PCCC1[a)]	2.39	0.0257	1.1
PCCC2[b)]	5.11	0.0363	0.7
Human serum 1	0.249	0.0185	7.4
Human serum 2	5.02	0.0355	0.7
Human serum 3	6.01	0.0369	0.6
Human serum 4	9.55	0.0432	0.5
Human serum 5	17.9	0.0959	0.5

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

Cholesterol values for human serum and plasma samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 75

Passing/Bablok[22]	Linear regression
y = 1.019x + 0.00509 mmol/L	y = 1.020x - 0.0158 mmol/L
τ = 0.985	r = 1.000

The sample concentrations were between 0.344 and 18.8 mmol/L.

CHOL2

Cholesterol Gen.2

Cholesterol values for human serum and plasma samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 66

Passing/Bablok[22]	Linear regression
y = 1.024x + 0.00124 mmol/L	y = 1.022x + 0.00775 mmol/L
τ = 0.993	r = 1.000

The sample concentrations were between 0.330 and 18.2 mmol/L.

Cholesterol values for human serum and plasma samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 91

Passing/Bablok[22]	Linear regression
y = 1.014x - 0.0144 mmol/L	y = 1.018x - 0.0486 mmol/L
τ = 0.987	r = 0.999

The sample concentrations were between 0.141 and 19.3 mmol/L.

References

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3 Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). NIH Publication No 01-3670; May 2001.

4 Liebermann NC. Über das Oxychinoterpen. Ber Dtsch chem Ges 1885;18:1803.

5 Burchard H. Beiträge zur Kenntnis der Cholesterine. Dissertation, Rostock 1889.

6 Abell LL, Levy BB, Kendall FE. Cholesterol in serum. In: Seligon D (ed.). Standard Methods of Clinical Chemistry. Vol 2. Academic Press, New York 1958;26-33.

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8 Roeschlau P, Bernt E, Gruber W. Enzymatic determination of total cholesterol in serum. Z Klin Chem Klin Biochem 1974;12(5):226.

9 Trinder P. Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. Ann Clin Biochem 1969;6:24-27.

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14 Recommendations for Improving Cholesterol Measurement: A Report from the Laboratory Standardization Panel of the National Cholesterol Education Program. NIH Publication No. 90-2964 1990.

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17 Siekmann L, Hüskes KP, Breuer H. Determination of cholesterol in serum using mass fragmentography - a reference method in clinical chemistry. Z Anal Chem 1976;279:145-146.

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20 Sonntag O, Scholer A. Drug interference in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.

21 Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240-1243.

22 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

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 navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com
+800 5505 6606

CK

Creatine Kinase

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057460190*	08057460500	Creatine Kinase (500 tests)	System-ID 2042 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057460214*	08057460500	Creatine Kinase (500 tests)	System-ID 2042 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

10759350190	Calibrator f.a.s. (12 x 3 mL)	Code 20401	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

CK2: ACN 20420

Intended use

In vitro test for the quantitative determination of creatine kinase (CK) in human serum and plasma on **cobas c** systems.

Summary

Measurements of creatine kinase (CK), performed with this assay in human serum and plasma, are used as an aid in diagnosis of muscular injuries and diseases.

CK is a dimeric enzyme occurring in 4 different forms: a mitochondrial isoenzyme and the cytosolic isoenzymes CK MM (skeletal muscle type), CK BB (brain type) and CK MB (myocardial type).[1] Elevated total CK is observed in patients with skeletal and heart muscle injuries and diseases.[2]

The determination of CK and CK isoenzyme activities is utilized in the diagnosis and monitoring of muscular injuries and diseases in the acute (e.g. rhabdomyolysis or acute myocardial injury) and chronic settings (e.g. myopathies such as the progressive Duchenne muscular dystrophy). In acute rhabdomyolysis, for example, serum CK activities above 200 times the upper reference limit may be found. Serum CK activity is elevated in all types of muscular dystrophy.[3] In progressive muscular dystrophy, enzyme activity in serum may be increased long before the disease is clinically apparent.[3]

Following injury to the myocardium, such as occurs with acute myocardial infarction,[1] CK is released from the damaged myocardial cells. In early cases, a rise in the CK activity can be found just 4 hours after an infarction.[1,4] The CK activity reaches a maximum after 12-24 hours and then falls back to the normal range after 3-4 days.[1,4] According to the 4th Universal Definition of Myocardial Infarction, cardiac troponins are the preferred biomarkers for the evaluation of myocardial injury, since other biomarkers are less specific and less sensitive.[5]

The determination of CK levels can also be used for evaluation of drug toxicity. The European Society of Cardiology and the European Atherosclerosis Society recommend measuring CK in patients before initiation of lipid-lowering drug therapy, and in patients on lipid-lowering drugs, presenting with muscle pain and weakness, in order to identify the limited number of patients where treatment is contraindicated.[6]

The assay method using creatine phosphate and ADP was first described by Oliver,[7] modified by Rosalki[8] and further improved for optimal test conditions by Szasz et al.[9] CK is rapidly inactivated by oxidation of the sulfhydryl groups in the active center. The enzyme can be reactivated by the addition of acetylcysteine (NAC).[9] Interference by adenylate kinase is prevented by the addition of diadenosine pentaphosphate[10] and AMP.[9,10]

Standardized methods for the determination of CK with activation by NAC were recommended by the German Society for Clinical Chemistry (DGKC)[10] in 1977 and the International Federation of Clinical Chemistry (IFCC)[11] in 1991. In 2002 the IFCC confirmed their recommendation and extended it to 37 °C.[12,13] The method described here is derived from the formulation recommended by the IFCC and was optimized for performance and stability.

Test principle

UV-test

$$\text{Creatine phosphate} + \text{ADP} \xrightarrow{\text{CK}} \text{creatine} + \text{ATP}$$

$$\text{ATP} + \text{D-glucose} \xrightarrow{\text{HK}} \text{ADP} + \text{G6P}$$

$$\text{G6P} + \text{NADP}^+ \xrightarrow{\text{G6PDH}} \text{D-6-phosphogluconate} + \text{NADPH} + \text{H}^+$$

Equimolar quantities of NADPH and ATP are formed at the same rate. The photometrically measured rate of formation of NADPH is directly proportional to the CK activity.

Reagents - working solutions

R1 Imidazole buffer: 123 mmol/L, pH 6.5 (37 °C); EDTA: 2.46 mmol/L; Mg^{2+}: 12.3 mmol/L; ADP: 2.46 mmol/L; AMP: 6.14 mmol/L; diadenosine pentaphosphate: 19 µmol/L; $NADP^+$ (yeast): 2.46 mmol/L; N-acetylcysteine: 24.6 mmol/L; HK (yeast): ≥ 36.7 µkat/L; G6PDH (E. coli): ≥ 23.4 µkat/L; preservative; stabilizers; additives.

R3 CAPSO* buffer: 20 mmol/L, pH 8.8 (37 °C); glucose: 120 mmol/L; EDTA: 2.46 mmol/L; creatine phosphate: 184 mmol/L; preservative; stabilizers.

*CAPSO: 3-(cyclohexylamine)-2-hydroxy-1-propanesulfonic acid

R1 is in position B and R3 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.

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

CK

Creatine Kinase

Danger

H360D	May damage the unborn child.

Prevention:

P201	Obtain special instructions before use.
P202	Do not handle until all safety precautions have been read and understood.
P280	Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection.

Response:

P308 + P313	IF exposed or concerned: Get medical advice/attention.

Storage:

P405	Store locked up.

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

Reagent handling

Ready for use

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	8 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: Nonhemolyzed serum is the specimen of choice and also recommended by IFCC.
Plasma: Li-heparin, K_2-, K_3-EDTA plasma.

Please note: Differences in the degree of hemolysis resulting from the blood sampling procedure used can lead to deviating results in serum and 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.

Centrifuge samples containing precipitates before performing the assay.

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

Stability in serum:[14]	2 days at 20-25 °C
	7 days at 4-8 °C
	4 weeks at -20 °C (± 5 °C)

Freeze only once.

Stability in EDTA/heparin plasma:	2 days at 15-25 °C
	7 days at 2-8 °C
	4 weeks at -20 °C (± 5 °C)

Freeze only once.

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

Test definition

Reporting time	10 min		
Wavelength (sub/main)	546/340 nm		
Reagent pipetting		Diluent (H_2O)	
R1	79 µL	–	
R3	16 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.2 µL	–	–
Decreased	2.2 µL	10 µL	100 µL
Increased	2.2 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	Automatic full calibration - after reagent lot change
	Full calibration - 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 the IFCC Method for Creatine Kinase.[12]

Quality control

For quality control, use control materials as listed in the “Order information” section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory’s individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 8 weeks. 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.

CK

Creatine Kinase

cobas®

Calculation

cobas c systems automatically calculate the analyte activity of each sample in the unit U/L (µkat/L).

Conversion factor: U/L x 0.0167 = µkat/L

Limitations - interference

Criterion: Recovery within ± 10 % of initial value at a creatine kinase activity of 140 U/L.

Icterus:[15] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[15] No significant interference up to an H index of 100 (approximate hemoglobin concentration: 62.1 µmol/L or 100 mg/dL). The level of interference may be variable depending on the exact content of erythrocytes.

Lipemia (Intralipid):[15] No significant interference up to an L index of 1000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration. Highly lipemic specimens (L index > 1000) may cause high absorbance flagging.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[16,17] Cyanokit (hydroxocobalamin) at therapeutic concentrations interferes with the test.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[18]

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions refer to the operator's manual.

Limits and ranges

Measuring range

7-2000 U/L (0.12-33.4 µkat/L)

Determine samples having higher activities via the rerun function. Dilution of samples via the rerun function is a 1:11 dilution. Results from samples diluted by the rerun function are automatically multiplied by a factor of 11.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 7 U/L (0.12 µkat/L)
Limit of Detection	= 7 U/L (0.12 µkat/L)
Limit of Quantitation	= 7 U/L (0.12 µkat/L)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the activity below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low activity samples.

The Limit of Detection corresponds to the lowest analyte activity which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte activity that can be reproducibly measured with a total error of 20 %. It has been determined using low activity creatine kinase samples.

Expected values

Reference intervals strongly depend on the patient group and the specific clinical situation.

U/L

For healthy people, according to Klein et al.:[19]

CK	Men	39-308 U/L
	Women	26-192 U/L

Consensus values:[20]

CK	Men	< 190 U/L
	Women	< 170 U/L
CK-MB	Men/women	< 25 U/L

Myocardial infarction: There is a high probability of myocardial damage when the following 3 conditions are fulfilled:[21]

1	CK_{men}	> 190 U/L
	CK_{women}	> 167 U/L
2	CK-MB	> 24 U/L
3	The CK-MB activity accounts for 6-25 % of the total CK-activity.	

According to Tietz:[22]

CK	Adult males > 19 years	20-200 U/L
	Adult females > 19 years	20-180 U/L

µkat/L

For healthy people, according to Klein et al.:[19]*

CK	Men	0.65-5.14 µkat/L
	Women	0.43-3.21 µkat/L

*calculated by unit conversion factor

Consensus values:[20]

CK	Men	< 3.20 µkat/L
	Women	< 2.85 µkat/L
CK-MB	Men/women	< 0.42 µkat/L

Myocardial infarction: There is a high probability of myocardial damage when the following 3 conditions are fulfilled:[21]

1	CK_{men}	> 3.17 µkat/L
	CK_{women}	> 2.79 µkat/L
2	CK-MB	> 0.40 µkat/L
3	The CK-MB activity accounts for 6-25 % of the total CK-activity.	

According to Tietz:[22]*

CK	Adult males > 19 years	0.33-3.34 µkat/L
	Adult females > 19 years	0.33-3.01 µkat/L

*calculated by unit conversion factor

The reference values according to Klein et al. are based on the 95th percentile of a group of healthy persons (202 men and 217 women) not involved in high-intensity athletic activities.

In order to ensure high sensitivity in the diagnosis of heart diseases the values given by Tietz are recommended. The loss of diagnostic specificity thereby incurred can be compensated for by additionally determining CK-MB and/or troponin T. When myocardial infarction is suspected the diagnostic strategy proposals in the consensus document of European and American cardiologists should in general be followed.[23]

If despite the suspicion of myocardial infarction the values found remain below the stated limits, a fresh infarction may be involved. In such cases, the determinations should be repeated after 4 hours.

CK varies with physical activity level and race in healthy individuals.[22,24]

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

CK

Creatine Kinase

cobas®

Specific performance data

Representative performance data on the analyzers are given below. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean* *U/L*	*SD* *U/L*	*CV* *%*
PCCC1[a)]	155	0.764	0.5
PCCC2[b)]	287	0.988	0.3
Human serum 1	19.5	0.524	2.7
Human serum 2	85.7	0.510	0.6
Human serum 3	176	1.12	0.6
Human serum 4	900	3.28	0.4
Human serum 5	1588	4.52	0.3

Intermediate precision	*Mean* *U/L*	*SD* *U/L*	*CV* *%*
PCCC1[a)]	155	1.04	0.7
PCCC2[b)]	287	2.02	0.7
Human serum 1	19.4	0.582	3.0
Human serum 2	85.7	1.01	1.2
Human serum 3	176	1.96	1.1
Human serum 4	895	10.7	1.2
Human serum 5	1588	18.7	1.2

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

Creatine kinase values for human serum and plasma samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 80

Passing/Bablok[25]	Linear regression
y = 0.988x + 1.20 U/L	y = 0.993x - 0.788 U/L
τ = 0.996	r = 1.000

The sample activities were between 8.20 and 1938 U/L.

Creatine kinase values for human serum and plasma samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 110

Passing/Bablok[25]	Linear regression
y = 1.006x + 0.553 U/L	y = 1.013x - 1.03 U/L
τ = 0.990	r = 1.000

The sample activities were between 11.0 and 1959 U/L.

Creatine kinase values for human serum and plasma samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 75

Passing/Bablok[25]	Linear regression
y = 1.004x + 0.512 U/L	y = 1.006x - 0.256 U/L
τ = 0.995	r = 1.000

The sample concentrations were between 51.2 and 1939 U/L.

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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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

Symbols

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

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com
+800 5505 6606

CKMB

Creatine Kinase-MB

cobas®

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057486190*	08057486500	Creatine Kinase-MB (150 tests)	System-ID 2043 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057486214*	08057486500	Creatine Kinase-MB (150 tests)	System-ID 2043 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

11447394216	Calibrator f.a.s. CK-MB (3 x 1 mL)	Code 20402	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

CKMB2: ACN 20430

Intended use

In vitro test for the quantitative determination of the catalytic activity of creatine kinase MB subunit (CK-MB) in human serum and plasma on **cobas** c systems.

Summary

Measurements of CK-MB, performed with this assay in human serum and plasma, are used as an aid in diagnosis of myocardial infarction.

Creatine kinase (CK) appears as 3 isoenzymes which are dimers composed of 2 types of monomer subunits. The isoenzymes comprise all 3 combinations of monomers, M (for skeletal muscle derived) and B (for brain derived), as represented by the notations MM, MB, and BB.[1,2]

Many organs contain CK, but the distribution of isoenzymes is different in each one. Skeletal muscle is very rich in the MM isoenzyme, while brain, stomach, intestine, bladder, and lung contain primarily the BB isoenzyme. The MB isoenzyme has been found in appreciable amounts only in myocardial tissue (15 to 20 percent of the total myocardial CK).[3] Therefore, total serum CK activity is elevated in a number of diseases. This lack of specificity limits its diagnostic value. However, the striking difference in the CK isoenzyme patterns from different organs has made CK one of the most useful enzymes for diagnostic purposes in acute myocardial infarction. CK-MB appears in serum reflecting its unique presence in myocardial tissue. It is in supporting the diagnosis of suspected myocardial infarction that serial determinations of CK isoenzymes find their most frequent application in the clinical laboratory.[1,2]

Because of their higher sensitivity and specificity, cardiac troponins, measured by high-sensitivity assays, are the preferred biomarkers to define myocardial infarction,[4] and if a troponin assay is not available, the best alternative is CK-MB measured by a mass assay.[4]

After immunoinhibition with antibodies to the CK-M subunit,[5] the CK-B activity is determined with a standardized method for the determination of CK with activation by NAC as recommended by the German Society for Clinical Chemistry (DGKC)[6] and the International Federation of Clinical Chemistry (IFCC)[7,8] in 1977 and 2002 respectively. This assay meets the recommendations of the IFCC and DGKC, but was optimized for performance and stability.

Test principle

Immunological UV assay

- Sample and addition of R1 (buffer/enzymes/coenzyme)
- Addition of R2 (buffer/substrate/antibody) and start of reaction.

Human CK-MB is composed of 2 subunits, CK-M and CK-B which both have an active site. With the aid of specific antibodies to CK-M, the catalytic activity of CK-M subunits in the sample is inhibited to 99.6 % without affecting the CK-B subunits. The remaining CK-B activity, corresponding to half the CK-MB activity, is determined by the total CK method. As the CK-BB isoenzyme only rarely appears in serum and the catalytic activity of the CK-M and CK-B subunits hardly differ, the catalytic activity of the CK-MB isoenzyme can be calculated from the measured CK-B activity by multiplying the result by 2.

Reagents - working solutions

R1 Imidazole buffer: 123 mmol/L, pH 6.5 (37 °C); EDTA: 2.46 mmol/L; Mg^{2+}: 12.3 mmol/L; ADP: 2.46 mmol/L; AMP: 6.14 mmol/L; diadenosine pentaphosphate: 19 µmol/L; NADP (yeast): 2.46 mmol/L; N-acetylcysteine: 24.6 mmol/L; HK (yeast): ≥ 36.7 µkat/L; G6P-DH (E. coli): ≥ 23.4 µkat/L; preservative; stabilizers; additives.

R2 CAPSO* buffer: 20 mmol/L, pH 8.8 (37 °C); glucose: 120 mmol/L; EDTA: 2.46 mmol/L; creatine phosphate: 184 mmol/L; 4 monoclonal anti-CK-M antibodies (mouse), inhibiting capacity: > 99.6 % up to 66.8 µkat/L (4000 U/L) (37 °C) CK-M subunit; preservative; stabilizers; additive.

*CAPSO: 3-(cyclohexylamino)-2-hydroxy-1-propanesulfonic acid

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.

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

Danger

H360D May damage the unborn child.

Prevention:

P201 Obtain special instructions before use.

P202 Do not handle until all safety precautions have been read and understood.

P280	Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection.

Response:

P308 + P313	IF exposed or concerned: Get medical advice/attention.

Storage:

P405	Store locked up.

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

Reagent handling

Ready for use

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	8 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: Nonhemolyzed serum is the specimen of choice and also recommended by IFCC.
Plasma: Li-heparin, K_2-, K_3-EDTA plasma.

Li-heparin in the usual concentration does not interfere with the test, but IFCC warns against its use.[7]

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.

Centrifuge samples containing precipitates before performing the assay.

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

Stability in serum:[9]	8 hours at 20-24 °C
	8 days at 2-8 °C
	4 weeks at -20 °C (± 5 °C)

Freeze only once.

Stability in heparin plasma:[9]	8 hours at 20-24 °C
	5 days at 2-8 °C
	8 days at -20 °C (± 5 °C)

Freeze only once.

Stability in EDTA plasma:[10]	2 days at 20-25 °C
	7 days at 4-8 °C
	1 year at -20 °C (± 5 °C)

Freeze only once.

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

Test definition

Reporting time	10 min		
Wavelength (sub/main)	546/340 nm		
Reagent pipetting		Diluent (H_2O)	
R1	79 µL	–	
R2	16 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	3.9 µL	–	–
Decreased	11.7 µL	10 µL	80 µL
Increased	3.9 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s. CK-MB
Calibration mode	Linear
Calibration frequency	Automatic full calibration - after reagent lot change
	Full calibration - 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 the IFCC Method for Creatine Kinase[8] with addition of antibodies.

Quality control

For quality control, use control materials as listed in the “Order information” section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory’s individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 8 weeks. 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

cobas c systems automatically calculate the analyte activity of each sample in the unit U/L (µkat/L).

Conversion factor: U/L × 0.0167 = µkat/L

Limitations - interference

The total CK activity of the specimen should be determined prior to performing the CK-MB assay. The amount of anti-human CK-M subunit antibody in the CK-MB reagent is sufficient for the complete inhibition of up to 4000 U/L CK-M activity. If the total CK activity exceeds 4000 U/L, the specimen requires dilution because complete inhibition of the CK-M subunit is no longer assured. In patients with a disposition to macro-CK formation, implausibly high CK-MB values may be measured in relation to the total CK, since the macroforms mainly consist of CK-B subunits. As these patients

have generally not suffered a myocardial infarction, additional diagnostic measures are necessary.[11]

Criterion: Recovery within ± 10 % of initial value at a CK-MB activity of ≥ 25 U/L.

Icterus:[12] No significant interference up to an I index of 60 for conjugated and 20 for unconjugated bilirubin (approximate conjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL and approximate unconjugated bilirubin concentration: 342 µmol/L or 20 mg/dL).

Hemolysis:[12] No significant interference up to an H index of 20 (approximate hemoglobin concentration: 12.4 µmol/L or 20 mg/dL).

Lipemia (Intralipid):[12] No significant interference up to an L index of 500. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Adenylate kinase: Adenylate kinase (AK) may cause positive interference. Sources of AK in the blood are erythrocytes, muscle, and liver. In order to reduce AK interference to a minimum, AMP and Ap_5A are included in the reagent. The AMP/Ap_5A mixture causes 97 % inhibition of the AK from erythrocytes and muscle, and 95 % inhibition of the AK from liver.[6] The slight residual AK activity does not influence the assay of total CK, but may affect the low CK-MB activities.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[13,14] Exceptions: Cyanokit (hydroxocobalamin) and cefoxitin at therapeutic concentrations interfere with the test.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[15]

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Limits and ranges

Measuring range

3-2000 U/L (0.05-33.4 µkat/L)

Determine samples having higher activities via the rerun function. Dilution of samples via the rerun function is a 1:3 dilution. Results from samples diluted by the rerun function are automatically multiplied by a factor of 3.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 3 U/L (0.05 µkat/L)
Limit of Detection	= 3 U/L (0.05 µkat/L)
Limit of Quantitation	= 5 U/L (0.08 µkat/L)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the activity below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low activity samples.

The Limit of Detection corresponds to the lowest analyte activity which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte activity that can be reproducibly measured with a total error of 20 %. It has been determined using low activity CK-MB samples.

Expected values

Reference intervals strongly depend on the patient group regarded and the specific clinical situation.

U/L

For healthy people: Reference range (37 °C) according to Klein et al.[16] and consensus values:[17]

< 25 U/L

For myocardial infarction diagnosis using the combination CK and CK-MB (activity), and representing a CK consensus value based on long-term experience:[17,18]

1. CK_{men} > 190 U/L
   CK_{women} > 167 U/L
2. CK-MB > 24 U/L
3. The CK-MB activity accounts for 6-25 % of the total CK activity.

µkat/L

For healthy people: Reference range (37 °C) according to Klein et al.[16] and consensus values:[17]*

< 0.418 µkat/L

*calculated by unit conversion factor

For myocardial infarction diagnosis using the combination CK and CK-MB (activity), and representing a CK consensus value based on long-term experience:[17,18]

1. CK_{men} > 3.17 µkat/L
   CK_{women} > 2.79 µkat/L
2. CK-MB > 0.40 µkat/L
3. The CK-MB activity accounts for 6-25 % of the total CK activity.

When myocardial infarction is suspected the diagnostic strategy proposals in the consensus document of European and American cardiologists should in general be followed.[19]

If despite the suspicion of myocardial infarction the values found remain below the stated limits, a fresh infarction may be involved. In such cases the determinations should be repeated after 4 hours.

Maximum diagnostic efficiency of the CK-MB determination will be obtained when a sequential sampling protocol is used and consideration is given to the time pattern of activity over a 6 to 48 hour period. When only CK-MB activity is used, the diagnostic efficiency will be lower and will vary with the sampling time.[1,11]

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean* *U/L*	*SD* *U/L*	*CV* *%*
PCCC1[a)]	42.9	0.380	0.9
PCCC2[b)]	96.9	0.365	0.4
Human serum 1	16.0	0.358	2.2
Human serum 2	22.6	0.365	1.6
Human serum 3	190	0.779	0.4
Human serum 4	997	2.61	0.3

Human serum 5	1782	4.80	0.3
Intermediate precision	*Mean U/L*	*SD U/L*	*CV %*
PCCC1[a)]	42.9	0.557	1.3
PCCC2[b)]	96.6	0.712	0.7
Human serum 1	15.5	0.507	3.3
Human serum 2	22.3	0.560	2.5
Human serum 3	190	3.24	1.7
Human serum 4	997	11.1	1.1
Human serum 5	1784	28.1	1.6

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

CK-MB values for human serum and plasma samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 69

Passing/Bablok[20]	Linear regression
y = 1.014x - 1.73 U/L	y = 1.013x - 1.24 U/L
τ = 0.964	r = 1.000

The sample activities were between 4.90 and 1876 U/L.

CK-MB values for human serum and plasma samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 69

Passing/Bablok[20]	Linear regression
y = 1.015x + 0.202 U/L	y = 1.023x + 0.108 U/L
τ = 0.932	r = 1.000

The sample activities were between 3.50 and 1970 U/L.

CK-MB values for human serum and plasma samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 66

Passing/Bablok[20]	Linear regression
y = 1.003x + 1.52 U/L	y = 1.005x + 1.43 U/L
τ = 0.975	r = 1.000

The sample concentrations were between 3.81 and 1857 U/L.

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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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

Symbols

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

CONTENT	Contents of kit

CKMB

Creatine Kinase-MB

→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com
+800 5505 6606

Cl Electrode

cobas®

Chloride

Order information

REF	CONTENT	Analyzer(s) on which the electrode can be used
03246353001	Cl Electrode 1 (electrode)	**cobas c** 311 analyzer **cobas** 6000 analyzer series: **cobas c** 501 module **cobas** 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module **cobas pure** integrated solutions: **cobas c** 303 analytical unit **cobas pro** integrated solutions: **cobas pro** ISE analytical unit, **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Materials required (but not provided):

03149501001	REF Electrode (1 electrode)	
11360981216	ISE Reference Electrolyte (5 x 300 mL) ①②	
10820652216	ISE Reference Electrolyte (1 x 500 mL) ③④	
08392013190	ISE Reference Electrolyte (2 x 2000 mL) ⑤⑥	
04522320190	ISE Internal Standard Gen.2 (5 x 600 mL) ①②	
04880455190	ISE Internal Standard Gen.2 (2 x 2000 mL) ③④⑤	
09137742190	ISE Internal Standard Gen.2 conc. (1 x 510 mL) ⑥	
05979854190	Internal Standard Insert - ISE (Set of 20) ①②	
04522630190	ISE Diluent Gen.2 (5 x 300 mL) ①②	
04880480190	ISE Diluent Gen.2 (2 x 2000 mL) ③④⑤	
11298500316	ISE Cleaning Solution (5 x 100 mL)	
20763071122	ISE Deproteinizer (6 x 21 mL) ④⑤⑥	
03110435180	Deproteinizer (1 x 125 mL) ⑥	
04663632190	Activator (9 x 12 mL)	
11183974216	ISE Standard Low (10 x 3 mL)	Code 20502
11183982216	ISE Standard High (10 x 3 mL)	Codes 20503, 20763
12149435122	Precinorm U Plus (10 x 3 mL)	Code 20300
12149443122	Precipath U Plus (10 x 3 mL)	Code 20301
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392

ISE reagents on:

① **cobas c** 311 analyzer

② **cobas** 6000 analyzer series: **cobas c** 501 module

③ **cobas** 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module

④ **cobas pure** integrated solutions: **cobas c** 303 analytical unit

⑤ **cobas pro** integrated solutions: **cobas pro** ISE analytical unit

⑥ **cobas pro** integrated solutions: **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

English

System information

	ACN (Serum/ plasma)	ACN (Urine)	ACN (Plasma)	ACN (Serum)
	ISE CL	**ISE CL-U**	**ISE CL-P**	**ISE CL-S**
cobas c 311 analyzer, **cobas c** 501 module, **cobas** 8000 ISE 900 / 1800 module	991	991	---	---

	ACN (Serum/ plasma)	ACN (Urine)	ACN (Plasma)	ACN (Serum)
	ISE CL	**ISE CL-U**	**ISE CL-P**	**ISE CL-S**
cobas c 303 analytical unit, **cobas pro** ISE analytical unit	29090	29091	29092	29093

Cl Electrode

Chloride

	ACN (Serum/ plasma)	ACN (Urine)	ACN (Plasma)	ACN (Serum)
	CL	**CL-U**	**CL-P**	**CL-S**
cobas ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit	29250	29251	29252	29253

Intended use

The Cl Electrode is a device intended for the in-vitro quantitative determination of chloride in human origin serum, plasma and urine.

Summary[1]

The chloride ion measurements may be useful for the diagnosis and monitoring purposes as an aid in diagnosing and monitoring chloride imbalance, including hypo- and hyperchloremia that can be associated with, or observed during a number of underlying disease states or pathological conditions.

Test principle

Ion-selective electrode, using automatically diluted serum/plasma or urine specimens. The chloride electrode is based on an ion exchanger.[2]

Calculation

The equation given below is used for the calculation of sample and/or QC results:

$$C_S = C_{IS} \times 10^{\frac{E_S - E_{IS}}{\pm S}}$$

Where:

C_S	concentration of the ion in the sample
C_{IS}	concentration of the ion in the ISE Internal Standard
E_S	EMF of the sample
E_{IS}	EMF of the ISE Internal Standard
S	Slope of the electrode

The complete measurement system for a particular ion includes the ISE, a reference electrode and electronic circuits to measure and process the EMF to give the test ion concentration.

Precautions and warnings

For in vitro diagnostic use for trained laboratory technicians.

Warning

- Samples containing material of human origin are potentially infectious. Wear personal protective equipment when replacing or installing electrodes at analyzers. If any biohazardous material is spilled, wipe it up immediately and apply a disinfectant.
- If sample or waste contacts with your skin, wash the affected area immediately with soap and water, then apply a disinfectant. Consult a physician.
- When disposing of used electrodes, treat them as biohazardous.

Caution

- Do not use electrodes after the shelf life or on-board stability period has expired. Otherwise, it may lead to unstable sodium, potassium, and chloride results due to the unstable potential reading of electrodes.
- Perchlorate medication may result in falsely high chloride results due to an interference of perchlorate with the Cl Electrode determinations.
- Perform electrode flow path cleaning as stated in the Instructions for Use for applicable analyzers, at the end of a daily sample run. Improper electrode flow path cleaning may cause unstable reading of electrodes and it results in calibration failures.

As with any diagnostic test procedure, results should be interpreted taking all other test results and the clinical status of the patient into consideration.

In addition, pay attention to all precautions and warnings listed in the operator's manual of the analyzer.

NOTE: Boric acid (CAS Registry No. 10043-35-3) is contained in the gel solution inside the electrode at 0.2 % of the total weight as a preservative.

Storage and stability

Store at 7-40 °C.

See labels for expiration dates.

On-board stability

After installation the electrode is stable for the following time period:
2 months or 9000 tests, whichever comes first.

The electrodes should be replaced after this time period has expired. For replacement refer to instructions in the operator's manual of the applicable analyzers.

NOTE: When replacing the electrode in **cobas pro** or **cobas pure**, the user should scan the barcode affixed on the rear side of the package instead of the barcode placed on the product's label.

Slope range -40 to -68 mV/dec

NOTE: Due to the negative charge of the chloride ion, the slope is negative.

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.

It is important to follow tube manufacturers recommended procedures at and after blood collection.

Separate from cells if analysis is not performed within 4 hours.[3]

Serum

For chloride determinations, serum is the specimen of choice.

CAUTION: Serum separator tubes have to be used in accordance with the tube manufacturer's recommended procedures. If these procedures are not considered, it is possible to coat the sample probe with gel (interfering with proper sample level detection), or even to aspirate gel into the ISE system (resulting in a clogged system).

Plasma: Lithium heparin plasma

CAUTION: Inadequate mixing of plasma tubes can cause introduction of fibrin microclots into and subsequent clogging of the ISE.

NOTE: It is strongly recommended to avoid silicone-type gels, due to risk of silicon oil contaminations. In addition, tubes that exhibit a layer of clear liquid, which rises to the top of the serum after centrifugation, should not be used, in order to prevent coating the sample probes and interfering with ISE system. It is possible to clog the sample probes or the ISE tubing with gel or clots if these precautions are not taken.

Urine: Collect 24-hour urine without addition of preservatives and/or stabilizers. Store refrigerated during collection.

NOTE: Each laboratory should establish guidelines for determining acceptability of specimens and the corrective action to be taken if a specimen is considered unacceptable. Compile a laboratory-specific guideline.

Sample stability (serum, plasma): [4]

7 days at 15-25 °C

7 days at 2-8 °C

stable at (-15)-(-25) °C

Freeze/thaw only once.

Sample stability (urine): [4,5]

7 days at 15-25 °C

stable at (-15)-(-25) °C

up to 6 freeze-thaw cycles possible.[6]

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[4] 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

Cl Electrode

Chloride

and/or its own studies to determine specific stability criteria for its laboratory.

Materials provided

See “Order information” section

Materials required (but not provided)

See “Order information” section

General laboratory equipment

Application for serum, plasma and urine

Test definition

Serum/plasma

	Sample dilution	
Sample volume	*Sample*	*Diluent*
cobas c 311 analyzer, cobas c 501 module		
Normal	9.7 µL	291 µL / ISE Diluent
cobas 8000 ISE 900 / 1800 module, cobas c 303 analytical unit, cobas pro ISE analytical unit		
Normal	15 µL	450 µL / ISE Diluent
cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit		
Normal	15 µL	450 µL / System Water

Measuring range on *cobas c 311 analyzer, cobas c 501 module, cobas 8000 ISE 900 / 1800 module, cobas c 303 analytical unit, cobas pro ISE analytical unit, cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit:* 60-140 mmol/L

Analysis of chloride on ISE analytical units listed with serum and plasma specimens should yield a linear relationship from 60-140 mmol/L with a deviation from the linear line of less than 5 %.

The sample volumes given above under “Normal” are for samples, calibrators, and quality controls.

Urine

	Sample dilution	
Sample volume	*Sample*	*Diluent*
cobas c 311 analyzer, cobas c 501 module		
Normal	9.7 µL	291 µL / ISE Diluent
Decreased	6.5 µL	291 µL / ISE Diluent
cobas 8000 ISE 900 / 1800 module		
Normal	10 µL	450 µL / ISE Diluent
Increased	15 µL	450 µL / ISE Diluent
cobas c 303 analytical unit, cobas pro ISE analytical unit		
Normal	15 µL	450 µL / ISE Diluent
Decreased	10 µL	450 µL / ISE Diluent
cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit		
Normal	15 µL	450 µL / System Water
Decreased	10 µL	450 µL / System Water

Measuring range on *cobas c 311 analyzer, cobas c 501 module, cobas c 303 analytical unit, cobas pro ISE analytical unit, cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit:* 20-250 mmol/L

Analysis of chloride on ISE analytical units listed with urine specimens should yield a linear relationship from 20-250 mmol/L with a deviation from the linear line of less than 10 %.

Determine samples having higher concentrations via the rerun function. Dilution of samples via rerun function is a 1:46 dilution. Results from samples diluted using the rerun function are automatically multiplied by the dilution factor.

Measuring range on *cobas c 311 analyzer, cobas c 501 module, cobas c 303 analytical unit, cobas pro ISE analytical unit, cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit for urine samples with decreased sample volume (Rerun):* 251-375 mmol/L.

Analysis of chloride on ISE analytical units listed with urine specimens should yield a linear relationship from 251-375 mmol/L with a deviation from the linear line of less than 10 %.

The sample volumes given above under “Normal” are for samples, calibrators, and quality controls.

Measuring range on *cobas 8000 ISE 900 / 1800 module:* 60-350 mmol/L

Analysis of chloride on **cobas** 8000 ISE 900 / 1800 module with urine specimens should yield a linear relationship from 60-350 mmol/L with a deviation from the linear line of less than 10 %.

Determine samples having lower concentrations via the rerun function. Dilution of samples via rerun function is a 1:31 dilution. Results from samples diluted using the rerun function are automatically multiplied by the dilution factor.

Measuring range on *cobas 8000 ISE 900 / 1800 module for urine samples with increased sample volume (Rerun):* 20-59.9 mmol/L

Analysis of chloride on **cobas** 8000 ISE 900 / 1800 module with urine specimens should yield a linear relationship from 20-59.9 mmol/L with a deviation from the linear line of less than 10 %.

The sample volumes given above under “Normal” are for samples and quality controls.

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 10 mmol/L
Limit of Detection	= 10 mmol/L
Limit of Quantitation	= 20 mmol/L

The Limit of Blank, the Limit of Detection and the Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 30 %. It has been determined using low concentration chloride samples.

Values below Limit of Quantitation are not reliable due to possible higher uncertainty.

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.

Calibration

Calibration requires the following calibrators: ISE Standard Low (S1), ISE Standard High (S2), and ISE Standard High (S3).

The slope of the calibration curve is calculated from Standards 1 and 2. ISE Internal Standard / ISE Internal Standard conc. is measured to provide E_{IS} for all measurements. Refer to the operator’s manual of the analyzer for detailed calibration instructions.

Traceability: ISE Standard Low and ISE Standard High are prepared gravimetrically from highly purified inorganic salts.

Purity of these salts has been certified by argentometric titration, acidimetric titration or perchloric acid titration.

Calibration frequency

Cl Electrode

cobas®

Chloride

Calibration

- every 24 hours
- after ISE washing and maintenance
- after changing the reagent bottle ①
- after changing ISE Reference Electrolyte and/or Internal Standard conc. (depending on AutoCal settings) ②
- after replacing any electrode
- as required following quality control procedures

ISE reagents on:

① **cobas c** 311 analyzer, **cobas c** 501 module, **cobas** 8000 ISE 900 / 1800 module, **cobas c** 303 analytical unit, **cobas pro** ISE analytical unit

② **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Refer to the operator's manual for a detailed description of the Calibration/AutoCal function.

Quality control

For quality control, use control materials as listed in the "Order information" section. In addition, other suitable control material can be used.

Serum/plasma:	PreciControl ClinChem Multi 1, PreciControl ClinChem Multi 2
	Precinorm U Plus, Precipath U Plus
Urine:	Quantitative urine controls are recommended for routine quality control.

Quality controls should be performed daily and after every additional calibration.

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.

Refer to appropriate value sheets/package inserts for additional information.

Traceability: Each Roche Diagnostics control listed above has been standardized against ISE Standard Low and ISE Standard High.

Limitations - interference

Criterion: Recovery within ± 10 % of initial value.

Hemolysis - serum/plasma

Hemolysis:[7] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Hemolysis - urine

Hemolysis:[7] No significant interference up to a hemoglobin concentration of 621 µmol/L or 1000 mg/dL.

Icterus - serum/plasma

Icterus:[7] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Lipemia - serum/plasma

Lipemia (Intralipid, SMOFlipid):[7] No significant interference up to an L index of 2000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Drugs

The following drugs have been tested and caused no significant interference when added to aliquots of pooled normal human serum up to the indicated concentration. Falsely high chloride values have been reported from patients receiving perchlorate medication. This is due to an interference of perchlorate ions with chloride ISE determinations.

Serum/plasma

Acetaminophen (Paracetamol)	200 mg/L
Acetylsalicylic acid	1000 mg/L
Ampicillin-Na	1000 mg/L
Ascorbic acid	300 mg/L
Cefoxitin	2500 mg/L
Cyclosporine	5 mg/L
Doxycyclin	50 mg/L
Heparin	5000 IU/L
Ibuprofen	500 mg/L
Intralipid	10000 mg/L
Levodopa	20 mg/L
Methyldopa	20 mg/L
Metronidazole	200 mg/L
N-Acetylcysteine	1660 mg/L
Phenylbutazone	400 mg/L
Rifampicin	60 mg/L
Theophylline	100 mg/L

Urine

Acetaminophen (Paracetamol)	3000 mg/L
Ascorbic acid	4000 mg/L
Cefoxitin	12000 mg/L
Gentamycine sulfate	400 mg/L
Ibuprofen	4000 mg/L
Levodopa	1000 mg/L
Methyldopa	2000 mg/L
N-Acetylcysteine	10 mg/L
Ofloxacine	900 mg/L
Phenazopyridine	300 mg/L
Salicyluric acid	6000 mg/L
Tetracycline	300 mg/L

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Expected values[8]

Serum, Plasma	Adult	98-107 mmol/L
	>90 y	98-111 mmol/L
Urine 24 h	Infant	2-10 mmol/24 h
	Child <6 y	15-40 mmol/24 h
	6-10 y, M	36-110 mmol/24 h
	6-10 y, F	18-74 mmol/24 h
	10-14 y, M	64-176 mmol/24 h
	10-14 y, F	36-173 mmol/24 h
	Adult	110-250 mmol/24 h
	>60 y	95-195 mmol/24 h

The urinary excretion of chloride varies significantly with dietary intake. The values given here are typical of people on an average diet.

NOTE: It is recommended that each laboratory establishes and maintains its own reference ranges. The values given here are only to be used as a guideline.

Cl Electrode

Chloride

Precision

see precision data of the following analyzers in "Appendix 1: Precision":

cobas c 311 analyzer

cobas 6000 analyzer series: **cobas c** 501 module

cobas 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module

cobas pure integrated solutions: **cobas c** 303 analytical unit

cobas pro integrated solutions: **cobas pro** ISE analytical unit, **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Method comparison

see method comparison data of the following analyzers in "Appendix 2: Method comparison":

cobas c 311 analyzer

cobas 6000 analyzer series: **cobas c** 501 module

cobas 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module

cobas pure integrated solutions: **cobas c** 303 analytical unit

cobas pro integrated solutions: **cobas pro** ISE analytical unit, **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Maintenance

ISE washing procedure for cobas c 311 analyzer, cobas c 501 module, cobas 8000 ISE 900 / 1800 module, cobas c 303 and cobas pro ISE analytical unit.

The system maintenance procedures and frequencies stated in the operator's manual of the respective analyzer must be performed each day at the end of the daily sample run or after an elevated sample throughput.

cobas c 311:	The specially designated positions on the sample disk are used.
Position W1:	ISE Cleaning Solution
Position W2:	Activator

The ISE Wash procedure has to be manually selected out of maintenance items.

cobas c 501:	The specially labeled wash rack (green) is used.
Position 1:	Multiclean (not necessary when only the ISE is cleaned)
Position 2:	ISE Cleaning Solution
Position 3:	Activator

The system recognizes the wash rack and switches automatically to cleaning mode.

cobas 8000 ISE:	The specially labeled wash rack (green) is used.
Position 1:	Cell Cleaning Solution (not necessary when only the ISE is cleaned)
Position 2:	ISE Cleaning Solution
Position 3:	Activator

The system recognizes the wash rack and switches automatically to cleaning mode.

cobas c 303, cobas pro ISE:	The specially labeled wash rack (green) is used.
Position 1:	ISE Cleaning Solution (used for weekly wash rack)
Position 2:	ISE Cleaning Solution (used for daily wash rack)
Position 3:	Activator

The system recognizes the wash rack and switches automatically to cleaning mode.

The ISE systems require conditioning after cleaning and prior to calibration.

NOTE: Always use fresh solutions for cleaning.

ISE washing procedure for cobas ISE neo analytical unit

cobas ISE neo:	The ISE system wash tube holder is used.
Position CS:	ISE Cleaning Solution
Position A:	Activator

The maintenance task "ISE system wash" is scheduled and initiated automatically. For detailed description, refer to the operator's manual.

On-board stability of auxiliary reagents: ISE Cleaning Solution 4 days, Activator 4 days.

NOTE: Always exchange the tubes on the ISE tube holder, using new tubes for fresh reagents. **You must not refill them**, as this will lead to deterioration of the ISE measuring unit(s). Refer to the operator's manual for further information.

Appendix 1: Precision

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

cobas c 311 analyzer

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

cobas 6000 analyzer series: cobas c 501 module

Repeatability and intermediate precision were determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP5 requirements (2 aliquots per run, 2 runs per day, 21 days). The following results were obtained:

	Repeatability			Intermediate precision		
Sample (on a **cobas c** 501)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
Plasma low	68.5	0.2	0.3	68.5	0.6	0.8
Plasma medium	129.0	0.4	0.3	129.0	0.6	0.5
Plasma high	139.0	0.3	0.2	139.0	0.6	0.4
Precinorm U	86.2	0.2	0.3	86.2	0.5	0.6
Precipath U	119.2	0.3	0.2	119.2	0.5	0.4
Urine low	25.8	0.1	0.2	25.8	0.6	2.3
Urine medium	131.4	0.3	0.2	131.4	0.7	0.5
Urine high	243.4	0.6	0.2	243.4	1.8	0.7
Liquichek 1	97.5	0.2	0.2	97.5	1.6	1.6
Liquichek 2	198.2	0.4	0.2	198.2	2.3	1.2

cobas 8000 modular analyzer series: cobas 8000 ISE 900 / 1800 module

Repeatability and intermediate precision were determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP5 requirements (2 aliquots per run, 2 runs per day, 21 days). The following results were obtained:

	Repeatability			Intermediate precision		
Sample (on a **cobas** 8000)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
Plasma low	67.1	0.3	0.4	67.1	0.6	1.0
Plasma medium	128.4	0.4	0.3	128.4	0.7	0.6
Plasma high	138.0	0.6	0.4	138.0	0.9	0.7
Precinorm U	77.1	0.3	0.4	77.1	0.6	0.8

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Chloride

	Repeatability			Intermediate precision		
Sample (on a **cobas** 8000)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
Precipath U	111.8	0.3	0.3	111.8	0.6	0.6
Urine low[1)]	21.6	0.2	1.0	21.6	0.8	3.7
Urine medium[2)]	167.6	0.5	0.3	167.6	1.1	0.7
Urine high[2)]	333.5	1.6	0.5	333.5	3.5	1.0
Liquichek 1[2)]	97.5	0.5	0.5	97.5	0.9	0.9
Liquichek 2[2)]	193.2	1.5	0.8	193.2	2.0	1.0

1) Data obtained with urine rerun function.

2) Data obtained with default urine mode.

cobas pure integrated solutions: cobas c 303 analytical unit

The data obtained on **cobas pro** analyzer(s) are representative for **cobas c** 303 analyzer(s).

cobas pro integrated solutions: cobas pro ISE analytical unit

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas pro** ISE analytical unit.

	Repeatability			Intermediate precision		
Sample (on a **cobas pro** ISE analytical unit)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
PCCC1[a)]	82.5	0.31	0.4	82.5	1.22	1.5
PCCC2[b)]	112	0.46	0.4	112	1.15	1.0
Human plasma 1	71.2	0.31	0.4	71.2	1.20	1.7
Human plasma 2	112	0.51	0.5	112	1.06	0.9
Human plasma 3	91.6	0.44	0.5	91.6	1.38	1.5
Human plasma 4	123	0.50	0.4	123	0.85	0.7
Human plasma 5	137	0.53	0.4	137	1.03	0.7
Human serum 1	73.4	0.23	0.3	73.4	1.08	1.5
Human serum 2	111	0.53	0.5	111	0.98	0.9
Human serum 3	91.4	0.35	0.4	91.4	1.17	1.3
Human serum 4	124	0.54	0.4	124	0.89	0.7
Human serum 5	133	0.62	0.5	133	0.82	0.6
Liquichek 1	95.2	0.41	0.4	95.2	1.18	1.2
Liquichek 2	184	0.69	0.4	184	1.79	1.0
Human urine 1	28.5	0.14	0.5	28.5	1.08	3.8
Human urine 2	139	0.58	0.4	139	1.44	1.0
Human urine 3	115	0.55	0.5	115	1.37	1.2
Human urine 4	207	0.93	0.5	207	1.92	0.9
Human urine 5	236	0.99	0.4	236	2.59	1.1

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

cobas pro integrated solutions: cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas ISE neo** analytical unit.

	Repeatability			Intermediate precision		
Sample (on a **cobas** ISE neo analytical unit)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
PCCC1[a)]	85.9	0.56	0.7	85.9	1.21	1.4
PCCC2[b)]	107	0.65	0.6	107	1.27	1.2
Human serum 1	68.0	0.32	0.5	68.0	1.07	1.6
Human serum 2	104	0.46	0.4	104	0.94	0.9
Human serum 3	97.8	0.46	0.5	97.8	0.94	1.0
Human serum 4	114	0.61	0.5	114	1.05	0.9
Human serum 5	138	0.65	0.5	138	1.11	0.8
Human plasma 1	65.0	0.30	0.5	65.2	1.20	1.8
Human plasma 2	101	0.63	0.6	101	1.07	1.1
Human plasma 3	94.5	0.54	0.6	94.4	1.02	1.1
Human plasma 4	110	0.50	0.5	110	1.14	1.0
Human plasma 5	135	0.74	0.5	135	1.28	0.9
Liquichek 1	82.3	0.40	0.5	82.0	0.99	1.2
Liquichek 2	193	1.14	0.6	193	2.12	1.1
Human urine 1	23.5	0.54	2.3	23.5	1.63	7.0
Human urine 2	132	0.57	0.4	132	1.33	1.0
Human urine 3	105	0.66	0.6	106	1.10	1.0
Human urine 4	201	0.93	0.5	202	2.01	1.0
Human urine 5	247	1.02	0.4	247	3.51	1.4

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

Appendix 2: Method comparison

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

cobas c 311 analyzer

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

cobas 6000 analyzer series: cobas c 501 module

ISE values for human plasma and urine samples obtained on **cobas c** 501 analyzers (y) using ISE Standard High (compensated) as S3 Calibrator, were compared to those determined with the corresponding reference method (x) and with a **cobas c** 501 analyzer using ISE Compensator as S3 Calibrator.

The reference method used was: Chloride Analyzer 926S for chloride.

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[9]	Coeff. (r)
x: coulometry y: **cobas c** 501 (S3 = ISE Standard High)	Plasma / 105	62.0	136	y = 1.033x -1.800	0.998
Bias at 90 mmol/L = 1.170 (1.3 %) Bias at 112 mmol/L = 1.896 (1.7 %)					
x: **cobas c** 501 (S3 = ISE Compensator) y: **cobas c** 501 (S3 = ISE Standard High)	Plasma / 105	61.4	138	y = 1.000x + 0.500	0.999

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Chloride

Bias at 90 mmol/L = 0.500 (0.6 %) Bias at 112 mmol/L = 0.500 (0.4 %)					
x: coulometry y: **cobas c** 501 (S3 = ISE Standard High)	Urine / 105	22.0	248	y = 1.020x - 1.700	0.999
Bias at 60 mmol/L = -0.500 (-0.8 %) Bias at 170 mmol/L = 1.700 (1.0 %)					
x: **cobas c** 501 (S3 = ISE Compensator) y: **cobas c** 501 (S3 = ISE Standard High)	Urine / 105	21.2	250	y = 0.989x + 0.669	1.000
Bias at 60 mmol/L = 0.009 (0.0 %) Bias at 170 mmol/L = -1.201 (-0.7 %)					

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas 8000 modular analyzer series: cobas 8000 ISE 900 / 1800 module

ISE values for human plasma and urine samples obtained on a **cobas** 8000 analyzer (y) using ISE Standard High as S3 Calibrator, were compared with those determined using the corresponding reference method (x) and with **cobas c** 501 (x) using ISE Standard High as S3 Calibrator.

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[9]	Coeff. (r)
x: coulometry y: **cobas** 8000 (S3 = ISE Standard High)	Plasma / 100	65.0	123.0	y = 1.075x - 6.025	0.9902
Bias at 90 mmol/L = 0.725 (0.8 %) Bias at 112 mmol/L = 2.375 (2.1 %)					
x: **cobas c** 501 (S3 = ISE Standard High) y: **cobas** 8000 (S3 = ISE Standard High)	Plasma / 100	61.9	127.9	y = 0.987x + 1.858	0.9984
Bias at 90 mmol/L = 0.688 (0.8 %) Bias at 112 mmol/L = 0.402 (0.4 %)					
x: coulometry y: **cobas** 8000 (S3 = ISE Standard High)	Urine[2)] / 108	66.0	313.0	y = 1.036x - 4.891	0.9995
Bias at 60 mmol/L = -2.731 (-4.6 %) Bias at 170 mmol/L = 1.229 (0.7 %)					
x: **cobas c** 501 (S3 = ISE Standard High) y: **cobas** 8000 (S3 = ISE Standard High)	Urine[2)] / 108	62.0	349.8	y = 0.908x + 9.018	0.9999
Bias at 60 mmol/L = 3.497 (5.8 %) Bias at 170 mmol/L = -6.623 (-3.9 %)					
x: coulometry y: **cobas** 8000 (S3 = ISE Standard High)	Urine[1)] / 92	22.0	59.0	y = 0.973x - 0.927	0.9987
Bias at 30 mmol/L = -1.737 (-5.8 %)					
x: **cobas c** 501 (S3 = ISE Standard High) y: **cobas** 8000 (S3 = ISE Standard High)	Urine[1)] / 92	20.2	57.3	y = 0.981x + 0.728	0.9992
Bias at 30 mmol/L = 0.158 (0.5 %)					

1) Data obtained with urine rerun function.

2) Data obtained with default urine mode.

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas pure integrated solutions: cobas c 303 analytical unit

ISE values for human plasma and serum samples obtained on a **cobas c** 303 ISE unit (y) were compared with those determined using **cobas pro** ISE analytical unit (x) and with a **cobas c** 501 analyzer (x).

ISE values for human urine samples obtained on a **cobas c** 303 ISE unit (y) were compared with a **cobas pro** ISE analytical unit (x) and with a **cobas c** 501 analyzer (x).

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[9]	Coeff. (r)
x: **cobas pro** ISE y: **cobas c** 303 ISE	Plasma / 120	65.5	137	y = 0.996x + 0.276	0.999
Bias at 95 mmol/L = -0.110 (-0.1 %) Bias at 110 mmol/L = -0.171 (-0.2 %)					
x: **cobas c** 501 y: **cobas c** 303 ISE	Plasma / 120	66.4	138	y = 1.000x - 1.20	0.999
Bias at 95 mmol/L = -1.20 (-1.3 %) Bias at 110 mmol/L = -1.20 (-1.1 %)					
x: **cobas pro** ISE y: **cobas c** 303 ISE	Serum / 118	62.2	138	y = 1.000x - 0.200	1.000
Bias at 95 mmol/L = -0.200 (-0.2 %) Bias at 110 mmol/L = -0.200 (-0.2 %)					
x: **cobas c** 501 y: **cobas c** 303 ISE	Serum / 118	62.6	138	y = 1.003x - 1.01	1.000
Bias at 95 mmol/L = -0.761(-0.8 %) Bias at 110 mmol/L = -0.722 (-0.7 %)					

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x: **cobas pro** ISE y: **cobas c** 303 ISE	Urine / 119	21.3	243	y = 1.011x - 1.03	1.000
x: **cobas c** 501 y: **cobas c** 303 ISE	Urine / 118	21.3	249	y = 0.984x + 2.29	1.000

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas pro integrated solutions: cobas pro ISE analytical unit

ISE values for human plasma samples obtained on a **cobas pro** ISE analytical unit (y) were compared with a **cobas c** 501 analyzer (x).

ISE values for human urine samples obtained on a **cobas pro** ISE analytical unit (y) were compared with a **cobas c** 501 analyzer (x).

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[9]	Coeff. (r)
x: **cobas c** 501 y: **cobas pro** ISE	Plasma / 118	60.5	140	y = 0.997x - 0.127	1.000
Bias at 95 mmol/L = -0.384 (-0.4 %) Bias at 110 mmol/L = -0.423 (-0.4 %)					
x: **cobas c** 501 y: **cobas pro** ISE	Serum / 118	61.7	135	y = 1.000x - 0.600	1.000
Bias at 95 mmol/L = -0.600 (-0.6 %) Bias at 110 mmol/L = -0.600 (-0.5 %)					
x: **cobas c** 501 y: **cobas pro** ISE	Urine / 119	25.0	245	y = 1.023x - 2.09	1.000

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas pro integrated solutions: cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit

ISE values for human plasma and serum samples obtained on a **cobas** ISE neo analytical unit (y) were compared with a **cobas c** 501 analyzer (x) and with a **cobas pro** ISE analytical unit (x).

ISE values for human urine samples obtained on a **cobas** ISE neo analytical unit (y) were compared with a **cobas c** 501 analyzer (x) and with a **cobas pro** ISE analytical unit (x).

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[9]	Coeff. (r)
x: **cobas c** 501 y: **cobas** ISE neo	Serum / 115	63.1	139	y = 1.021x - 1.41	0.998
Bias at 95 mmol/L = 0.593 (0.6 %) Bias at 110 mmol/L = 0.908 (0.8 %)					
x: **cobas pro** ISE y: **cobas** ISE neo	Serum / 115	63.1	136	y = 1.045x - 2.77	0.998
Bias at 95 mmol/L = 1.52 (1.6 %) Bias at 110 mmol/L = 2.19 (2.0 %)					
x: **cobas c** 501 y: **cobas** ISE neo	Plasma / 119	62.4	137	y = 1.010x + 0.152	0.999
Bias at 95 mmol/L = 1.14 (1.2 %) Bias at 110 mmol/L = 1.29 (1.2 %)					
x: **cobas pro** ISE y: **cobas** ISE neo	Plasma / 118	60.7	135	y = 1.012x + 2.10	0.999
Bias at 95 mmol/L = 3.21 (3.4 %) Bias at 110 mmol/L = 3.38 (3.1 %)					
x: **cobas c** 501 y: **cobas** ISE neo	Urine / 118	22.6	248	y = 1.004x - 1.20	1.000
x: **cobas pro** ISE y: **cobas** ISE neo	Urine /114	30.0	248	y = 1.008x - 0.843	1.000

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

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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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

Cl Electrode

Chloride

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Symbols

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

Cont.	Quantity contained in the package
CONTENT	Quantity contained in the package
GTIN	Global Trade Item Number
INSTALL BEFORE	Latest date by which the electrode has to be installed on the analyzer
RoHS	Directive for the restriction of the use of certain hazardous substances in electrical and electronic equipment

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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CREJ2

Creatinine Jaffé Gen.2

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057532190*	08057532500	Creatinine Jaffé Gen.2 (2500 tests)	System-ID 2047 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057532214*	08057532500	Creatinine Jaffé Gen.2 (2500 tests)	System-ID 2047 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

10759350190	Calibrator f.a.s. (12 x 3 mL)	Code 20401	
03121313122	Precinorm PUC (4 x 3 mL)	Code 20240	
03121291122	Precipath PUC (4 x 3 mL)	Code 20241	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

CREJ2: ACN 20470 (Serum/plasma)

CREJ2U: ACN 20471 (Urine)

Intended use

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

Summary

Creatinine measurements, performed with this assay, in human serum, plasma and urine are used as an aid in diagnosis and monitoring of renal disease and in monitoring of renal dialysis. Creatinine measurements are also used for the calculation of the fractional excretion of other urine analytes (e. g., albumin, α-amylase).

Creatinine is a break-down product of creatine phosphate in muscle, and is usually produced at a fairly constant rate by the body (depending on muscle mass). It is freely filtered by the glomeruli and, under normal conditions, is not reabsorbed by the tubules to any appreciable extent. A small but significant amount is also actively secreted. Its concentration is thus, inversely related to glomerular filtration rate (GFR).[1,2]

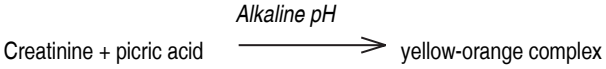
The assay of creatinine in serum or plasma is the most commonly used test to assess renal function. Chronic kidney disease is a worldwide problem that carries a substantial risk for cardiovascular morbidity and death. Current guidelines define chronic kidney disease as kidney damage or decreased glomerular filtration rate (GFR) (less than 60 mL/min per 1.73 m^2) for 3 months or more.[2,3]

Since a rise in blood creatinine is observed only with marked damage of the nephrons, it is not suited to detect early stage kidney disease. A considerably more sensitive test and better estimation of glomerular filtration rate (GFR) is given by the creatinine clearance test based on creatinine's concentration in urine and serum or plasma, and urine flow rate. For this test a precisely timed urine collection (usually 24 hours) and a blood sample are needed. However, since this test is prone to error due to the inconvenient collection of timed urine, mathematical attempts to estimate GFR (eGFR) based only on the creatinine concentration in serum or plasma have been made.[4] Among the various approaches suggested, three have found wide recognition: the Cockroft and Gault, the Modification of Diet in Renal Disease (MDRD) Study equation and the CKD-EPI (Chronic Kidney Disease Epidemiology) equation. While the Cockcroft and Gault equation was derived from data in which serum creatinine was measured with the conventional Jaffé method, the MDRD study equation measured serum creatinine using the Jaffé method calibrated to an isotope dilution mass spectrometry (IDMS).[5,6] These estimates of GFR are useful during monitoring of renal dialysis.[7,8] In children, the Bedside Schwartz formula should be used.[9,10,11]

In addition to the diagnosis and treatment of renal disease and the monitoring of renal dialysis, creatinine measurements are used for the calculation of the fractional excretion of other urine analytes (e. g., albumin, α-amylase). Numerous methods were described for determining creatinine. Automated assays established in the routine laboratory include the Jaffé alkaline picrate method in various modifications, as well as enzymatic tests.[2]

Test principle[12,13,14]

This kinetic colorimetric assay is based on the Jaffé method. In alkaline solution, creatinine forms a yellow-orange complex with picrate. The rate of dye formation is proportional to the creatinine concentration in the specimen. The assay uses "rate-blanking" to minimize interference by bilirubin. To correct for non-specific reaction caused by serum/plasma pseudo-creatinine chromogens, including proteins and ketones, the results for serum or plasma are corrected by -26 µmol/L (-0.3 mg/dL).



Reagents - working solutions

R1 Potassium hydroxide: 900 mmol/L; phosphate: 135 mmol/L; pH ≥ 13.5; preservative; stabilizer

R3 Picric acid: 38 mmol/L; pH 6.5; non reactive buffer

R1 is in position B and R3 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.

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

Danger

H314 Causes severe skin burns and eye damage.

Prevention:

P280	Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection.

Response:

P301 + P330 + P331	IF SWALLOWED: Rinse mouth. Do NOT induce vomiting.
P303 + P361 + P353	IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water.
P304 + P340 + P310	IF INHALED: Remove person to fresh air and keep comfortable for breathing. Immediately call a POISON CENTER/ doctor.
P305 + P351 + P338 + P310	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/ doctor.

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

Reagent handling

Ready for use

Storage and stability

Shelf life at 15-25 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	26 weeks

Specimen collection and preparation[15]

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 urine without using additives. If urine must be collected with a preservative for other analytes, only hydrochloric acid (14 to 47 mmol/L urine, e.g. 5 mL 10 % HCl or 5 mL 30 % HCl per liter urine) or boric acid (81 mmol/L, e.g. 5 g per liter urine) may be used. If stabilizers are added to the sample, the sample index feature must not be used.

Stability in *serum/plasma:*[16]	7 days at 15-25 °C
	7 days at 2-8 °C
	3 months at -20 °C (± 5 °C)

Freeze only once.

Stability in *urine* (without preservative):[16]	2 days at 15-25 °C
	6 days at 2-8 °C
	6 months at -20 °C (± 5 °C)

Freeze only once.

Stability in *urine* (with preservative):	3 days at 15-25 °C
	8 days at 2-8 °C
	3 weeks at -20 °C (± 5 °C)

Freeze only once.

Centrifuge samples containing precipitates before performing the assay.

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

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

Test definition

Reporting time	10 min	
Wavelength (sub/main)	570/505 nm	
Reagent pipetting		Diluent (H_2O)
R1	10 µL	58 µL
R3	13 µL	23 µL

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	7.5 µL	–	–
Decreased	7.5 µL	20 µL	80 µL
Increased	7.5 µL	–	–

Application for urine

Test definition

Reporting time	10 min	
Wavelength (sub/main)	570/505 nm	
Reagent pipetting		Diluent (H_2O)
R1	10 µL	58 µL
R3	13 µL	23 µL

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	7.5 µL	4 µL	96 µL
Decreased	7.5 µL	1.5 µL	135 µL
Increased	7.5 µL	4 µL	96 µL

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Application for serum/plasma (ACN 20470)

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear

Calibration frequency	Automatic full calibration - after reagent lot change Full calibration - every 8 weeks on-board - as required following quality control procedures

Application for urine (ACN 20471)
Transfer of calibration from serum/plasma application (ACN 20470)

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

Traceability: This method has been standardized against ID/MS.

Quality control

For quality control, use control materials as listed in the "Order information" section. In addition, other suitable control material can be used.

Serum/plasma:	PreciControl ClinChem Multi 1, PreciControl ClinChem Multi 2
Urine:	Precinorm PUC, Precipath PUC

The control intervals and limits should be adapted to each laboratory's individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 26 weeks.

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

cobas c systems automatically calculate the analyte concentration of each sample in the unit µmol/L (mg/dL, mmol/L, mg/L).

Conversion factors:	µmol/L x 0.0113 = mg/dL
	µmol/L x 0.001 = mmol/L
	µmol/L x 0.113 = mg/L

Limitations – interference

Criterion: Recovery within ± 10 % of initial value at a creatinine concentration of 80 µmol/L (0.90 mg/dL) in serum/plasma and 2.5 mmol/L (28.3 mg/dL) in urine.

Serum/plasma

Icterus (*CREJ2*):[17] No significant interference up to an I index of 5 for conjugated bilirubin and 10 for unconjugated bilirubin (approximate conjugated bilirubin concentration: 86 µmol/L or 5 mg/dL; approximate unconjugated bilirubin concentration: 171 µmol/L or 10 mg/dL).

Hemolysis:[17] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[17] No significant interference up to an L index of 800. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Pyruvate: No significant interference from pyruvate up to a concentration of 0.3 mmol/L (2.6 mg/dL).

Glucose: No significant interference from glucose up to a concentration of 25 mmol/L (450 mg/dL).

Ascorbic acid: No significant interference from ascorbic acid up to a concentration of 5 mmol/L (88 mg/dL).

Drugs: No interference was found at therapeutic concentrations using common drug panels.[18,19]

Exception: Antibiotics containing cephalosporin lead to significant false-positive values.[20,21] Cefoxitin causes artificially high creatinine results. Cyanokit (Hydroxocobalamin) may cause interference with results.

Values < 15 µmol/L (< 0.17 mg/dL) or negative results are reported in rare cases in children < 3 years and in elderly patients. In such cases use the Creatinine plus test to assay the sample.

Do not use Creatinine Jaffé for the testing of creatinine in hemolyzed samples from neonates, infants or adults with HbF levels ≥ 60 mg/dL for *CREJ2* applications.[22] In such cases, use the Creatinine plus test (≤ 600 mg/dL HbF) to assay the sample.

Estimation of the Glomerular Filtration Rate (GFR) on the basis of the Schwartz Formula can lead to an overestimation.[23]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[24]

The presence of ketone bodies can cause artificially high results in serum and plasma.

Urine

Icterus: No significant interference up to a conjugated bilirubin concentration of 855 µmol/L or 50 mg/dL.

Hemolysis: No significant interference up to an H index of 1000 (approximate hemoglobin concentration of 621 µmol/L or 1000 mg/dL).

Glucose: No significant interference from glucose up to a concentration of 120 mmol/L (2162 mg/dL).

Urea: No significant interference from urea up to a concentration of 2100 mmol/L (12612 mg/dL).

Urobilinogen: No significant interference from urobilinogen up to a concentration of 676 µmol/L (40 mg/dL).

Drugs: No interference was found at therapeutic concentrations using common drug panels.[19]

Exception: Cyanokit (Hydroxocobalamin) may cause interference with results.

High homogentisic acid concentrations in urine samples lead to false results.

The presence of ketone bodies can cause artificially high results in urine.

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Limits and ranges

Measuring range

Serum/plasma

15-2200 µmol/L (0.17-24.9 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:5 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 5.

Urine

0.375-55 mmol/L (4.2-622 mg/dL)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:3.6 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 3.6.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Serum/plasma (CREJ2)

Limit of Blank	= 15 µmol/L (0.17 mg/dL)
Limit of Detection	= 15 µmol/L (0.17 mg/dL)
Limit of Quantitation	= 15 µmol/L (0.17 mg/dL)

Urine (CREJ2U)

Limit of Blank	= 0.375 mmol/L (4.24 mg/dL)
Limit of Detection	= 0.375 mmol/L (4.24 mg/dL)
Limit of Quantitation	= 0.375 mmol/L (4.24 mg/dL)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 20 %. It has been determined using low concentration creatinine samples.

Expected values

µmol/L

Serum/plasma

Adults[25]

Females	44-80 µmol/L
Males	62-106 µmol/L

Children[26]

Neonates (premature)	25-91 µmol/L
Neonates (full term)	21-75 µmol/L
2-12 months	15-37 µmol/L
1- < 3 years	21-36 µmol/L
3- < 5 years	27-42 µmol/L
5- < 7 years	28-52 µmol/L
7- < 9 years	35-53 µmol/L
9- < 11 years	34-65 µmol/L
11- < 13 years	46-70 µmol/L
13- < 15 years	50-77 µmol/L

mmol/L

Urine

1st morning urine[25]

Females	2.47-19.2 mmol/L
Males	3.45-22.9 mmol/L

24-hour urine[27]

Females	7.0-14.0 mmol/24 h
Males	9.0-21.0 mmol/24 h

Creatinine clearance[27,28]	71-151 mL/min

Refer to reference for a prospective study on creatinine clearance in children.[29]

mg/dL

Serum/plasma

Adults[25]

Females	0.50-0.90 mg/dL
Males	0.70-1.20 mg/dL

Children[26]

Neonates (premature)	0.29-1.04 mg/dL
Neonates (full term)	0.24-0.85 mg/dL
2-12 months	0.17-0.42 mg/dL
1- < 3 years	0.24-0.41 mg/dL
3- < 5 years	0.31-0.47 mg/dL
5- < 7 years	0.32-0.59 mg/dL
7- < 9 years	0.40-0.60 mg/dL
9- < 11 years	0.39-0.73 mg/dL
11- < 13 years	0.53-0.79 mg/dL
13- < 15 years	0.57-0.87 mg/dL

Urine

1st morning urine[25]

Females	28-217 mg/dL
Males	39-259 mg/dL

24-hour urine[27]

Females	740-1570 mg/24 h
Males	1040-2350 mg/24 h

Creatinine clearance[27,28]	71-151 mL/min

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Serum/plasma (CREJ2)

Repeatability	*Mean*	*SD*	*CV*
	µmol/L	*µmol/L*	*%*
PCCC1[a)]	90.4	1.57	1.7
PCCC2[b)]	347	3.87	1.1
Human serum 1	48.2	1.40	2.9
Human serum 2	71.8	1.51	2.1
Human serum 3	480	4.15	0.9
Human serum 4	1064	12.0	1.1
Human serum 5	1791	20.6	1.2

Intermediate precision	*Mean*	*SD*	*CV*
	µmol/L	*µmol/L*	*%*
PCCC1[a)]	89.2	2.33	2.6
PCCC2[b)]	347	5.24	1.5
Human serum 1	48.2	1.64	3.4
Human serum 2	71.8	1.89	2.6
Human serum 3	480	7.59	1.6
Human serum 4	1064	16.1	1.5
Human serum 5	1791	30.0	1.7

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

CREJ2

Creatinine Jaffé Gen.2

Urine (CREJ2U)

Repeatability	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PN PUC[c)]	8.89	0.0922	1.0
PP PUC[d)]	4.56	0.0560	1.2
Human urine 1	1.19	0.0310	2.6
Human urine 2	2.41	0.0311	1.3
Human urine 3	22.5	0.210	0.9
Human urine 4	28.4	0.318	1.1
Human urine 5	49.7	0.480	1.0

Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PN PUC[c)]	8.89	0.151	1.7
PP PUC[d)]	4.56	0.0824	1.8
Human urine 1	1.19	0.0341	2.9
Human urine 2	2.43	0.0398	1.6
Human urine 3	22.5	0.339	1.5
Human urine 4	28.4	0.417	1.5
Human urine 5	49.7	0.745	1.5

c) Precinorm PUC

d) Precipath PUC

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

Creatinine values for human serum, plasma and urine samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Serum/plasma (CREJ2)

Sample size (n) = 71

Passing/Bablok[30]	Linear regression
y = 1.012x - 3.68 µmol/L	y = 1.010x - 3.19 µmol/L
τ = 0.980	r = 1.000

The sample concentrations were between 23.2 and 2133 µmol/L.

Urine (CREJ2U)

Sample size (n) = 72

Passing/Bablok[30]	Linear regression
y = 1.065x - 0.0368 mmol/L	y = 1.056x + 0.00514 mmol/L
τ = 0.984	r = 1.000

The sample concentrations were between 0.388 and 50.8 mmol/L.

Creatinine values for human serum, plasma and urine samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Serum/plasma (CREJ2)

Sample size (n) = 70

Passing/Bablok[30]	Linear regression
y = 1.018x - 5.48 µmol/L	y = 1.015x - 4.42 µmol/L
τ = 0.968	r = 1.000

The sample concentrations were between 24.1 and 2114 µmol/L.

Urine (CREJ2U)

Sample size (n) = 69

Passing/Bablok[30]	Linear regression
y = 1.088x - 0.0452 mmol/L	y = 1.093x - 0.0846 mmol/L
τ = 0.984	r = 1.000

The sample concentrations were between 0.787 and 49.1 mmol/L.

Creatinine values for human serum, plasma and urine samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Serum/plasma (CREJ2)

Sample size (n) = 86

Passing/Bablok[30]	Linear regression
y = 1.000x + 1.00 µmol/L	y = 0.997x + 2.37 µmol/L
τ = 0.983	r = 1.000

The sample concentrations were between 25.5 and 2120 µmol/L.

Urine (CREJ2U)

Sample size (n) = 89

Passing/Bablok[30]	Linear regression
y = 0.983x - 0.0174 mmol/L	y = 0.984x - 0.0442 mmol/L
τ = 0.993	r = 1.000

The sample concentrations were between 0.478 and 53.4 mmol/L.

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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 navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com
+800 5505 6606

CRP4

cobas®

Tina-quant C-Reactive Protein IV

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057591190	08057591500	Tina-quant C-Reactive Protein IV (500 tests)	System-ID 2050 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

11355279216	Calibrator f.a.s. Proteins (5 x 1 mL)	Code 20656	
20766321322	CRP T Control N (5 x 0.5 mL)	Code 20235	
10557897122	Precinorm Protein (3 x 1 mL)	Code 20302	
11333127122	Precipath Protein (3 x 1 mL)	Code 20303	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

English

System information

CRP4: ACN 20500

Intended use

Immunoturbidimetric assay for the in vitro quantitative determination of CRP in human serum and plasma on **cobas c** systems.

Summary

CRP measurements, performed with this assay in human serum or plasma, are used as aid in diagnosis, monitoring, prognosis, and management of suspected inflammatory disorders and associated diseases, acute infections and tissue injury.

C-reactive protein is the classic acute phase protein in inflammatory reactions.[1] It is synthesized by the liver and consists of 5 identical polypeptide chains that form a 5 membered ring having a molecular weight of 105000 daltons.[1,2,3,4] CRP is the most sensitive of the acute phase reactants and its concentration increases rapidly during inflammatory processes.[2,3] Complexed CRP activates the classical complement pathway. The CRP response frequently precedes clinical symptoms, including fever.[1,3] After onset of an acute phase response the serum CRP concentration rises rapidly and extensively.[2,3,4] The increase begins within 6 to 12 hours and the peak value is reached within 24 to 48 hours.[1,3,5] Levels above 100 mg/L are associated with severe stimuli such as major trauma and severe infection (sepsis).[5] CRP response may be less pronounced in patients suffering from liver disease.[6]

CRP assays are used to detect systemic inflammatory processes (apart from certain types of inflammation such as systemic lupus erythematosus (SLE) and Colitis ulcerosa);[1,3,4,6] to assess treatment of bacterial infections with antibiotics;[1,4,6,7] to detect intrauterine infections with concomitant premature amniorrhexis;[4,6] to differentiate between active and inactive forms of disease with concurrent infection, e.g. in patients suffering from SLE or Colitis ulcerosa;[3,4,6] to therapeutically monitor rheumatic disease and assess anti-inflammatory therapy;[1,4,6] to determine the presence of post-operative complications at an early stage, such as infected wounds, thrombosis and pneumonia, and to distinguish between infection and bone marrow transplant rejection.[1,4,6]

Various assay methods are available for CRP determination, such as nephelometry and turbidimetry.[8,9] The Roche CRP assay is based on the principle of particle-enhanced immunological agglutination.

Test principle[10,8]

Particle-enhanced immunoturbidimetric assay

Human CRP agglutinates with latex particles coated with monoclonal anti-CRP antibodies. The aggregates are determined turbidimetrically.

Reagents - working solutions

R1 TRIS[a)] buffer with bovine serum albumin; preservatives

R3 Latex particles coated with anti-CRP (mouse) in glycine buffer; immunoglobulins (mouse); preservative

a) TRIS = Tris(hydroxymethyl)-aminomethane

R1 is in position B and R3 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.

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

Warning

H317 May cause an allergic skin reaction.

Prevention:

P261 Avoid breathing mist or vapours.

P272 Contaminated work clothing should not be allowed out of the workplace.

P280 Wear protective gloves.

Response:

P333 + P313 If skin irritation or rash occurs: Get medical advice/attention.

P362 + P364 Take off contaminated clothing and wash it before reuse.

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

Reagent handling

Ready for use

Carefully invert reagent container several times prior to use to ensure that the reagent components are mixed.

CRP4

Tina-quant C-Reactive Protein IV

cobas®

Storage and stability

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, K_2-EDTA, K_3-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.

Centrifuge samples containing precipitates before performing the assay.

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

Stability in serum and Li-heparin plasma:	2 weeks at 15-25 °C
	3 weeks at 2-8 °C
	12 months at -20 °C (± 5 °C)
Stability in K_2- and K_3-EDTA plasma:	1 day at 15-25 °C
	3 weeks at 2-8 °C
	12 months at -20 °C (± 5 °C)

Freeze only once.

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

Test definition

Reporting time	10 min	
Wavelength (sub/main)	800/570 nm	
Reagent pipetting		Diluent (H_2O)
R1	98 µL	
R3	31 µL	16 µL

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.3 µL	–	–
Decreased	2.6 µL	20 µL	60 µL
Increased	1.3 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2: Calibrator f.a.s. Proteins
Calibration mode	Non-linear
Calibration frequency	Full calibration - after reagent lot change - every 3 weeks on-board - every 6 months during shelf life - as required following quality control procedures

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

This method has been standardized against the certified reference material in human serum of the IRMM (Institute for Reference Materials and Measurements) ERM-DA474/IFCC.[11]

Quality control

For quality control, use control materials as listed in the “Order information” section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory’s individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 12 weeks. 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

cobas c systems automatically calculate the analyte concentration of each sample in the unit mg/L (nmol/L, mg/dL).

Conversion factors:	mg/L × 9.52 = nmol/L
	mg/L × 0.1 = mg/dL

Limitations - interference

Criterion: Recovery within ± 0.5 mg/L of initial values of samples ≤ 5.0 mg/L and within ± 10 % for samples > 5 mg/L.

Icterus:[12] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 60 mg/dL or 1026 µmol/L).

Hemolysis:[12] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 622 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[12] No significant interference up to an L index of 1000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Rheumatoid factors: No significant interference from rheumatoid factors up to a concentration of 1200 IU/mL.

Immunoglobulins: No significant interference from immunoglobulins up to a concentration of 50 g/L (334 µmol/L) (simulated by human immunoglobulin G).

High-dose hook effect: No false result occurs up to a CRP concentration of 1200 mg/L.

In vitro tests were performed on commonly used pharmaceuticals. In addition, special pharmaceuticals were tested. Among them, the following substance caused interference:

Substance	No significant interference up to
Ticarcillin	225 mg/L

Drug interferences are measured based on recommendations given in CLSI guidelines EP07 and EP37 and other published literature. Effects of concentrations exceeding these recommendations have not been characterized.

As with any assay employing mouse antibodies, the possibility exists for interference by human anti-mouse antibodies (HAMA) in the sample, which could cause falsely lowered results.

CRP4

Tina-quant C-Reactive Protein IV

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

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Limits and ranges

Measuring range

0.6-350 mg/L (5.7-3332 nmol/L)

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

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 0.2 mg/L (1.9 nmol/L)
Limit of Detection	= 0.3 mg/L (2.9 nmol/L)
Limit of Quantitation	= 0.6 mg/L (5.7 nmol/L)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 20 %. It has been determined using low concentration C-reactive protein samples.

Expected values

Consensus reference interval for adults:[14] < 5 mg/L (< 47.6 nmol/L*)

*calculated by unit conversion factor

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP5-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean mg/L*	*SD mg/L*	*CV %*
CRP T Control N	3.33	0.0313	0.9
Precinorm Protein	9.72	0.0516	0.5
Precipath Protein	53.9	0.275	0.5
Human serum 1	1.11	0.0276	2.5
Human serum 2	4.09	0.0338	0.8
Human serum 3	82.9	0.474	0.6
Human serum 4	174	1.37	0.8
Human serum 5	305	2.10	0.7

Intermediate precision	*Mean mg/L*	*SD mg/L*	*CV %*
CRP T Control N	3.33	0.0375	1.1
Precinorm Protein	9.72	0.0708	0.7
Precipath Protein	53.9	0.854	1.6
Human serum 1	1.11	0.0296	2.7
Human serum 2	4.09	0.0397	1.0
Human serum 3	82.9	1.61	1.9
Human serum 4	174	3.94	2.3
Human serum 5	305	5.79	1.9

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

CRP values for human serum and plasma samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 157

Passing/Bablok[15]	Linear regression
y = 0.990x + 0.124 mg/L	y = 0.978x + 0.428 mg/L
τ = 0.995	r = 1.000

The sample concentrations were between 0.791 and 333 mg/L.

CRP values for human serum and plasma samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 79

Passing/Bablok[15]	Linear regression
y = 0.976x - 0.0226 mg/L	y = 0.973x + 0.340 mg/L
τ = 0.989	r = 1.000

The sample concentrations were between 0.920 and 348 mg/L.

CRP values for human serum and plasma samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 101

Passing/Bablok[15]	Linear regression
y = 1.013x + 0.0240 mg/L	y = 1.018x - 0.501 mg/L
τ = 0.992	r = 1.000

The sample concentrations were between 0.620 and 335 mg/L.

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.

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Symbols

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

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com
+800 5505 6606

ECO-D

EcoTergent

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08063354190*	08063354500	EcoTergent (40 mL)	System-ID 2901 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08063354214*	08063354500	EcoTergent (40 mL)	System-ID 2901 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

* Some kits shown may not be available in all countries.

English

System information

ECO-D: ACN 29010

Intended use

EcoTergent is an additive to the reaction bath to reduce surface tension on **cobas c** systems.

Summary

EcoTergent is added to the reaction bath. It acts as a surfactant to minimize the formation of bubbles that could potentially interfere with the photometer readings.

Reagents - working solutions

Detergent

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.

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

Danger

H314	Causes severe skin burns and eye damage.
H410	Very toxic to aquatic life with long lasting effects.

Prevention:

P273	Avoid release to the environment.
P280	Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection.

Response:

P303 + P361 + P353	IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water.
P304 + P340 + P310	IF INHALED: Remove person to fresh air and keep comfortable for breathing. Immediately call a POISON CENTER/ doctor.
P305 + P351 + P338 + P310	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/ doctor.
P391	Collect spillage.

Hazardous components:

- 1-Decanaminium, N-decyl-N, N-dimethyl-, hexanedioate (2:1)
- alcohols, C12-14-secondary, ethoxylated
- N-(3-aminopropyl)-N-dodecylpropane-1, 3-diamine

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590

Reagent handling

Ready for use

Storage and stability

Store protected from light.

Shelf life at 15-25 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	4 weeks

Materials provided

See “Reagents – working solutions” section for reagents.

Materials required (but not provided)

See “Order information” section

Assay

Use EcoTergent as specified in the respective instructions for use of the system reagents.

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 navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE

Roche Diagnostics GmbH
Sandhofer Strasse 116
68305 Mannheim, Germany
www.roche.com
+800 5505 6606

GGT-2

cobas®

γ-Glutamyltransferase ver.2 - Standardized against IFCC / Szasz

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057796190*	08057796500	γ-Glutamyltransferase ver.2 (400 tests)	System-ID 2060 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057796214*	08057796500	γ-Glutamyltransferase ver.2 (400 tests)	System-ID 2060 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

10759350190	Calibrator f.a.s. (12 x 3 mL)	Code 20401	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

GGT2-I: ACN 20600: assay standardized against IFCC

GGT2-S: ACN 20601: assay standardized against Szasz

Intended use

In vitro test for the quantitative determination of γ-glutamyltransferase (GGT) in human serum and plasma on **cobas c** systems.

Summary

Measurements of γ-glutamyltransferase (GGT) performed with this assay in human serum and plasma are used in the diagnosis and monitoring of hepatobiliary diseases, as well as a screening test for occult alcoholism.

Mature GGT is a dimeric glycoprotein weighing 68 kDa. It is found in the kidneys, liver, pancreas, and intestine, with the highest abundance in renal tissue. However, the primary source of GGT activity in the serum is the liver.[1]

In clinical practice, GGT serum levels are typically measured alongside a full blood count, bilirubin, albumin, transaminases (ALT and AST), and alkaline phosphatases (ALP) as an initial investigation for potential liver disease.[2] GGT is considered one of the most reliable indicators for the development of liver disease.[3] Multiple guidelines recommend GGT testing as part of the diagnostic workup and monitoring for various liver diseases. Additionally, GGT serves as a well-established marker for alcohol-related liver disease and excessive alcohol consumption.[4,5,6,7,8,9,10] Increased GGT is observed as a result of obesity, excess alcohol consumption or may be induced by drugs, including phenobarbital and phenytoin.[1]

In 1969, Szasz published the first kinetic procedure for GGT in serum using γ-glutamyl-p-nitroanilide as substrate and glycylglycine as acceptor.[11] In order to circumvent the poor solubility of γ-glutamyl-p-nitroanilide, Persijn and van der Slik investigated various derivatives and found the water-soluble substrate L-γ-glutamyl-3-carboxy-4-nitroanilide to be superior in terms of stability and solubility.[12] The results correlate with those derived using the original substrate.

In 2002, the International Federation of Clinical Chemistry (IFCC) recommended the standardized method for determining GGT including optimization of substrate concentrations, employment of NaOH, glycylglycine buffer and sample start.[13,14] The GGT liquid reagent follows the formulation recommendation according to Szasz, but was optimized for performance and stability. The assay is optionally standardized against the original IFCC and Szasz methods. The performance claims and data presented here are independent from the standardization.

Test principle[15]

Enzymatic colorimetric assay

γ-glutamyltransferase transfers the γ-glutamyl group of L-γ-glutamyl-3-carboxy-4-nitroanilide to glycylglycine.

L-γ-glutamyl-3-carboxy-4-nitroanilide + glycylglycine $\xrightarrow{\text{GGT}}$ L-γ-glutamyl-glycylglycine + 5-amino-2-nitrobenzoate

The amount of 5-amino-2-nitrobenzoate liberated is proportional to the GGT activity in the sample. It is determined by measuring the increase in absorbance photometrically.

Reagents - working solutions

R1 TRIS: 492 mmol/L, pH 8.25; glycylglycine: 492 mmol/L; preservative; additive

R3 L-γ-glutamyl-3-carboxy-4-nitroanilide: 22.5 mmol/L; acetate: 10 mmol/L, pH 4.5; stabilizer; preservative

R1 is in position B and R3 is in position C.

Precautions and warnings

For in vitro diagnostic use for laboratory 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.

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

Warning

H317 May cause an allergic skin reaction.

Prevention:

P261 Avoid breathing mist or vapours.

P272 Contaminated work clothing should not be allowed out of the workplace.

P280 Wear protective gloves.

Response:

P333 + P313 If skin irritation or rash occurs: Get medical advice/attention.

P362 + P364 Take off contaminated clothing and wash it before reuse.

Disposal:

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

GGT-2

γ-Glutamyltransferase ver.2 - Standardized against IFCC / Szasz

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590

Reagent handling

Ready for use

Storage and stability

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: Collect serum using standard sampling tubes.
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.

Centrifuge samples containing precipitates before performing the assay.

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

Stability:[16,17]	7 days at 15-25 °C
	7 days at 2-8 °C
	1 year at -20 °C (± 5 °C)

Freeze only once.

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

Test definition

Reporting time	10 min		
Wavelength (sub/main)	700/415 nm		
Reagent pipetting		Diluent (H_2O)	
R1	19 µL	57 µL	
R3	15 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	2.3 µL	–	–
Decreased	2.3 µL	10 µL	100 µL
Increased	2.3 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	Full 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 the original IFCC formulation (2002)[13] and against the GGT method published by Persijn and van der Slik (1976)[12], respectively.

Use the appropriate calibrator value for the corresponding application.

Quality control

For quality control, use control materials as listed in the "Order information" section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory's individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 12 weeks. 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

cobas c systems automatically calculate the analyte activity of each sample in the unit U/L (µkat/L).

Conversion factor: U/L × 0.0167 = µkat/L

Limitations - interferences

Criterion: Recovery within ± 4 U/L of initial values of samples ≤ 40 U/L and within ± 10 % for samples > 40 U/L.

Icterus:[18] No significant interference up to an I index of 50 for conjugated and 20 for unconjugated bilirubin (approximate conjugated bilirubin concentration: 855 µmol/L or 50 mg/dL and approximate unconjugated bilirubin concentration: 342 µmol/L or 20 mg/dL).

Hemolysis:[18] No significant interference up to an H index of 200 (approximate hemoglobin concentration: 124 µmol/L or 200 mg/dL).

Lipemia (Intralipid):[18] No significant interference up to an L index of 1500. 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.[19,20]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[21]

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Limits and ranges

Measuring range

3-1200 U/L (0.05-20.0 µkat/L)

Determine samples having higher activities via the rerun function. Dilution of samples via the rerun function is a 1:11 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 11.

GGT-2

γ-Glutamyltransferase ver.2 - Standardized against IFCC / Szasz

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 3 U/L (0.05 µkat/L)
Limit of Detection	= 3 U/L (0.05 µkat/L)
Limit of Quantitation	= 3 U/L (0.05 µkat/L)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the activity below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low activity samples.

The Limit of Detection corresponds to the lowest analyte activity which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte activity that can be reproducibly measured with a total error of 20 %. It has been determined using low activity γ-glutamyltransferase samples.

Expected values

U/L

Standardized against Szasz (Persijn, van der Slik)[22]

Men	8-61 U/L
Women	5-36 U/L

Standardized against IFCC

Reference Interval Study at 37 °C (corrected in 2005)[22,23]

Men (n = 216)	10-71 U/L
Women (n = 228)	6-42 U/L

Consensus values (IFCC)[24]

Men	< 60 U/L
Women	< 40 U/L

µkat/L

Standardized against Szasz (Persijn, van der Slik)[22,*]

Men	0.13-1.02 µkat/L
Women	0.08-0.60 µkat/L

Standardized against IFCC

Reference Interval Study at 37 °C (corrected in 2005)[22,23,*]

Men (n = 216)	0.17-1.19 µkat/L
Women (n = 228)	0.10-0.70 µkat/L

Consensus values (IFCC)[24]

Men	< 1.00 µkat/L
Women	< 0.67 µkat/L*

*calculated by unit conversion factor

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean* *U/L*	*SD* *U/L*	*CV* *%*
PCCC1[a)]	45.8	0.382	0.8
PCCC2[b)]	207	0.772	0.4
Human serum 1	8.57	0.449	5.2
Human serum 2	30.9	0.646	2.1
Human serum 3	62.7	0.679	1.1
Human serum 4	598	3.55	0.6
Human serum 5	1155	6.04	0.5

Intermediate precision	*Mean* *U/L*	*SD* *U/L*	*CV* *%*
PCCC1[a)]	45.6	0.463	1.0
PCCC2[b)]	207	1.67	0.8
Human serum 1	7.97	0.420	5.3
Human serum 2	30.6	0.703	2.3
Human serum 3	62.7	0.708	1.1
Human serum 4	598	3.69	0.6
Human serum 5	1161	10.6	0.9

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

γ-glutamyltransferase values for human serum and plasma samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 65

Passing/Bablok[25]	Linear regression
y = 1.014x - 1.98 U/L	y = 1.023x - 1.96 U/L
τ = 0.981	r = 0.999

The sample activities were between 4.81 and 941 U/L.

γ-glutamyltransferase values for human serum and plasma samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 75

Passing/Bablok[25]	Linear regression
y = 1.010x + 1.44 U/L	y = 1.019x + 0.534 U/L
τ = 0.982	r = 1.000

The sample activities were between 3.10 and 1001 U/L.

γ-glutamyltransferase values for human serum and plasma samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 77

Passing/Bablok[25]	Linear regression
y = 1.014x + 0.823 U/L	y = 1.013x + 1.03 U/L
τ = 0.962	r = 1.000

The sample concentrations were between 3.35 and 1157 U/L.

GGT-2

cobas®

γ-Glutamyltransferase ver.2 - Standardized against IFCC / Szasz

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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:

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

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+800 5505 6606

Roche

GLUC3

cobas®

Glucose HK Gen.3

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057800190*	08057800500	Glucose HK Gen.3 (3300 tests)	System-ID 2063 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057800214*	08057800500	Glucose HK Gen.3 (3300 tests)	System-ID 2063 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

10759350190	Calibrator f.a.s. (12 x 3 mL)	Code 20401	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

GLUC3: ACN 20630 (Serum/plasma)

GLUC3U: ACN 20631 (Urine)

GLUC3C: ACN 20632 (CSF)

Intended use

In vitro test for the quantitative determination of glucose in human serum, plasma, urine and CSF on **cobas c** systems.

Summary

Glucose measurement in serum and plasma with this device can be used to aid in diagnosis and monitoring of hypo- and hyperglycemia, in the context of an altered carbohydrate metabolism state.

In urine, glucose measurement with this device can be used as an aid in diagnosing glycosuria in the context of altered carbohydrate metabolism states and/or kidney disease.

In CSF glucose measurement with this device can be used to aid in diagnosis and monitoring of central nervous system infections, such as meningitis and encephalitis of different etiologies.

Glucose is the major carbohydrate present in the peripheral blood.[1] Oxidation of glucose is the major source of cellular energy in the body.[2] Glucose derived from dietary sources is converted to glycogen for storage in the liver or to fatty acids for storage in adipose tissue. The concentration of glucose in blood is controlled within narrow limits by many hormones, the most important of which are produced by the pancreas.[1,2] The most frequent cause of hyperglycemia is diabetes mellitus resulting from a deficiency in insulin secretion or action.[1] Hypoglycemia is less frequently observed.[2] A variety of conditions may cause low blood glucose levels such as insulinoma, insulin induced hypoglycemia, or hypopituitarism.[2,3]

Under normal circumstances, almost all the glucose filtered by the glomerulus is reabsorbed in the proximal convoluted tubule.[4] In case of hyperglycemia, as occurs in diabetes mellitus, the tubular transport capacity of glucose is overwhelmed and glucose appears in the urine (glycosuria).[5] Furthermore, glycosuria occurs in the absence of hyperglycemia when the reabsorption of glucose by renal tubules is compromised.[4]

CSF glucose level and the corresponding plasma glucose ratio are usually altered (low) in certain types of central nervous system infections, such as bacterial and tuberculous meningitis. Whereas CSF glucose level and plasma glucose ratio concentration is typically normal during most viral CNS infections.[6,7] However, the spectrum of CSF glucose levels in bacterial meningitis is wide and there is substantial overlap with the findings in viral infection. Therefore, clinical evaluation and other laboratory tests are needed to guide treatment decisions besides the results of the CSF glucose and CSF plasma glucose ratio.[8]

Test principle

Enzymatic reference method with hexokinase.[9,10]

Hexokinase catalyzes the phosphorylation of glucose to glucose-6-phosphate by ATP.

$$\text{Glucose} + \text{ATP} \xrightarrow{\text{HK}} \text{G-6-P} + \text{ADP}$$

Glucose-6-phosphate dehydrogenase oxidizes glucose-6-phosphate in the presence of NADP to gluconate-6-phosphate. No other carbohydrate is oxidized. The rate of NADPH formation during the reaction is directly proportional to the glucose concentration and is measured photometrically.

$$\text{G-6-P} + \text{NADP}^+ \xrightarrow{\text{G-6-PDH}} \text{gluconate-6-P} + \text{NADPH} + \text{H}^+$$

Reagents - working solutions

R1 MES buffer: 5.0 mmol/L, pH 6.0; Mg^{2+}: 24 mmol/L; ATP: ≥ 4.5 mmol/L; NADP: ≥ 7.0 mmol/L; preservative

R3 HEPES buffer: 200 mmol/L, pH 8.0; Mg^{2+}: 4 mmol/L; HK (yeast): ≥ 300 µkat/L; G-6-PDH (E. coli): ≥ 300 µkat/L; preservative

R1 is in position B and R3 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.

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

Warning

H315 Causes skin irritation.

H319 Causes serious eye irritation.

Prevention:

P264 Wash skin thoroughly after handling.

P280 Wear protective gloves/ eye protection/ face protection.

Response:

P302 + P352 IF ON SKIN: Wash with plenty of water.

P332 + P313 If skin irritation occurs: Get medical advice/attention.

P337 + P313 If eye irritation persists: Get medical advice/attention.

P362 + P364 Take off contaminated clothing and wash it before reuse.

Product safety labeling follows EU GHS guidance.
Contact phone: all countries: +49-621-7590

Reagent handling
Ready for use

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	26 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, K_2-EDTA, NaF/Na_2EDTA, KF/Na_2EDTA, NaF/K-Oxalate and NaF/citrate/Na_2-EDTA.

The stability of glucose in specimens is affected by storage temperature, bacterial contamination, and glycolysis. Plasma or serum samples without preservative (NaF) should be separated from the cells or clot within half an hour of being drawn. When blood is drawn and permitted to clot and to stand uncentrifuged at room temperature, the average decrease in serum glucose is ~ 7 % in 1 hour (0.28 to 0.56 mmol/L or 5 to 10 mg/dL). This decrease is the result of glycolysis. Glycolysis can be inhibited by collecting the specimen in fluoride tubes.[11]

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.

Centrifuge samples containing precipitates before performing the assay.

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

Stability:[10]	8 hours at 15-25 °C
	72 hours at 2-8 °C
Stability in fluoride plasma:[12]	3 days at 15-25 °C

Urine
Collect urine in a dark bottle. For 24-hour urine collections, glucose may be preserved by adding 5 mL of glacial acetic acid to the container before collection. Unpreserved urine samples may lose up to 40 % of their glucose after 24-hour storage at room temperature.[13] Therefore, keep samples on ice during collection.[10] If stabilizers are added to the sample, the sample index feature must not be used.

CSF
Cerebrospinal fluid may be contaminated with bacteria and often contains other cellular constituents. CSF samples should therefore be analyzed for glucose immediately or stored at 2-8 °C or -20 °C (± 5 °C).[13,10]

Freeze only once.

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, plasma, urine and CSF

Test definition

Reporting time	10 min	
Wavelength (sub/main)	700/340 nm	
Reagent pipetting		Diluent (H_2O)
R1	21 µL	106 µL
R3	8 µL	15 µL

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.5 µL	–	–
Decreased	3 µL	20 µL	60 µL
Increased	1.5 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Application for serum/plasma (ACN 20630)

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

Application for urine (ACN 20631)
Transfer of calibration from serum/plasma application (ACN 20630)

Application for CSF (ACN 20632)
Transfer of calibration from serum/plasma application (ACN 20630)

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

Traceability: This method has been standardized against ID/MS.

Quality control
For quality control, use control materials as listed in the "Order information" section. In addition, other suitable control material can be used.

Serum/plasma:	PreciControl ClinChem Multi 1, PreciControl ClinChem Multi 2
Urine:	Quantitative urine controls are recommended for routine quality control.
CSF:	Quantitative CSF controls are recommended for routine quality control.

The control intervals and limits should be adapted to each laboratory's individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 26 weeks.

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
cobas c systems automatically calculate the analyte concentration of each sample in the unit mmol/L (mg/dL, g/L).

Conversion factors:	mmol/L x 18.02 = mg/dL
	mmol/L x 0.1802 = g/L

GLUC3

Glucose HK Gen.3

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Limitations - interference

Serum/plasma

Criterion: Recovery within ± 0.39 mmol/L of initial values of samples ≤ 3.9 mmol/L and within ± 10 % of samples > 3.9 mmol/L.

Icterus:[14] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[14] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[14] No significant interference up to an L index of 1000. 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.[15,16]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[17]

Urine

Criterion: Recovery within ± 0.11 mmol/L of initial values of samples ≤ 1.1 mmol/L and within ± 10 % of samples > 1.1 mmol/L.

Urea: No significant interference from urea up to a concentration of 1800 mmol/L (10811 mg/dL).

Hemolysis: No significant interference up to an H index of 750 (approximate hemoglobin concentration: 466 µmol/L or 750 mg/dL).

Drugs: No interference was found at therapeutic concentrations using common drug panels.[16]

Tetracycline at therapeutic concentration gives falsely low results in urine samples.

CSF

Criterion: Recovery within ± 0.22 mmol/L of initial values of samples ≤ 2.2 mmol/L and within ± 10 % of samples > 2.2 mmol/L.

Icterus: No significant interference up to an I index of 60 for conjugated bilirubin (approximate conjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL.

Hemolysis: No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

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

NOTE: Glucose values achieved on some proficiency testing materials, when evaluated against a glucose oxidase-oxygen electrode comparison method, demonstrate an approximate 3 % positive bias on average.

ACTION REQUIRED

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

Limits and ranges

Measuring range

Serum, plasma, urine and CSF

0.11-41.6 mmol/L (2-750 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

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 0.11 mmol/L (2 mg/dL)
Limit of Detection	= 0.11 mmol/L (2 mg/dL)
Limit of Quantitation	= 0.11 mmol/L (2 mg/dL)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 20 %. It has been determined using low concentration glucose samples.

Expected values

mmol/L

Plasma[18]

Fasting	4.11-6.05 mmol/L

*Urine**[19]

1st morning urine	0.3-1.1 mmol/L
24-hour urine	0.3-0.96 mmol/L
	(average of 1350 mL urine/24 h)

* calculated by unit conversion factor

acc. to Tietz:[10]

Serum, plasma

Adults	4.11-5.89 mmol/L
60-90 years	4.56-6.38 mmol/L
> 90 years	4.16-6.72 mmol/L
Children	3.33-5.55 mmol/L
Neonates (1 day)	2.22-3.33 mmol/L
Neonates (> 1 day)	2.78-4.44 mmol/L

Urine

24-hour urine	< 2.78 mmol/24 h
Random urine	0.06-0.83 mmol/L

CSF

Children	3.33-4.44 mmol/L
Adults	2.22-3.89 mmol/L

mg/dL

Plasma[18]

Fasting	74-109 mg/dL

*Urine**[19]

1st morning urine	6-20 mg/dL
24-hour urine	6-17 mg/dL
	(average of 1350 mL urine/24 h)

* calculated by unit conversion factor

acc. to Tietz:[10]

Serum, plasma

Adults	74-106 mg/dL
60-90 years	82-115 mg/dL
> 90 years	75-121 mg/dL
Children	60-100 mg/dL
Neonates (1 day)	40-60 mg/dL
Neonates (> 1 day)	50-80 mg/dL

GLUC3

Glucose HK Gen.3

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Urine	
24-hour urine	< 2.78 mmol/24 h (< 0.5 g/24 h)
Random urine	1-15 mg/dL

CSF	
Children	60-80 mg/dL
Adults	40-70 mg/dL

CSF glucose values should be approximately 60 % of the plasma values and must always be compared with concurrently measured plasma values for adequate clinical interpretation.

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Serum/plasma

Repeatability	*Mean* *mmol/L*	*SD* *mmol/L*	*CV* *%*
PCCC1[a)]	5.61	0.0315	0.6
PCCC2[b)]	12.6	0.0523	0.4
Human serum 1	0.188	0.0174	9.2
Human serum 2	3.57	0.0181	0.5
Human serum 3	5.46	0.0233	0.4
Human serum 4	19.6	0.121	0.6
Human serum 5	38.6	0.188	0.5

Intermediate precision	*Mean* *mmol/L*	*SD* *mmol/L*	*CV* *%*
PCCC1[a)]	5.61	0.0559	1.0
PCCC2[b)]	12.8	0.106	0.8
Human serum 1	0.188	0.0188	10.0
Human serum 2	3.57	0.0212	0.6
Human serum 3	5.46	0.0297	0.5
Human serum 4	19.6	0.136	0.7
Human serum 5	38.6	0.216	0.6

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

Urine

Repeatability	*Mean* *mmol/L*	*SD* *mmol/L*	*CV* *%*
Control 1[c)]	1.09	0.0215	2.0
Control 2[c)]	16.4	0.0655	0.4
Human urine 1	0.227	0.0188	8.3
Human urine 2	0.733	0.0143	1.9
Human urine 3	4.10	0.0418	1.0
Human urine 4	22.0	0.182	0.8
Human urine 5	40.6	0.173	0.4

Intermediate precision	*Mean* *mmol/L*	*SD* *mmol/L*	*CV* *%*
Control 1[c)]	1.09	0.0278	2.5
Control 2[c)]	16.4	0.122	0.7
Human urine 1	0.215	0.0183	8.5
Human urine 2	0.744	0.0180	2.4
Human urine 3	4.07	0.0478	1.2
Human urine 4	22.0	0.452	2.1
Human urine 5	40.4	0.344	0.8

CSF

Repeatability	*Mean* *mmol/L*	*SD* *mmol/L*	*CV* *%*
Control 1[c)]	3.31	0.0119	0.4
Control 2[c)]	1.66	0.00970	0.6
Human CSF 1	0.273	0.00831	3.0
Human CSF 2	2.16	0.0180	0.8
Human CSF 3	3.81	0.0172	0.5
Human CSF 4	20.2	0.0824	0.4
Human CSF 5	39.9	0.193	0.5

Intermediate precision	*Mean* *mmol/L*	*SD* *mmol/L*	*CV* *%*
Control 1[c)]	3.34	0.0163	0.5
Control 2[c)]	1.66	0.0109	0.7
Human CSF 1	0.273	0.00966	3.5
Human CSF 2	2.16	0.0212	1.0
Human CSF 3	3.81	0.0240	0.6
Human CSF 4	20.2	0.0994	0.5
Human CSF 5	39.9	0.230	0.6

c) commercially available control material

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

Glucose values for human serum, plasma, urine and CSF samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Serum/plasma

Sample size (n) = 74

Passing/Bablok[20]	Linear regression
y = 1.000x - 0.0200 mmol/L	y = 0.997x - 0.00454 mmol/L
τ = 0.987	r = 1.000

The sample concentrations were between 0.320 and 40.3 mmol/L.

Urine

Sample size (n) = 67

Passing/Bablok[20]	Linear regression
y = 0.995x - 0.0447 mmol/L	y = 0.995x - 0.0402 mmol/L
τ = 0.982	r = 1.000

The sample concentrations were between 0.170 and 40.9 mmol/L.

CSF
Sample size (n) = 75

Passing/Bablok[20]	Linear regression
y = 1.000x + 0.00400 mmol/L	y = 1.001x + 0.0287 mmol/L
τ = 0.957	r = 0.999

The sample concentrations were between 0.200 and 40.8 mmol/L.

Glucose values for human serum, plasma, urine and CSF samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Serum/plasma
Sample size (n) = 69

Passing/Bablok[20]	Linear regression
y = 1.006x - 0.00351 mmol/L	y = 1.009x - 0.0366 mmol/L
τ = 0.977	r = 1.000

The sample concentrations were between 0.110 and 40.3 mmol/L.

Urine
Sample size (n) = 71

Passing/Bablok[20]	Linear regression
y = 1.012x - 0.0233 mmol/L	y = 1.022x - 0.0527 mmol/L
τ = 0.982	r = 1.000

The sample concentrations were between 0.130 and 40.3 mmol/L.

CSF
Sample size (n) = 66

Passing/Bablok[20]	Linear regression
y = 1.019x + 0.0138 mmol/L	y = 1.020x + 0.0122 mmol/L
τ = 0.975	r = 1.000

The sample concentrations were between 0.290 and 39.4 mmol/L.

Glucose values for human serum, plasma, urine and CSF samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Serum/plasma
Sample size (n) = 74

Passing/Bablok[20]	Linear regression
y = 0.995x - 0.00537 mmol/L	y = 0.999x - 0.0120 mmol/L
τ = 0.990	r = 1.000

The sample concentrations were between 0.200 and 40.2 mmol/L.

Urine
Sample size (n) = 67

Passing/Bablok[20]	Linear regression
y = 0.998x + 0.0116 mmol/L	y = 1.001x + 0.0119 mmol/L
τ = 0.950	r = 1.000

The sample concentrations were between 0.113 and 40.5 mmol/L.

CSF
Sample size (n) = 75

Passing/Bablok[20]	Linear regression
y = 0.987x - 0.0130 mmol/L	y = 0.989x - 0.0144 mmol/L
τ = 0.984	r = 1.000

The sample concentrations were between 0.154 and 40.5 mmol/L.

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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:

CONTENT	Contents of kit
→	Volume for reconstitution

GLUC3

Glucose HK Gen.3

GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH
Sandhofer Strasse 116
68305 Mannheim, Germany
www.roche.com
+800 5505 6606

HDLC4

cobas®

HDL-Cholesterol Gen.4

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057877190	08057877500	HDL-Cholesterol Gen.4 (700 tests)	System-ID 2071 002	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057877214*	08057877500	HDL-Cholesterol Gen.4 (700 tests)	System-ID 2071 002	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

12172623122	Calibrator f.a.s. Lipids (3 x 1 mL)	Code 20424	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

HDLC4: ACN 20710

Intended use

In vitro diagnostic test for the quantitative determination of the HDL-cholesterol concentration in human serum and plasma on **cobas c** systems.

Summary

Measurements of HDL-cholesterol, performed with this assay in human serum or plasma, are used for screening, aid in diagnosis and monitoring of dyslipidaemias as well as for assessment of cardiovascular risk such as in ASCVD and CHD.

High density lipoproteins (HDL) are responsible for the reverse transport of cholesterol from the peripheral cells to the liver. In the liver, cholesterol is transformed to bile acids which are then excreted into the intestine via the biliary tract.
Monitoring of HDL-cholesterol in serum or plasma is of clinical relevance as the HDL-cholesterol concentration is important in the assessment of atherosclerotic cardiovascular risk (ASCVD).[1] Elevated HDL-cholesterol concentrations protect against coronary heart disease (CHD), whereas reduced HDL-cholesterol concentrations, particularly in conjunction with elevated triglycerides, increase cardiovascular risk.[1]

Two cholesterol related variables that are predictive of cardiovascular disease (CVD) have emerged. These are non-HDL-cholesterol[2,3,4] (= total cholesterol - HDL-cholesterol) and the rate of cholesterol transfer from the macrophages to HDL, also described as cholesterol efflux capacity.[5]
Whereas both cholesterol and HDL-cholesterol can be readily determined with high accuracy, currently, non-HDL-cholesterol appears to be best suited for patient management.[2,3,4]

A variety of methods are available to determine HDL-cholesterol, including ultracentrifugation (reference method in combination with cholesterol measurement by the Abell-Kendall method), electrophoresis, high performance liquid chromatography (HPLC), precipitation, and direct methods.[6,7] Of these, the direct methods are used routinely. Roche HDLC4 is also a direct method. The automated HDLC4 assay uses detergents, cholesterol esterase (CHER), cholesterol oxidase (CHOD) and peroxidase to form a colored pigment that is measured optically.[8,9]

The HDLC4 assay meets the 1998 National Institutes of Health (NIH) / National Cholesterol Education Program (NCEP) goals for precision and accuracy.[10,11]

Test principle[8,9]

Homogeneous enzymatic colorimetric test.

Non-HDL lipoproteins such as LDL, VLDL and chylomicrons are combined with polyanions and a detergent forming a water-soluble complex. In this complex the enzymatic reaction of CHER and CHOD towards non-HDL lipoproteins is blocked.
Finally only HDL-particles can react with CHER and CHOD. The concentration of HDL-cholesterol is determined enzymatically by CHER and CHOD.
Cholesterol esters are broken down quantitatively into free cholesterol and fatty acids by CHER.

$$\text{HDL-cholesterol esters} + H_2O \xrightarrow{\text{CHER}} \text{HDL-cholesterol} + \text{RCOOH}$$

In the presence of oxygen, cholesterol is oxidized by cholesterol oxidase to Δ^4-cholestenone and hydrogen peroxide.

$$\text{HDL-cholesterol} + O_2 \xrightarrow{\text{CHOD}} \Delta^4\text{-cholestenone} + H_2O_2$$

In the presence of peroxidase, the hydrogen peroxide generated reacts with 4-amino-antipyrine and EMSE[a)] to form a dye. The color intensity of this dye is directly proportional to the cholesterol concentration and is measured photometrically.

$$2\ H_2O_2 + \text{4-amino-antipyrine} + \text{EMSE} + H^+ \xrightarrow{\text{Peroxidase}} \text{colored pigment} + 5\ H_2O$$

a) N-ethyl-N-(3-methylphenyl)-N'-succinylethylenediamine

Reagents - working solutions

R1 TAPSO[b)] buffer: 62.1 mmol/L, pH 7.77; polyanion: 1.25 g/L; EMSE: 1.08 mmol/L; ascorbate oxidase (cucurbita): ≥ 50 µkat/L; peroxidase (horseradish): ≥ 166.7 µkat/L; detergent; BSA: 2.0 g/L; preservative

R3 Bis-Tris[c)] buffer: 20.1 mmol/L, pH 6.70; cholesterol esterase (microorganism): ≥ 7.5 µkat/L; cholesterol oxidase (recombinant E. coli): ≥ 7.17 µkat/L; cholesterol oxidase (microorganism): ≥ 76.7 µkat/L; peroxidase (horseradish): ≥ 333 µkat/L; 4-amino-antipyrine: 1.48 mmol/L; BSA: 3.0 g/L; detergents; preservative

b) 2-Hydroxy-N-tris(hydroxymethyl)methyl-3-aminopropanesulfonic acid

c) Bis(2-hydroxyethyl)iminotris(hydroxymethyl)methane

R1 is in position B and R3 is in position C.

Precautions and warnings

For in vitro diagnostic use for laboratory 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.

Reagent handling

Ready for use

The intrinsic color of the reagent does not interfere with the test.

HDLC4

HDL-Cholesterol Gen.4

cobas®

Storage and stability

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, K_2- and K_3-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.

Centrifuge samples containing precipitates before performing the assay.

Collect blood by using an evacuated tube or syringe. Specimens should preferably be analyzed on the day of collection.

Fasting and non-fasting samples can be used.[12,13]

Stability in serum:	72 hours at 15-25 °C[13]
	7 days at 2-8 °C[13]
	12 months at -20 °C (± 5 °C)[14]
	24 months at -70 °C (± 5 °C)[15]

Freeze only once.

Stability in Li-heparin, K_2- and K_3-EDTA plasma:	72 hours at 15-25 °C[13]
	7 days at 2-8 °C[13]
	3 months at -20 °C (± 5 °C) °C[13]
	18 months at -70 °C (± 5 °C)[13]
	18 months at -80 °C (± 5 °C)[16]

Freeze only once.

It is reported that EDTA stabilizes lipoproteins.[17]

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

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

Test definition

Reporting time	10 min	
Wavelength (sub/main)	700/600 nm	
Reagent pipetting		Diluent (H_2O)
R1	80 µL	–
R3	27 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.6 µL	–	–
Decreased	8.0 µL	10 µL	90 µL
Increased	1.6 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s. Lipids
Calibration mode	Linear
Calibration frequency	Automatic full calibration - after reagent lot change
	Full calibration - 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 the designated CDC reference method (ultracentrifugation method).[10] The standardization meets the requirements of the “HDL Cholesterol Method Evaluation Protocol for Manufacturers” of the US National Reference System for Cholesterol, CRMLN (Cholesterol Reference Method Laboratory Network), November 1994.

Quality control

For quality control, use control materials as listed in the “Order information” section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory’s individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 12 weeks.

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

cobas c systems automatically calculate the analyte concentration of each sample in the unit mmol/L (mg/dL, g/L).

Conversion factors:	mmol/L x 38.66 = mg/dL
	mmol/L x 0.3866 = g/L

Limitations - interference[18]

Criterion: Recovery within ±0.1 mmol/L of initial values of samples ≤ 1 mmol/L (38.7 mg/dL) and within ±10 % for samples > 1 mmol/L.

Icterus:[19] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[19] No significant interference up to an H index of 1200 (approximate hemoglobin concentration: 745 µmol/L or 1200 mg/dL).

Lipemia (Intralipid):[19] No significant interference up to an L index of 2000. No significant interference from native triglycerides up to 13.7 mmol/L or 1200 mg/dL. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Other: Elevated concentrations of free fatty acids and denatured proteins may cause falsely elevated HDL-cholesterol results.

Ascorbic acid: No significant interference from ascorbic acid up to a concentration of 2.84 mmol/L (50 mg/dL).

Abnormal liver function affects lipid metabolism; consequently, HDL and LDL results are of limited diagnostic value. In some patients with abnormal liver function, the HDL-cholesterol result may significantly differ from the DCM (designated comparison method) result due to the presence of lipoproteins with abnormal lipid distribution.[20]

Drugs: No interference was found at therapeutic concentrations using common drug panels.[21,22]

Statins (Simvastatin) and fibrates (Bezafibrate) tested at therapeutic concentration ranges did not interfere.

N-acetylcysteine: No significant interference from N-acetylcysteine up to a concentration of 2.76 mmol/L (450 mg/L).

Acetaminophen intoxications are frequently treated with N-acetylcysteine. N-acetylcysteine at the therapeutic concentration when used as an antidote and the acetaminophen metabolite N-acetyl-p-benzoquinone imine (NAPQI) independently may cause falsely low HDL-cholesterol results.

Metamizole: Venipuncture should be performed prior to the administration of metamizole. Venipuncture immediately after or during the administration of metamizole may lead to falsely low results.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[23]

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Limits and ranges

Measuring range

0.08-3.88 mmol/L (3.09-150 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

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 0.08 mmol/L (3.09 mg/dL)
Limit of Detection	= 0.08 mmol/L (3.09 mg/dL)
Limit of Quantitation	= 0.08 mmol/L (3.09 mg/dL)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a precision of ≤ 30 % CV. It has been determined using low concentration HDL-cholesterol samples.

Expected values

mmol/L

	No risk	Moderate risk	High risk
Females[7,24,25]	> 1.68 mmol/L	1.15-1.68 mmol/L	< 1.15 mmol/L
Males[7,24,25]	> 1.45 mmol/L	0.90-1.45 mmol/L	< 0.90 mmol/L

mg/dL

	No risk	Moderate risk	High risk
Females[7,24,25]	> 65 mg/dL	45-65 mg/dL	< 45 mg/dL
Males[7,24,25]	> 55 mg/dL	35-55 mg/dL	< 35 mg/dL

National Cholesterol Education Program (NCEP) guidelines:[26]

< 40 mg/dL: Low HDL-cholesterol (major risk factor for CHD)

≥ 60 mg/dL: High HDL-cholesterol ("negative" risk factor for CHD)

HDL-cholesterol is affected by a number of factors, e.g. smoking, exercise, hormones, sex and age.

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

National Cholesterol Education Program (NCEP) guidelines are based on serum values. When classifying patients, serum or serum equivalent values should be used. Therefore the NCEP recommends using a factor of 1.03 to convert EDTA plasma values to serum values. A later study found EDTA plasma concentrations to be 4.7 % lower than those in serum.[27] To comply with the 1998 NCEP goal of a bias < 5 % it is recommended that each laboratory validates this conversion factor and enters it into the test parameters for HDL-cholesterol.[28]

Treatment goals for non-HDL-cholesterol have been proposed:[2]

	NCEP ATP III	ADA/AHA Guidelines for patients with increased cardiometabolic risk
Optional goal for very-high/highest risk patients (known CVD, diabetes with elevated risk)	< 3.37 mmol/L (< 130 mg/dL)	< 2.59 mmol/L (< 100 mg/dL)
Optional goal for those with established cardiovascular disease and multiple major risk factors	< 2.59 mmol/L (< 100 mg/dL)	
Optional goal for high-risk patients, CHD-risk-equivalent (Framingham 10-year risk score > 20 %/10 years, diabetes without other major risk factors)	< 3.37 mmol/L (< 130 mg/dL)	< 3.37 mmol/L (< 130 mg/dL)
Optional goal for moderately-high/intermediate risk patients (≥ 2 major CVD risk factors, Framingham 10-year risk score from 10-20 %)	< 4.14 mmol/L (< 160 mg/dL)	< 3.37 mmol/L (< 130 mg/dL)
Optional goal for high-risk patients, CHD-risk-equivalent (Framingham 10-year risk score > 20 %/10 years, diabetes without other major risk factors)	< 3.37 mmol/L (< 130 mg/dL)	

Specific performance data

Representative performance data on the analyzers are given below. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PCCC1[d)]	0.764	0.00290	0.4
PCCC2[e)]	1.45	0.00690	0.5

Human serum 1	0.148	0.00177	1.2
Human serum 2	1.07	0.00512	0.5
Human serum 3	1.49	0.00673	0.5
Human serum 4	1.92	0.00715	0.4
Human serum 5	3.53	0.0152	0.4
Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PCCC1[d)]	0.760	0.00630	0.8
PCCC2[e)]	1.44	0.00974	0.7
Human serum 1	0.148	0.00229	1.5
Human serum 2	1.07	0.00708	0.7
Human serum 3	1.49	0.0105	0.7
Human serum 4	1.92	0.0145	0.8
Human serum 5	3.54	0.0249	0.7

d) PreciControl ClinChem Multi 1

e) PreciControl ClinChem Multi 2

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

HDL-cholesterol values for human serum and plasma samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 70

Passing/Bablok[29]	Linear regression
y = 1.001x - 0.0175 mmol/L	y = 1.012x - 0.0274 mmol/L
τ = 0.976	r = 1.000

The sample concentrations were between 0.110 and 3.57 mmol/L.

HDL-cholesterol values for human serum and plasma samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 70

Passing/Bablok[29]	Linear regression
y = 1.011x - 0.0242 mmol/L	y = 1.028x - 0.0389 mmol/L
τ = 0.977	r = 1.000

The sample concentrations were between 0.110 and 3.57 mmol/L.

HDL-cholesterol values for human serum and plasma samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 75

Passing/Bablok[29]	Linear regression
y = 1.000x - 0.0200 mmol/L	y = 1.000x - 0.0159 mmol/L
τ = 0.994	r = 1.000

The sample concentrations were between 0.115 and 3.67 mmol/L.

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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:

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

Roche Diagnostics GmbH
Sandhofer Strasse 116
68305 Mannheim, Germany
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IRON2

cobas®

Iron Gen.2

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08057931190*	08057931500	Iron Gen.2 (700 tests)	System-ID 2077 001	**cobas c** 303, **cobas c** 503, **cobas c** 703
08057931214*	08057931500	Iron Gen.2 (700 tests)	System-ID 2077 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

10759350190	Calibrator f.a.s. (12 x 3 mL)	Code 20401	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

* Some kits shown may not be available in all countries.

English

System information

IRON2: ACN 20770

Intended use

In vitro test for the quantitative determination of iron in human serum and plasma on **cobas c** systems.

Summary

Iron measurements performed with this assay in human serum and plasma are used as an aid in diagnosis and monitoring of iron deficiency and iron overload disorders.

Iron is essential for many metabolic and biochemical processes. Similar to other micronutrients in the human body, iron is supplied with food. Ingested iron is mainly absorbed in the form of Fe^{2+} in the duodenum and proximal jejunum. The trivalent form and the heme-bound Fe^{3+}-component of iron in food has to be reduced by duodenal cytochrome B. About 1-2 mg of iron is absorbed and lost daily. Upon reaching the mucosal cells, Fe^{2+} ions become bound to transport proteins. In the cellular phase iron is either stored in cellular ferritin or transported to the circulation. Iron export into the circulation requires Fe^{2+} oxidation to Fe^{3+} by hephaestin (on cellular membrane) or ceruloplasmin (in the circulation), for loading onto transferrin. Circulating Fe ions are transported by transferrin-iron complexes. A maximum of 2 Fe^{3+} ions per protein molecule can be transported.[1]

Serum iron fluctuates with dietary intake and normal diurnal variation. Clinically, dysregulation of serum/plasma iron levels can be divided into iron deficiency and iron overload.[1,2] Iron deficiency disorders can be due to increased demands (e.g. growth, pregnancy), limited external supply (e.g. malnutrition, inappropriate diet, malabsorption), increased loss (e.g. hemorrhage, hemodialysis, blood donation), or other conditions, such as chronic kidney disease resulting in renal anemia, inflammatory bowel disease, heart failure, obesity, bone marrow disease.[2,3] Iron deficiency occurs in several stages, defined by the extent of depletion, first of iron stores and then of iron available for hemoglobin synthesis. In the first stage, iron stores can be completely depleted without causing anemia. Further iron loss causes anemia (iron deficiency anemia, IDA), which is initially normocytic, with a normal absolute reticulocyte count. Deeper deficiency results in classic anemia findings with hypochromic (low mean corpuscular hemoglobin) and microcytic (low mean corpuscular volume) red blood cells.[3,4,5] Another type of anemia is macrocytic anemia (elevated mean corpuscular volume), which is not directly due to iron deficiency but is rather related to other causes, such as vitamin B12 and folate deficiency, bone marrow disorders (myelodysplasia), use of certain medications, alcohol abuse, liver disease, marked reticulocytosis, and hypothyroidism. Iron measurements can help define different causes of anemia.[6,7]

Iron overload disorders normally result in increased serum/plasma iron concentration, and can be due to a number of underlying conditions, most commonly hereditary haemochromatosis (excess iron derived from increased gastrointestinal absorption due to inactivating mutations in components of the hepcidin pathway) and thalassemia (increased concentrations of iron mainly caused from regular red blood cell transfusions and to a lesser extent by increased iron absorption).[2,8]

Numerous photometric methods have been described for the determination of iron. All have the following in common:

- Liberation of Fe^{3+} ions from the transferrin complex using acids or detergents.
- Reduction of Fe^{3+} ions to Fe^{2+} ions.
- Reaction of the Fe^{2+} ions to give a colored complex.[1]

The method described here is based on the FerroZine method without deproteinization.

Test principle

Colorimetric assay.

$$\text{Transferrin-Fe-complex} \xrightarrow{\text{pH} < 2.0} \text{apotransferrin} + Fe^{3+}$$

$$Fe^{3+} \xrightarrow{\text{ascorbate}} Fe^{2+}$$

$$Fe^{2+} + \text{FerroZine} \longrightarrow \text{colored complex}$$

Under acidic conditions, iron is liberated from transferrin. Lipemic samples are clarified by the detergent. Ascorbate reduces the released Fe^{3+} ions to Fe^{2+} ions which then react with FerroZine to form a colored complex. The color intensity is directly proportional to the iron concentration and can be measured photometrically.

Reagents - working solutions

R1	Citric acid: 200 mmol/L; thiourea: 115 mmol/L; detergent
R3	Sodium ascorbate: 150 mmol/L; FerroZine: 6 mmol/L; preservative

R1 is in position B and R3 is in position C.

Precautions and warnings

For in vitro diagnostic use for laboratory 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.

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

IRON2

Iron Gen.2

Danger

H318 Causes serious eye damage.

Prevention:

P280 Wear eye protection/ face protection.

Response:

P305 + P351 + P338 + P310 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/ doctor.

Hazardous components:

- Poly(oxy-1,2-ethanediyl), .alpha.-isotridecyl-.omega.-hydroxy-

EUH 208 Contains DIAZOLIDINYL UREA. May produce an allergic reaction.

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590

Reagent handling

Ready for use

Storage and stability

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

When removing the **cobas c** pack from the instrument during use, please immediately store at 2-8 °C.

Do not shake the **cobas c** pack to avoid foaming.

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 plasma. Do not use EDTA or oxalate plasma.

Separate serum or plasma from the clot or cells within 1 hour.

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.

Centrifuge samples containing precipitates before performing the assay.

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

Stability:[9,10]	7 days at 15-25 °C
	3 weeks at 2-8 °C
	several years at -20 °C (± 5 °C)

Freeze only once.

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

Test definition

Reporting time	10 min		
Wavelength (sub/main)	700/570 nm		
Reagent pipetting		Diluent (H_2O)	
R1	75 µL	–	
R3	15 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	Diluent (NaCl)
Normal	6.4 µL	–	–
Decreased	9.0 µL	25 µL	50 µL
Increased	6.4 µL	–	–

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	Full calibration - after reagent lot change
	1-point recalibration using S1 - after **cobas c** pack green change - every 7 days on-board

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

Traceability: This method has been standardized against a primary reference material (SRM 937).

Quality control

For quality control, use control materials as listed in the “Order information” section. In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory’s individual requirements. It is recommended to perform quality control always after lot calibration and subsequently at least every 12 weeks.

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

cobas c systems automatically calculate the analyte concentration of each sample in the unit µmol/L (µg/dL, mg/L).

Conversion factors:	µmol/L x 5.59 = µg/dL
	µmol/L x 0.0559 = mg/L

Limitations - interference

Criterion: Recovery within ± 2.7 µmol/L of initial values of samples ≤ 26.9 µmol/L and within ± 10 % for samples > 26.9 µmol/L.

Icterus:[11] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Hemolysis:[11] No significant interference up to an H index of 200 (approximate hemoglobin concentration: 125 µmol/L or 200 mg/dL). Higher hemoglobin concentrations lead to artificially increased values due to contamination of the sample with hemoglobin-bound iron.

Lipemia (Intralipid):[11] No significant interference up to an L index of 1500. 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.[12, 13]

In patients treated with iron supplements or metal-binding drugs, the drug-bound iron may not properly react in the test, resulting in artificially low values.

In the presence of high ferritin concentrations > 1200 µg/L the assumption that serum iron is almost completely bound to transferrin is not valid anymore. Therefore, such iron results should not be used to calculate Total Iron Binding Capacity (TIBC) or percent transferrin saturation (% SAT).[14]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[15]

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Limits and ranges

Measuring range

0.90-179 µmol/L (5.00-1000 µg/dL, 0.05-10.0 mg/L)

Determine samples having higher concentrations via the rerun function. For samples with higher concentrations, the rerun function decreases the sample volume by a factor of 2.1. The results are automatically multiplied by this factor.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 0.9 µmol/L (5.03 µg/dL)
Limit of Detection	= 0.9 µmol/L (5.03 µg/dL)
Limit of Quantitation	= 0.9 µmol/L (5.03 µg/dL)

The Limit of Blank, Limit of Detection and Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 20 %. It has been determined using low concentration iron samples.

Expected values[16]

µmol/L

Adults:	5.83-34.5 µmol/L

µg/dL

Adults:	33-193 µg/dL

The concentration of iron in serum/plasma is dependent on ingestion of iron and is subject to circadian variations.[17]

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. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

Precision

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas c** 503 analyzer.

Repeatability	*Mean* µmol/L	*SD* µmol/L	*CV* %
PCCC1[a)]	18.6	0.111	0.6
PCCC2[b)]	41.4	0.163	0.4
Human serum 1	2.37	0.0817	3.4
Human serum 2	6.01	0.0830	1.4
Human serum 3	35.1	0.135	0.4
Human serum 4	89.2	0.307	0.3
Human serum 5	158	0.655	0.4

Intermediate precision	*Mean* µmol/L	*SD* µmol/L	*CV* %
PCCC1[a)]	18.6	0.212	1.1
PCCC2[b)]	41.6	0.369	0.9
Human serum 1	2.32	0.120	5.2
Human serum 2	5.95	0.149	2.5
Human serum 3	35.1	0.187	0.5
Human serum 4	89.2	0.337	0.4
Human serum 5	158	0.673	0.4

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

The data obtained on **cobas c** 503 analyzer(s) are representative for **cobas c** 303 analyzer(s) and **cobas c** 703 analyzer(s).

Method comparison

Iron values for human serum and plasma samples obtained on a **cobas c** 503 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 74

Passing/Bablok[18]	Linear regression
y = 1.004x + 0.0354 µmol/L	y = 1.003x + 0.00110 µmol/L
τ = 0.985	r = 1.000

The sample concentrations were between 1.20 and 169 µmol/L.

Iron values for human serum and plasma samples obtained on a **cobas c** 303 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 501 analyzer (x).

Sample size (n) = 98

Passing/Bablok[18]	Linear regression
y = 1.011x - 0.0750 µmol/L	y = 1.011x - 0.0772 µmol/L
τ = 0.993	r = 1.000

The sample concentrations were between 1.72 and 172 µmol/L.

IRON2

Iron Gen.2

Iron values for human serum and plasma samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 75

Passing/Bablok[18]	Linear regression
y = 1.000x + 0.0000 µmol/L	y = 1.001x - 0.0494 µmol/L
τ = 0.998	r = 1.000

The sample concentrations were between 2.55 and 176 µmol/L.

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13 Sonntag O, Scholer A. Drug interference in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.

14 Tietz NW, Rinker AD, Morrison SR. When Is a Serum Iron Really a Serum Iron? A Follow-up Study on the Status of Iron Measurements in Serum. Clin Chem 1996;42(1):109-111.

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18 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

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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:

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

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ISE Clean. Soln./Elecsys SysClean

ISE Cleaning Solution / Elecsys SysClean

Order information

REF		CONTENT	Analyzer(s) on which kit(s) can be used
11298500316	11298500500	ISE Cleaning Solution / Elecsys SysClean (5 × 100 mL)	**cobas c** **cobas e**

English

Intended use

For the cleaning of ISE units on Roche/Hitachi analyzers.
For the cleaning of Elecsys and **cobas e** immunoassay analyzers.

Summary

ISE Cleaning Solution / Elecsys SysClean is an alkaline cleaning solution with antimicrobial properties.

Reagents - working solutions

5 bottles, each containing 100 mL
Components:
Sodium hydroxide 3 mol/L
Sodium hypochlorite solution (< 2 % active chlorine)
Additive

Precautions and warnings

For in vitro diagnostic use for laboratory 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.

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

Danger

H290	May be corrosive to metals.
H314	Causes severe skin burns and eye damage.
H410	Very toxic to aquatic life with long lasting effects.

Prevention:

P273	Avoid release to the environment.
P280	Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection.

Response:

P303 + P361 + P353	IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water.
P304 + P340 + P310	IF INHALED: Remove person to fresh air and keep comfortable for breathing. Immediately call a POISON CENTER/ doctor.
P305 + P351 + P338 + P310	IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/ doctor.
P391	Collect spillage.

Hazardous components:

- sodium hydroxide
- sodium hypochlorite

Product safety labeling follows EU GHS guidance.
Contact phone: all countries: +49-621-7590

Reagent handling

Ready for use

Storage and stability

Store protected from light.
The solution is stable up to the stated expiration date when stored at 2-8 °C.

Materials provided

See “Reagents – working solutions” section for reagents.

Materials required (but not provided)

- General laboratory equipment
- **cobas c** or **cobas e** analyzers

Assay

For further information, please refer to the appropriate operator’s manual for the analyzer concerned, the respective application sheets and method sheets of all necessary components.

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:

CONTENT	Contents of kit
SYSTEM	Analyzers/Instruments on which reagents can be used
REAGENT	Reagent
CALIBRATOR	Calibrator
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE

Roche Diagnostics GmbH
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+800 5505 6606

ISE Deproteinizer

cobas®

Order information

REF		CONTENT	Analyzer(s) on which kit(s) can be used
20763071122	20763071500	ISE Deproteinizer (6 × 21 mL)	COBAS INTEGRA 400 plus **cobas c** systems
04838181001	20763071500	ISE Deproteinizer (2 × 11 mL)	**cobas c** 111

English

Intended use

ISE Deproteinizer is a cleaning solution intended for use with the COBAS INTEGRA and **cobas c** 111 ISE modules for cleaning the ion-selective electrodes and intended for use with **cobas c** systems for cleaning the ISE flow path.

Summary

ISE Deproteinizer is a cleaning solution used on the ISE module for the cleaning of the ion-selective electrodes, the mixing tower, and tubing during ISE maintenance. It is also used in the cleaning of the sample probe(s).

Reagents - working solutions

Sodium hypochlorite (NaOCl): approximately 1.2 %

Precautions and warnings

For in vitro diagnostic use for healthcare 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.

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

Warning

H315	Causes skin irritation.
H319	Causes serious eye irritation.
H410	Very toxic to aquatic life with long lasting effects.

Prevention:

P264	Wash skin thoroughly after handling.
P273	Avoid release to the environment.
P280	Wear protective gloves/ eye protection/ face protection.

Response:

P337 + P313	If eye irritation persists: Get medical advice/attention.
P391	Collect spillage.

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

COBAS INTEGRA 400 plus:
Place the ISE Deproteinizer on the ISE rack.

cobas c 111 **analyzer:**
Place the aliquoted ISE Deproteinizer in the sample area as indicated on the touch screen of the analyzer.

cobas c systems:
Use ISE Deproteinizer according to instructions of the **cobas c** operator's manual.

Storage and stability

COBAS INTEGRA 400 plus:

Shelf life at 2-8 °C:	See expiration date on the label
On-board in use and refrigerated on the analyzer:	4 weeks

Replace the bottle when alerted to do so by the system. Discard any remaining solution.

cobas c 111 **analyzer:**

Shelf life at 2-8 °C:	See expiration date on the label
after opening:	8 weeks at 2-8 °C or until the printed expiration date, whichever comes first

cobas c systems:

Shelf life at 2-8 °C:	See expiration date on the label

ISE Deproteinizer for maintenance on **cobas c systems** is to be prepared freshly.

Materials provided

See “Reagents – working solutions” section for reagents.

Materials required (but not provided)

Refer to the appropriate instructions for use and to the operator's manual for additional required materials.

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.

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 navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
REAGENT	Reagent
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

ISE Deproteinizer

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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ISE Diluent Gen.2

cobas®

ISE Diluent Gen.2

Order information

REF		CONTENT	Analyzer(s) on which reagents can be used
04880480190	04880480500	ISE Diluent Gen.2 (2 × 2000 mL)	**cobas c** 303 ISE, **cobas pro** ISE, **cobas** 8000 ISE
04880480214	04880480500	ISE Diluent Gen.2 (2 × 2000 mL)	**cobas c** 303 ISE, **cobas pro** ISE, **cobas** 8000 ISE
04522630190	04880480500	ISE Diluent Gen.2 (5 × 300 mL)	**cobas c** 311, **cobas c** 501

* Some kits shown may not be available in all countries.

English

Intended use

ISE Diluent Gen.2 is intended for diluting samples on the **cobas c** ISE analytical unit for the quantitative determination of sodium, potassium, and chloride in human origin serum, plasma or urine.

Reagents - ready for use

HEPES buffer: 10 mmol/L

Triethanolamine: 7 mmol/L

Preservatives

Precautions and warnings

For in vitro diagnostic use for laboratory 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.

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

EUH 208 Contains mixture of 5-chloro-2-methyl-4-isothiazolin-3-one and 2-methyl-2H-isothiazol-3-one (3:1). May produce an allergic reaction.

Contact phone: all countries: +49-621-7590

Product safety labeling follows EU GHS guidance.

Reagent handling

Use the ISE Diluent Gen.2 according to the instructions in the ISE section of the corresponding Operator's Manual/User Guide.

Storage and stability

Store at 15-25 °C.

Stability:

Unopened: up to expiration date at 15-25 °C.

See label for expiration date.

On-board stability:

cobas c 303 ISE, **cobas pro** ISE, **cobas** 8000 ISE: 6 weeks

cobas c 311, **cobas c** 501: 2 weeks

NOTE: Dissolved gases can cause performance problems if present in high amounts in the ISE Diluent Gen.2. In this case, mix the contents of the bottle gently before use.

NOTE: If one of the reagent bottles is nearly empty, do not just refill the bottle with new reagent. Discard the old reagent bottle, including any remaining reagent.

Materials provided

See "Reagents – working solutions" section for reagents.

Materials required (but not provided)

Roche system reagents and clinical chemistry analyzer with **cobas c** ISE analytical units.

General laboratory equipment

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:

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

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Roche

ISE Reference Electrolyte

cobas®

Order information

REF		CONTENT	Analyzer(s) on which reagents can be used
08392013190	08392013500	ISE Reference Electrolyte (2 × 2000 mL)	**cobas** ISE neo, **cobas** pro ISE
08392013214	08392013500	ISE Reference Electrolyte (2 × 2000 mL)	**cobas** ISE neo, **cobas** pro ISE
10820652216	08392013500	ISE Reference Electrolyte (1 × 500 mL)	**cobas c** 303 ISE, **cobas** 8000 ISE
11360981216	08392013500	ISE Reference Electrolyte (5 × 300 mL)	**cobas c** 311, **cobas c** 501

* Some kits shown may not be available in all countries.

English

Intended use

The ISE Reference Electrolyte is intended for closing the measurement circuit on the **cobas c** ISE analytical unit for the quantitative determination of sodium, potassium, and chloride in human origin serum, plasma or urine.

Reagents - working solutions

1 mol/L potassium chloride

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.

Reagent handling

Use the ISE Reference Electrolyte according to the instructions in the ISE section of the corresponding Operator's Manual/User Guide.

Storage and stability

Store at 15-25 °C.

Stability:

Unopened: up to expiration date at 15-25 °C. See label for expiration date.

On-board stability:

cobas ISE neo, **cobas** pro ISE, **cobas c** 303 ISE, **cobas** 8000 ISE: up to expiration date

cobas c 311, **cobas c** 501: 4 weeks

NOTE: Dissolved gases can cause performance problems if present in high amounts in the Reference Electrolyte. In this case, mix the contents of the bottle gently before use.

NOTE: If one of the reagent bottles is nearly empty, do not just refill the bottle with new reagent. Discard the old reagent bottle, including any remaining reagent.

Materials provided

See "Reagents – working solutions" section for reagents.

Materials required (but not provided)

Roche system reagents and clinical chemistry analyzer with **cobas c** ISE analytical units.

General laboratory equipment

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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

Symbols

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

CONTENT	Contents of kit
REAGENT	Reagent
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

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Additions, deletions or changes are indicated by a change bar in the margin.

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Roche

11183982500V11.0

ISE Standard High

cobas®

REF 11183982216 | 11183982500 | 10 x 3 mL

English

System information

For use on **cobas c** analyzers the calibrator codes are:
ISE Standard High as S2: Code 503
ISE Standard High as S3: Code 763

Intended use

ISE Standard High is for use in the calibration of Ion Selective Electrodes on **cobas c** analyzers.

Summary

ISE Standard High is an aqueous preparation with gravimetrically defined electrolyte levels.

This product is for use in standardization of sodium, potassium, and chloride values on the Ion Selective Electrodes.

Some methods specified in the relevant value sheet may not be available in all countries.

Reagents – working solutions

10 ampules, 3 mL each

Reactive components:

160 mmol/L Na^+, 7 mmol/L K^+, 120 mmol/L Cl^-

For ISE Standard High as S2: For the **cobas c** analyzers (except for the **cobas c** 111 analyzer) the values are encoded in electronic files sent via the **cobas** link to the analyzers.

For ISE Standard High as S3: The values must be entered manually.

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.

Handling

Wear protective glasses when handling the ampule. Using a piece of gauze or other protective material, carefully snap the top off the ampule.

Use according to instructions in the ISE sections of your **cobas c** operator's manual.

The enclosed barcoded labels are intended exclusively for **cobas c** systems to identify the calibrator. Attach the barcoded labels to the tubes carrying the sample cups containing the calibrator material.

NOTE: When the barcoded tube is only used as a cup holder, it can be reused if the lot number of the ISE standard does not change.

Storage and stability

Store at 15-25 °C.

Stability:

Unopened:	Up to the stated expiration date at 15-25 °C.
After opening:	Opened ampules should be used immediately. Remaining content must not be stored to avoid inaccurate calibrations.

Materials provided

- See “Reagents – working solutions” section
- Barcoded labels

Materials required (but not provided)

- Roche system reagents and clinical chemistry analyzers
- General laboratory equipment

Assay

Use ISE Standard High as specified in the **cobas c** operator's manual.

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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

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
CALIBRATOR	Calibrator
→	Volume for reconstitution
GTIN	Global Trade Item Number

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Roche

Distribution in USA by:
Roche Diagnostics, Indianapolis, IN
US Customer Technical Support 1-800-428-2336

ISE Standard Low

cobas®

REF 11183974216 11183974500 10 x 3 mL

English

System information

For use on **cobas c** analyzers the calibrator code is 502.

Intended use

ISE Standard Low is for use in the calibration of Ion Selective Electrodes on **cobas c** analyzers.

Summary

ISE Standard Low is an aqueous preparation with gravimetrically defined electrolyte levels.

This product is for use in standardization of sodium, potassium, and chloride values on the Ion Selective Electrodes.

Some methods specified in the relevant value sheet may not be available in all countries.

Reagents – working solutions

10 ampules, 3 mL each

Reactive components:

120 mmol/L Na^+, 3 mmol/L K^+, 80 mmol/L Cl^-

For the **cobas c** analyzers (except for the **cobas c** 111 analyzer) the values are encoded in electronic files sent via the **cobas** link to the analyzers.

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.

Handling

Wear protective glasses when handling the ampule. Using a piece of gauze or other protective material, carefully snap the top off the ampule.

Use according to instructions in the ISE sections of your **cobas c** operator's manual.

The enclosed barcoded labels are intended exclusively for **cobas c** systems to identify the calibrator. Attach the barcoded labels to the tubes carrying the sample cups containing the calibrator material.

NOTE: When the barcoded tube is only used as a cup holder, it can be reused if the lot number of the ISE standard does not change.

Storage and stability

Store at 15-25 °C.

Stability:

Unopened:	Up to the stated expiration date at 15-25 °C.
After opening:	Opened ampules should be used immediately. Remaining content must not be stored to avoid inaccurate calibrations.

Materials provided

- See “Reagents – working solutions” section
- Barcoded labels

Materials required (but not provided)

- Roche system reagents and clinical chemistry analyzers
- General laboratory equipment

Assay

Use ISE Standard Low as specified in the **cobas c** operator's manual.

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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

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
CALIBRATOR	Calibrator
→	Volume for reconstitution
GTIN	Global Trade Item Number

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Distribution in USA by:
Roche Diagnostics, Indianapolis, IN
US Customer Technical Support 1-800-428-2336

K Electrode

cobas®

Potassium

Order information

REF	CONTENT	Analyzer(s) on which the electrode can be used
10825441001	K Electrode 1 (electrode)	**cobas c** 311 analyzer **cobas** 6000 analyzer series: **cobas c** 501 module **cobas** 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module **cobas pure** integrated solutions: **cobas c** 303 analytical unit **cobas pro** integrated solutions: **cobas pro** ISE analytical unit, **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Materials required (but not provided):

03149501001	REF Electrode (1 electrode)	
11360981216	ISE Reference Electrolyte (5 x 300 mL) ①②	
10820652216	ISE Reference Electrolyte (1 x 500 mL) ③④	
08392013190	ISE Reference Electrolyte (2 x 2000 mL) ⑤⑥	
04522320190	ISE Internal Standard Gen.2 (5 x 600 mL) ①②	
04880455190	ISE Internal Standard Gen.2 (2 x 2000 mL) ③④⑤	
09137742190	ISE Internal Standard Gen.2 conc. (1 x 510 mL) ⑥	
05979854190	Internal Standard Insert - ISE (Set of 20) ①②	
04522630190	ISE Diluent Gen.2 (5 x 300 mL) ①②	
04880480190	ISE Diluent Gen.2 (2 x 2000 mL) ③④⑤	
11298500316	ISE Cleaning Solution (5 x 100 mL)	
20763071122	ISE Deproteinizer (6 x 21 mL) ④⑤⑥	
03110435180	Deproteinizer (1 x 125 mL) ⑥	
04663632190	Activator (9 x 12 mL)	
11183974216	ISE Standard Low (10 x 3 mL)	Code 20502
11183982216	ISE Standard High (10 x 3 mL)	Codes 20503, 20763
12149435122	Precinorm U Plus (10 x 3 mL)	Code 20300
12149443122	Precipath U Plus (10 x 3 mL)	Code 20301
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392

ISE reagents on:

① **cobas c** 311 analyzer

② **cobas** 6000 analyzer series: **cobas c** 501 module

③ **cobas** 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module

④ **cobas pure** integrated solutions: **cobas c** 303 analytical unit

⑤ **cobas pro** integrated solutions: **cobas pro** ISE analytical unit

⑥ **cobas pro** integrated solutions: **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

English

System information

	ACN (Serum/ plasma)	ACN (Urine)	ACN (Plasma)	ACN (Serum)
	ISE K	**ISE K-U**	**ISE K-P**	**ISE K-S**
cobas c 311 analyzer, **cobas c** 501 module, **cobas** 8000 ISE 900 / 1800 module	990	990	---	---

	ACN (Serum/ plasma)	ACN (Urine)	ACN (Plasma)	ACN (Serum)
	ISE K	**ISE K-U**	**ISE K-P**	**ISE K-S**
cobas c 303 analytical unit, **cobas pro** ISE analytical unit	29080	29081	29082	29083

K Electrode

cobas®

Potassium

	ACN (Serum/ plasma)	ACN (Urine)	ACN (Plasma)	ACN (Serum)
	K	**K-U**	**K-P**	**K-S**
cobas ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit	29240	29241	29242	29243

Intended use

The K Electrode is a device intended for the in-vitro quantitative determination of potassium in human origin serum, plasma and urine.

Summary[1]

Electrolytes are involved in most major metabolic functions in the body.

Potassium is the major intracellular cation and is critical to neural and muscle cell activity.

Some causes of decreased potassium levels include reduced intake of dietary potassium or excessive loss of potassium from the body due to diarrhea, prolonged vomiting, or increased kidney excretion. Increased potassium levels may be caused by dehydration or shock, severe burns, diabetic ketoacidosis, and retention of potassium by the kidney.

Test principle

Ion-selective electrode, using automatically diluted serum/plasma or urine specimens. The potassium electrode is based on a neutral carrier (Valinomycin).[2]

Calculation

The equation given below is used for the calculation of sample and/or QC results:

$$C_S = C_{IS} \times 10^{\frac{E_S - E_{IS}}{\pm S}}$$

Where:

C_S	concentration of the ion in the sample
C_{IS}	concentration of the ion in the ISE Internal Standard
E_S	EMF of the sample
E_{IS}	EMF of the ISE Internal Standard
S	Slope of the electrode

The complete measurement system for a particular ion includes the ISE, a reference electrode and electronic circuits to measure and process the EMF to give the test ion concentration.

Precautions and warnings

For in vitro diagnostic use for trained laboratory technicians.

Warning

- Samples containing material of human origin are potentially infectious. Wear personal protective equipment when replacing or installing electrodes at analyzers. If any biohazardous material is spilled, wipe it up immediately and apply a disinfectant.
- If sample or waste contacts with your skin, wash the affected area immediately with soap and water, then apply a disinfectant. Consult a physician.
- When disposing of used electrodes, treat them as biohazardous.

Caution

- Do not use electrodes after the shelf life or on-board stability period has expired. Otherwise, it may lead to unstable sodium, potassium, and chloride results due to the unstable potential reading of electrodes.
- Do not use hemolyzed samples because of falsely higher potassium results. Potassium concentration in erythrocytes is 25 times higher than in normal plasma.
- Perform electrode flow path cleaning as stated in the Instructions for Use for applicable analyzers, at the end of a daily sample run. Improper electrode flow path cleaning may cause unstable reading of electrodes and it results in calibration failures.

As with any diagnostic test procedure, results should be interpreted taking all other test results and the clinical status of the patient into consideration.

In addition, pay attention to all precautions and warnings listed in the operator's manual of the analyzer.

NOTE: Boric acid (CAS Registry No. 10043-35-3) is contained in the gel solution inside the electrode at 0.2 % of the total weight as a preservative.

Storage and stability

Store at 7-40 °C.

See labels for expiration dates.

On-board stability

After installation the electrode is stable for the following time period:
2 months or 9000 tests, whichever comes first.

The electrodes should be replaced after this time period has expired. For replacement refer to instructions in the operator's manual of the applicable analyzers.

NOTE: When replacing the electrode in **cobas pro** or **cobas pure**, the user should scan the barcode affixed on the rear side of the package instead of the barcode placed on the product's label.

Slope range 50 to 68 mV/dec

NOTE: The slope ranges for newly installed electrodes should be in the upper half of the recommended electrode slope range.

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.

It is important to follow tube manufacturers recommended procedures at and after blood collection.

Separate from cells if analysis is not performed within 2 hours.[3]

Serum

Use serum free of hemolysis and gross lipemia, collected by standard venipuncture technique.

As described in the literature, potassium values in serum are increased compared to plasma. Serum potassium is released from platelets during clotting. The higher the platelet count, the greater the error.[4]

Plasma is preferable to serum as sample material for potassium determinations.

CAUTION: Serum separator tubes have to be used in accordance with the tube manufacturer's recommended procedures. If these procedures are not considered, it is possible to coat the sample probe with gel (interfering with proper sample level detection), or even to aspirate gel into the ISE system (resulting in a clogged system).

Plasma: Lithium heparin plasma

For potassium determinations, plasma is the specimen of choice.

CAUTION: Inadequate mixing of plasma tubes can cause introduction of fibrin microclots into and subsequent clogging of the ISE.

NOTE: For certain types of hematological neoplasias, (severe) pseudohyperkalemia using lithium heparin samples has been reported.[5,6,7]

NOTE: It is strongly recommended to avoid silicone-type gels, due to risk of silicon oil contaminations. In addition, tubes that exhibit a layer of clear liquid, which rises to the top of the serum after centrifugation, should not be used, in order to prevent coating the sample probes and interfering with ISE system. It is possible to clog the sample probes or the ISE tubing with gel or clots if these precautions are not taken.

Urine: Collect 24-hour urine without addition of preservatives and/or stabilizers. Store refrigerated during collection.

NOTE: Each laboratory should establish guidelines for determining acceptability of specimens and the corrective action to be taken if a specimen is considered unacceptable. Compile a laboratory-specific guideline.

Sample stability (serum, plasma): [8]

14 days at 15-25 °C

14 days at 2-8 °C

stable at (-15)-(-25) °C

K Electrode

Potassium

up to 10 freeze-thaw cycles possible.[9]

Sample stability (urine):[8,10]

14 days at 15-25 °C

stable at (-15)-(-25) °C

up to 6 freeze-thaw cycles possible.[11]

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[8] 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 "Order information" section

Materials required (but not provided)

See "Order information" section

General laboratory equipment

Application for serum, plasma and urine

Test definition

Serum/plasma

	Sample dilution	
Sample volume	*Sample*	*Diluent*
cobas c 311 analyzer, cobas c 501 module		
Normal	9.7 µL	291 µL / ISE Diluent
cobas 8000 ISE 900 / 1800 module, cobas c 303 analytical unit, cobas pro ISE analytical unit		
Normal	15 µL	450 µL / ISE Diluent
cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit		
Normal	15 µL	450 µL / System Water

Measuring range on *cobas c 311 analyzer, cobas c 501 module, cobas 8000 ISE 900 / 1800 module, cobas c 303 analytical unit, cobas pro ISE analytical unit, cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit:* 1.5-10 mmol/L

Analysis of potassium on ISE analytical units listed with serum and plasma specimens should yield a linear relationship from 1.5-10 mmol/L with a deviation from the linear line of less than 5 %.

The sample volumes given above under "Normal" are for samples, calibrators, and quality controls.

Urine

	Sample dilution	
Sample volume	*Sample*	*Diluent*
cobas c 311 analyzer, cobas c 501 module		
Normal	9.7 µL	291 µL / ISE Diluent
Decreased	6.5 µL	291 µL / ISE Diluent
cobas 8000 ISE 900 / 1800 module		
Normal	10 µL	450 µL / ISE Diluent
Increased	not applicable	not applicable
cobas c 303 analytical unit, cobas pro ISE analytical unit		
Normal	15 µL	450 µL / ISE Diluent
Decreased	10 µL	450 µL / ISE Diluent
cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit		
Normal	15 µL	450 µL / System Water
Decreased	10 µL	450 µL / System Water

Measuring range on *cobas c 311 analyzer, cobas c 501 module, cobas c 303 analytical unit, cobas pro ISE analytical unit, cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit:* 3-100 mmol/L

Analysis of potassium on ISE analytical units listed with urine specimens should yield a linear relationship from 3-100 mmol/L with a deviation from the linear line of less than 10 %.

Determine samples having higher concentrations via the rerun function. Dilution of samples via rerun function is a 1:46 dilution. Results from samples diluted using the rerun function are automatically multiplied by the dilution factor.

Measuring range on *cobas c 311 analyzer, cobas c 501 module, cobas c 303 analytical unit, cobas pro ISE analytical unit, cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit* for urine samples with decreased sample volume (Rerun): 101-150 mmol/L.

Analysis of potassium on ISE analytical units listed with urine specimens should yield a linear relationship from 101-150 mmol/L with a deviation from the linear line of less than 10 %.

The sample volumes given above under "Normal" are for samples, calibrators, and quality controls.

Measuring range on *cobas 8000 ISE 900 / 1800 module:* 3-100 mmol/L

Analysis of potassium on **cobas** 8000 ISE 900 / 1800 module with urine specimens should yield a linear relationship from 3-100 mmol/L with a deviation from the linear line of less than 10 %.

A diluton of samples via rerun function is not applicable for urinary potassium.

The sample volumes given above under "Normal" are for samples and quality controls.

For further information about the assay test definitions refer to the application parameters setting screen of the corresponding analyzer and assay.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 1 mmol/L
Limit of Detection	= 1 mmol/L
Limit of Quantitation	= 1.5 mmol/L
Limit of Quantitation for urinary potassium on **cobas** 8000 ISE 900 / 1800 module	= 3.0 mmol/L

The Limit of Blank, the Limit of Detection and the Limit of Quantitation were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A2 requirements.

The Limit of Blank is the 95th percentile value from n ≥ 60 measurements of analyte-free samples over several independent series. The Limit of Blank corresponds to the concentration below which analyte-free samples are found with a probability of 95 %.

The Limit of Detection is determined based on the Limit of Blank and the standard deviation of low concentration samples.

The Limit of Detection corresponds to the lowest analyte concentration which can be detected (value above the Limit of Blank with a probability of 95 %).

The Limit of Quantitation is the lowest analyte concentration that can be reproducibly measured with a total error of 30 %. It has been determined using low concentration potassium samples.

Values below Limit of Quantitation are not reliable due to possible higher uncertainty.

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.

Calibration

Calibration requires the following calibrators: ISE Standard Low (S1), ISE Standard High (S2), and ISE Standard High (S3).

K Electrode

cobas®

Potassium

The slope of the calibration curve is calculated from Standards 1 and 2. ISE Internal Standard / ISE Internal Standard conc. is measured to provide E_{IS} for all measurements. Refer to the operator's manual of the analyzer for detailed calibration instructions.

Traceability: ISE Standard Low and ISE Standard High are prepared gravimetrically from highly purified inorganic salts.

Purity of these salts has been certified by argentometric titration, acidimetric titration or perchloric acid titration.

Calibration frequency

Calibration

- every 24 hours
- after ISE washing and maintenance
- after changing the reagent bottle ①
- after changing ISE Reference Electrolyte and/or Internal Standard conc. (depending on AutoCal settings) ②
- after replacing any electrode
- as required following quality control procedures

ISE reagents on:

① **cobas c** 311 analyzer, **cobas c** 501 module, **cobas** 8000 ISE 900 / 1800 module, **cobas c** 303 analytical unit, **cobas pro** ISE analytical unit

② **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Refer to the operator's manual for a detailed description of the Calibration/AutoCal function.

Quality control

For quality control, use control materials as listed in the "Order information" section. In addition, other suitable control material can be used.

Serum/plasma:	PreciControl ClinChem Multi 1, PreciControl ClinChem Multi 2
	Precinorm U Plus, Precipath U Plus
Urine:	Quantitative urine controls are recommended for routine quality control.

Quality controls should be performed daily and after every additional calibration.

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.

Refer to appropriate value sheets/package inserts for additional information.

Traceability: Each Roche Diagnostics control listed above has been standardized against ISE Standard Low and ISE Standard High.

Limitations - interference

Criterion: Recovery within ± 10 % of initial value.

Hemolysis - serum/plasma

Do not use hemolyzed samples.

Potassium concentration in erythrocytes is 25 times higher than in normal plasma. The level of interference may be variable depending on the exact content of erythrocytes.

An H index of ≤ 20 equals an increase of the potassium concentration of ≤ 0.1 mmol/L.[12]

Hemolysis - urine

Hemolysis:[13] No significant interference up to a hemoglobin concentration of 12.4 µmol/L or 20 mg/dL.

Icterus - serum/plasma

Icterus:[13] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

Lipemia - serum/plasma

Lipemia (Intralipid, SMOFlipid):[13] No significant interference up to an L index of 2000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

NOTE: Grossly lipemic specimens should be cleared by ultracentrifugation.[14]

Drugs

The following drugs have been tested and caused no significant interference when added to aliquots of pooled normal human serum up to the indicated concentration.

Serum/plasma

Acetaminophen (Paracetamol)	200 mg/L
Acetylsalicylic acid	1000 mg/L
Ampicillin-Na	1000 mg/L
Ascorbic acid	300 mg/L
Cefoxitin	2500 mg/L
Cyclosporine	5 mg/L
Doxycyclin	50 mg/L
Heparin	5000 IU/L
Ibuprofen	500 mg/L
Intralipid	10000 mg/L
Levodopa	20 mg/L
Methyldopa	20 mg/L
Metronidazole	200 mg/L
N-Acetylcysteine	1660 mg/L
Phenylbutazone	400 mg/L
Rifampicin	60 mg/L
Theophylline	100 mg/L

Urine

Acetaminophen (Paracetamol)	3000 mg/L
Ascorbic acid	4000 mg/L
Cefoxitin	12000 mg/L
Gentamycine sulfate	400 mg/L
Ibuprofen	4000 mg/L
Levodopa	1000 mg/L
Methyldopa	2000 mg/L
N-Acetylcysteine	10 mg/L
Ofloxacine	900 mg/L
Phenazopyridine	300 mg/L
Salicyluric acid	6000 mg/L
Tetracycline	300 mg/L

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 **cobas c** systems. All special wash programming necessary for avoiding carry-over is available via the **cobas** link. The latest version of the carry-over evasion list can be found with the NaOHD/SMS/SCCS Method Sheet. For further instructions, refer to the operator's manual.

Expected values[15]

Serum	Infant	4.1-5.3 mmol/L
	Child	3.4-4.7 mmol/L
	Adult	3.5-5.1 mmol/L

K Electrode

Potassium

Plasma	M	3.5-4.5 mmol/L
	F	3.4-4.4 mmol/L

Plasma potassium levels are reported to be lower than serum levels.

Urine 24 h	6-10 y, M	17-54 mmol/24 h
	6-10 y, F	8-37 mmol/24 h
	10-14 y, M	22-57 mmol/24 h
	10-14 y, F	18-58 mmol/24 h
	Adult	25-125 mmol/24 h

The urinary excretion of potassium varies significantly with dietary intake. The values given here are typical of people on an average diet.

NOTE: It is recommended that each laboratory establishes and maintains its own reference ranges. The values given here are only to be used as a guideline.

Precision

see precision data of the following analyzers in "Appendix 1: Precision":

cobas c 311 analyzer

cobas 6000 analyzer series: **cobas c** 501 module

cobas 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module

cobas pure integrated solutions: **cobas c** 303 analytical unit

cobas pro integrated solutions: **cobas pro** ISE analytical unit, **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Method comparison

see method comparison data of the following analyzers in "Appendix 2: Method comparison":

cobas c 311 analyzer

cobas 6000 analyzer series: **cobas c** 501 module

cobas 8000 modular analyzer series: **cobas** 8000 ISE 900 / 1800 module

cobas pure integrated solutions: **cobas c** 303 analytical unit

cobas pro integrated solutions: **cobas pro** ISE analytical unit, **cobas** ISE neo 900 analytical unit, **cobas** ISE neo 1800 analytical unit

Maintenance

ISE washing procedure for cobas c 311 analyzer, cobas c 501 module, cobas 8000 ISE 900 / 1800 module, cobas c 303 and cobas pro ISE analytical unit.

The system maintenance procedures and frequencies stated in the operator's manual of the respective analyzer must be performed each day at the end of the daily sample run or after an elevated sample throughput.

cobas c 311:	The specially designated positions on the sample disk are used.
Position W1:	ISE Cleaning Solution
Position W2:	Activator

The ISE Wash procedure has to be manually selected out of maintenance items.

cobas c 501:	The specially labeled wash rack (green) is used.
Position 1:	Multiclean (not necessary when only the ISE is cleaned)
Position 2:	ISE Cleaning Solution
Position 3:	Activator

The system recognizes the wash rack and switches automatically to cleaning mode.

cobas 8000 ISE:	The specially labeled wash rack (green) is used.
Position 1:	Cell Cleaning Solution (not necessary when only the ISE is cleaned)
Position 2:	ISE Cleaning Solution
Position 3:	Activator

The system recognizes the wash rack and switches automatically to cleaning mode.

cobas c 303, cobas pro ISE:	The specially labeled wash rack (green) is used.
Position 1:	ISE Cleaning Solution (used for weekly wash rack)
Position 2:	ISE Cleaning Solution (used for daily wash rack)
Position 3:	Activator

The system recognizes the wash rack and switches automatically to cleaning mode.

The ISE systems require conditioning after cleaning and prior to calibration.

NOTE: Always use fresh solutions for cleaning.

ISE washing procedure for cobas ISE neo analytical unit

cobas ISE neo:	The ISE system wash tube holder is used.
Position CS:	ISE Cleaning Solution
Position A:	Activator

The maintenance task "ISE system wash" is scheduled and initiated automatically. For detailed description, refer to the operator's manual.

On-board stability of auxiliary reagents: ISE Cleaning Solution 4 days, Activator 4 days.

NOTE: Always exchange the tubes on the ISE tube holder, using new tubes for fresh reagents. **You must not refill them**, as this will lead to deterioration of the ISE measuring unit(s). Refer to the operator's manual for further information.

Appendix 1: Precision

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

cobas c 311 analyzer

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

cobas 6000 analyzer series: cobas c 501 module

Repeatability and intermediate precision were determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP5 requirements (2 aliquots per run, 2 runs per day, 21 days). The following results were obtained:

	Repeatability			Intermediate precision		
Sample (on a **cobas c** 501)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
Plasma low	1.62	0.01	0.7	1.62	0.03	1.6
Plasma medium	4.97	0.04	0.7	4.97	0.04	0.8
Plasma high	9.46	0.06	0.6	9.46	0.07	0.7
Precinorm U	3.57	0.03	0.8	3.57	0.04	1.0
Precipath U	6.59	0.04	0.6	6.59	0.05	0.7
Urine low	5.12	0.03	0.6	5.12	0.04	0.7
Urine medium	52.08	0.32	0.6	52.08	0.67	1.3
Urine high	90.34	0.67	0.7	90.34	1.38	1.5
Liquichek 1	31.48	0.19	0.6	31.48	0.53	1.7
Liquichek 2	70.56	0.43	0.6	70.56	1.17	1.7

K Electrode

Potassium

cobas 8000 modular analyzer series: cobas 8000 ISE 900 / 1800 module

Repeatability and intermediate precision were determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP5 requirements (2 aliquots per run, 2 runs per day, 21 days). The following results were obtained:

	Repeatability			Intermediate precision		
Sample (on a **cobas** 8000)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
Plasma low	2.03	0.01	0.5	2.03	0.03	1.6
Plasma medium	5.01	0.02	0.3	5.01	0.03	0.7
Plasma high	9.56	0.03	0.3	9.56	0.06	0.6
Precinorm U	3.60	0.02	0.4	3.60	0.03	0.9
Precipath U	6.61	0.02	0.3	6.61	0.04	0.5
Urine low	3.47	0.01	0.3	3.47	0.04	1.1
Urine medium	50.70	0.26	0.5	50.70	0.63	1.2
Urine high	93.48	0.58	0.6	93.48	1.82	1.9
Liquichek 1	30.64	0.20	0.6	30.64	0.32	1.0
Liquichek 2	66.22	0.61	0.9	66.22	1.14	1.7

cobas pure integrated solutions: cobas c 303 analytical unit

The data obtained on **cobas pro** analyzer(s) are representative for **cobas c** 303 analyzer(s).

cobas pro integrated solutions: cobas pro ISE analytical unit

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas pro** ISE analytical unit.

	Repeatability			Intermediate precision		
Sample (on a **cobas pro** ISE analytical unit)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
PCCC1[a)]	3.66	0.02	0.4	3.66	0.04	1.1
PCCC2[b)]	6.77	0.02	0.3	6.77	0.05	0.8
Human plasma 1	1.65	0.01	0.7	1.65	0.05	2.9
Human plasma 2	5.82	0.02	0.4	5.82	0.04	0.6
Human plasma 3	2.97	0.01	0.5	2.97	0.05	1.6
Human plasma 4	7.50	0.03	0.4	7.50	0.06	0.8
Human plasma 5	9.52	0.04	0.4	9.52	0.11	1.1
Human serum 1	1.59	0.01	0.7	1.59	0.04	2.3
Human serum 2	5.96	0.02	0.4	5.96	0.03	0.5
Human serum 3	2.96	0.01	0.4	2.96	0.04	1.2
Human serum 4	7.79	0.03	0.4	7.79	0.05	0.7
Human serum 5	9.86	0.05	0.5	9.86	0.08	0.8
Liquichek 1	31.1	0.24	0.8	31.1	0.55	1.8
Liquichek 2	69.9	0.45	0.6	69.9	1.56	2.2
Human urine 1	3.31	0.03	0.8	3.31	0.05	1.5
Human urine 2	50.8	0.30	0.6	50.8	1.01	2.0
Human urine 3	32.4	0.26	0.8	32.4	0.58	1.8
Human urine 4	82.4	0.85	1.0	82.4	2.07	2.5
Human urine 5	95.7	1.20	1.3	95.7	2.56	2.7

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

cobas pro integrated solutions: cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit

Precision was determined using human samples and controls in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP05-A3 requirements with repeatability (n = 84) and intermediate precision (2 aliquots per run, 2 runs per day, 21 days). Results for repeatability and intermediate precision were obtained on the **cobas ISE neo** analytical unit.

	Repeatability			Intermediate precision		
Sample (on a **cobas** ISE neo analytical unit)	Mean mmol/L	SD mmol/L	CV %	Mean mmol/L	SD mmol/L	CV %
PCCC1[a)]	3.65	0.02	0.7	3.65	0.06	1.7
PCCC2[b)]	7.44	0.04	0.5	7.48	0.09	1.3
Human serum 1	1.76	0.01	0.6	1.76	0.06	3.2
Human serum 2	5.08	0.03	0.6	5.12	0.07	1.3
Human serum 3	3.64	0.02	0.5	3.64	0.04	1.2
Human serum 4	5.65	0.03	0.5	5.65	0.06	1.0
Human serum 5	9.70	0.05	0.5	9.76	0.08	0.8
Human plasma 1	1.69	0.01	0.7	1.69	0.06	3.5
Human plasma 2	5.03	0.03	0.7	5.06	0.06	1.1
Human plasma 3	3.50	0.02	0.6	3.52	0.05	1.4
Human plasma 4	5.56	0.03	0.5	5.56	0.06	1.0
Human plasma 5	9.70	0.05	0.5	9.77	0.09	0.9
Liquichek 1	30.3	0.21	0.7	30.3	0.60	2.0
Liquichek 2	71.7	0.67	0.9	71.7	2.00	2.8
Human urine 1	3.51	0.05	1.5	3.51	0.06	1.7
Human urine 2	48.5	0.39	0.8	48.5	1.22	2.5
Human urine 3	29.3	0.24	0.8	29.3	0.51	1.7
Human urine 4	75.2	0.61	0.8	75.2	2.09	2.8
Human urine 5	88.3	0.77	0.9	88.3	2.67	3.0

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

Appendix 2: Method comparison

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

cobas c 311 analyzer

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

cobas 6000 analyzer series: cobas c 501 module

ISE values for human plasma and urine samples obtained on **cobas c** 501 analyzers (y) using ISE Standard High (compensated) as S3 Calibrator, were compared to those determined with the corresponding reference method (x) and with a **cobas c** 501 analyzer using ISE Compensator as S3 Calibrator.

The reference method used was: Flame Photometer IL 943 for potassium.

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[16]	Coeff. (r)
x: flame photom. y: **cobas c** 501 (S3 = ISE Standard High)	Plasma / 106	1.59	9.56	y = 1.007x - 0.019	1.000

K Electrode

Potassium

Bias at 3.0 mmol/L = 0.002 (0.1 %) Bias at 5.8 mmol/L = 0.022 (0.4 %)					
x: **cobas c** 501 (S3 = ISE Compen-sator) y: **cobas c** 501 (S3 = ISE Standard High)	Plasma / 106	1.52	9.45	y = 1.006x + 0.024	1.000
Bias at 3.0 mmol/L = 0.042 (1.4 %) Bias at 5.8 mmol/L = 0.059 (1.0 %)					
x: flame photom. y: **cobas c** 501 (S3 = ISE Standard High)	Urine / 105	4.00	97.2	y = 1.018x - 0.397	1.000
Bias at 20 mmol/L = 0.757 (3.8 %) Bias at 80 mmol/L = 1.837 (2.3 %)					
x: **cobas c** 501 (S3 = ISE Compen-sator) y: **cobas c** 501 (S3 = ISE Standard High)	Urine / 105	4.05	97.4	y = 0.997x + 0.062	0.999
Bias at 20 mmol/L = 0.002 (0.0 %) Bias at 80 mmol/L = -0.178 (-0.2 %)					

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas 8000 modular analyzer series: cobas 8000 ISE 900 / 1800 module

ISE values for human plasma and urine samples obtained on a **cobas** 8000 analyzer (y) using ISE Standard High as S3 Calibrator, were compared with those determined using the corresponding reference method (x) and with **cobas c** 501 (x) using ISE Standard High as S3 Calibrator.

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[16]	Coeff. (r)
x: flame photom. y: **cobas** 8000 (S3 = ISE Standard High)	Plasma / 100	1.54	10.57	y = 1x + 0.05	0.9994
Bias at 3.0 mmol/L = 0.050 (1.7 %) Bias at 5.8 mmol/L = 0.050 (0.9 %)					
x: **cobas c** 501 (S3 = ISE Standard High) y: **cobas** 8000 (S3 = ISE Standard High)	Plasma / 100	1.59	10.59	y = 0.99x + 0.032	0.9999
Bias at 3.0 mmol/L = 0.002 (0.1 %) Bias at 5.8 mmol/L = -0.026 (-0.4 %)					
x: flame photom. y: **cobas** 8000 (S3 = ISE Standard High)	Urine / 101	3.1	99.5	y = 1.014x + 0.506	0.9997
Bias at 20 mmol/L = 0.786 (3.9 %) Bias at 80 mmol/L = 1.626 (2.0 %)					
x: **cobas c** 501 (S3 = ISE Standard High) y: **cobas** 8000 (S3 = ISE Standard High)	Urine / 101	2.97	102.04	y = 1.001x + 0.266	0.9998
Bias at 20 mmol/L = 0.286 (1.4 %) Bias at 80 mmol/L = 0.346 (0.4 %)					

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas pure integrated solutions: cobas c 303 analytical unit

ISE values for human plasma and serum samples obtained on a **cobas c** 303 ISE unit (y) were compared with a **cobas pro** ISE analytical unit (x) and with a **cobas c** 501 analyzer (x).

ISE values for human urine samples obtained on a **cobas c** 303 ISE unit (y) were compared with a **cobas pro** ISE analytical unit (x) and with a **cobas c** 501 analyzer (x).

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[16]	Coeff. (r)
x: **cobas pro** ISE y: **cobas c** 303 ISE	Plasma / 120	1.52	9.95	y = 0.990x + 0.029	1.000
Bias at 3.5 mmol/L = -0.006 (-0.2 %) Bias at 5.5 mmol/L = -0.025 (-0.5 %)					
x: **cobas c** 501 y: **cobas c** 303 ISE	Plasma / 120	1.55	10.0	y = 0.997x - 0.029	1.000
Bias at 3.5 mmol/L = -0.041 (-1.2 %) Bias at 5.5 mmol/L = -0.047 (-0.9 %)					

K Electrode

Potassium

x: **cobas pro** ISE y: **cobas c** 303 ISE	Serum / 116	1.62	9.81	y = 0.990x - 0.004	1.000
Bias at 3.5 mmol/L = -0.038 (-1.1 %) Bias at 5.5 mmol/L = -0.058 (-1.1 %)					
x: **cobas c** 501 y: **cobas c** 303 ISE	Serum / 116	1.56	9.78	y = 0.984x + 0.059	1.000
Bias at 3.5 mmol/L = 0.002 (0.1 %) Bias at 5.5 mmol/L = -0.031 (-0.6 %)					
x: **cobas pro** ISE y: **cobas c** 303 ISE	Urine / 120	3.55	98.9	y = 0.983x + 0.290	1.000
x: **cobas c** 501 y: **cobas c** 303 ISE	Urine / 119	3.49	93.0	y = 0.950x + 0.628	1.000

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas pro integrated solutions: cobas pro ISE analytical unit

ISE values for human plasma samples obtained on a **cobas pro** ISE analytical unit (y) were compared with a **cobas c** 501 analyzer (x).

ISE values for human urine samples obtained on a **cobas pro** ISE analytical unit (y) were compared with a **cobas c** 501 analyzer (x).

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[16]	Coeff. (r)
x: **cobas c** 501 y: **cobas pro** ISE	Plasma / 120	1.71	9.57	y = 0.998x - 0.00004	1.000
Bias at 3.5 mmol/L = -0.041 (-1.2 %) Bias at 5.5 mmol/L = -0.064 (-1.2 %)					
x: **cobas c** 501 y: **cobas pro** ISE	Serum / 120	1.62	10.0	y = 1.004x - 0.082	1.000
Bias at 3.5 mmol/L = -0.068 (-1.9 %) Bias at 5.5 mmol/L = -0.060 (-1.1 %)					
x: **cobas c** 501 y: **cobas pro** ISE	Urine / 119	3.28	96.4	y = 1.035x - 0.507	1.000

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

cobas pro integrated solutions: cobas ISE neo 900 analytical unit, cobas ISE neo 1800 analytical unit

ISE values for human plasma and serum samples obtained on a **cobas** ISE neo analytical unit (y) were compared with a **cobas c** 501 analyzer (x) and with a **cobas pro** ISE analytical unit (x).

ISE values for human urine samples obtained on a **cobas** ISE neo analytical unit (y) were compared with a **cobas c** 501 analyzer (x) and with a **cobas pro** ISE analytical unit (x).

Instruments	Sample Type/ N	Min.x	Max.x	P/B Regression[16]	Coeff. (r)
x: **cobas c** 501 y: **cobas** ISE neo	Serum / 118	1.62	9.93	y = 1.000x - 0.0400	1.000
Bias at 3.5 mmol/L = -0.0400 (-1.1 %) Bias at 5.5 mmol/L = -0.0400 (-0.7 %)					
x: **cobas pro** ISE y: **cobas** ISE neo	Serum / 119	1.52	9.89	y = 1.000x - 0.0200	1.000
Bias at 3.5 mmol/L = -0.0200 (-0.6 %) Bias at 5.5 mmol/L = -0.0200 (-0.4 %)					
x: **cobas c** 501 y: **cobas** ISE neo	Plasma / 116	1.64	9.69	y = 1.000x - 0.0400	0.999
Bias at 3.5 mmol/L = -0.0400 (-1.1 %) Bias at 5.5 mmol/L = -0.0400 (-0.7 %)					
x: **cobas pro** ISE y: **cobas** ISE neo	Plasma / 115	1.62	9.72	y = 1.008x - 0.0378	1.000
Bias at 3.5 mmol/L = -0.00840 (-0.2 %) Bias at 5.5 mmol/L = 0.00840 (0.2 %)					
x: **cobas c** 501 y: **cobas** ISE neo	Urine / 113	3.41	93.4	y = 1.056x - 0.777	1.000
x: **cobas pro** ISE y: **cobas** ISE neo	Urine / 113	3.43	94.3	y = 1.035x - 0.489	1.000

Bias at the medical decision level (MDL) was calculated as follows:

Bias [mmol/L] = intercept + (slope x MDL) - MDL

Bias [%] = (Bias [mmol/L] x 100) / MDL

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K Electrode

Potassium

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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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

Symbols

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

Cont.	Quantity contained in the package
CONTENT	Quantity contained in the package
GTIN	Global Trade Item Number
INSTALL BEFORE	Latest date by which the electrode has to be installed on the analyzer
RoHS	Directive for the restriction of the use of certain hazardous substances in electrical and electronic equipment

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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