UREAL

cobas®

Urea/BUN

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08058806190	08058806500	Urea/BUN (600 tests)	System-ID 2119 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	

English

System information

UREAL: ACN 21191 (Serum/plasma)

URELU: ACN 21190 (Urine)

U-BUN: ACN 21192 (Serum/plasma)

UBUNU: ACN 21193 (Urine)

Intended use

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

Summary

Measurements of urea/urea nitrogen in human serum, plasma and urine, performed with this assay are used as screening tests and as an aid in diagnosis and monitoring of renal function.

Urea is the major end product of protein nitrogen metabolism. It is synthesized by the urea cycle in the liver from ammonia which is produced by amino acid deamination. Urea is excreted mostly by the kidneys but minimal amounts are also excreted in sweat and degraded in the intestines by bacterial action.[1]

Serum urea mass concentration is either specified for the complete urea molecule or for nitrogen equivalents [blood urea nitrogen (BUN)].[2] Determination of blood urea nitrogen is primarily used as a screening test for renal function. When used in conjunction with serum creatinine determinations it can aid in the differential diagnosis of the three types of azotemia: prerenal, renal, and postrenal. The urea to creatinine ratio has been proposed as a crude discriminator between prerenal and intrinsic azotemia.[1]

Elevations in blood urea nitrogen concentration are seen in inadequate renal perfusion, shock, diminished blood volume (prerenal causes), chronic nephritis, nephrosclerosis, tubular necrosis, glomerular-nephritis (renal causes), and urinary tract obstruction (postrenal causes). Transient elevations may also be seen during periods of high protein intake. Liver diseases may lead to unpredictable blood urea nitrogen concentrations, including abnormally low levels. Low blood urea nitrogen concentrations are not common, but can be found in cases such as malnutrition, lack of protein in the diet, or overhydration.[1,3]

Test principle

Kinetic test with urease and glutamate dehydrogenase.[4,5,6,7]

Urea is hydrolyzed by urease to form ammonium and carbonate.

$$\text{Urea} + 2\ H_2O \xrightarrow{\text{Urease}} 2\ NH_4^+ + CO_3^{2-}$$

In the second reaction 2-oxoglutarate reacts with ammonium in the presence of glutamate dehydrogenase (GLDH) and the coenzyme NADH to produce L-glutamate. In this reaction 2 moles of NADH are oxidized to NAD^+ for each mole of urea hydrolyzed.

$$NH_4^+ + \text{2-oxoglutarate} + NADH \xrightarrow{\text{GLDH}} \text{L-glutamate} + NAD^+ + H_2O$$

The rate of decrease in the NADH concentration is directly proportional to the urea concentration in the specimen and is measured photometrically.

Reagents - working solutions

R1 NaCl 9 %

R3 TRIS buffer: 220 mmol/L, pH 8.6; 2-oxoglutarate: 73 mmol/L; NADH: 2.5 mmol/L; ADP: 6.5 mmol/L; urease (jack bean): ≥ 300 µkat/L; GLDH (bovine liver): ≥ 80 µkat/L; preservative; nonreactive stabilizers

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.

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
Plasma: Li-heparin and K_2-EDTA plasma. Do not use ammonium heparin.

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

Bacterial growth in the specimen and high atmospheric ammonia concentrations as well as contamination by ammonium ions may cause erroneously elevated results. If stabilizers are added to the sample, the sample index feature must not be used.

Stability in *serum/plasma:*[8]	7 days at 15-25 °C
	7 days at 2-8 °C
	1 year at (-15)-(-25) °C

Freeze only once.

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Stability in *urine*:[8]	2 days at 15-25 °C 7 days at 2-8 °C 1 month at (-15)-(-25) °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)	700/340 nm		
Reagent pipetting		Diluent (H_2O)	
R1	8 µL	66 µL	
R3	28 µL	81 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.5 µL	–	–
Decreased	1.5 µL	25 µL	50 µL
Increased	1.5 µL	–	–

Application for urine

Test definition

Reporting time	10 min		
Wavelength (sub/main)	700/340 nm		
Reagent pipetting		Diluent (H_2O)	
R1	8 µL	66 µL	
R3	28 µL	81 µL	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.5 µL	2.0 µL	98 µL
Decreased	1.5 µL	1.3 µL	116 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 21191/21192)

Calibrators	S1: H_2O S2: C.f.a.s.
Calibration mode	Linear
Calibration frequency	Full calibration - after reagent lot change - every 4 weeks on-board - as required following quality control procedures

Application for urine (ACN 21190/21193)

Transfer of calibration from serum/plasma application (ACN 21191/21192)

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.

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 concentration of each sample in the unit mmol/L (mg/dL, g/L).

Conversion factors:

mmol/L urea x 6.006 = mg/dL urea

mmol/L urea x 0.06006 = g/L urea

mmol/L urea nitrogen x 2.801 = mg/dL urea nitrogen

mmol/L urea nitrogen x 0.02801 = g/L urea nitrogen

mg/dL urea x 0.467 = mg/dL urea nitrogen

When 24-hour urine is used as the specimen, multiply the result by the 24-hour volume to obtain values in g or mmol/24 hours.

Limitations - interference

Criterion: Recovery within ± 0.83 mmol/L of initial values of samples ≤ 8.3 mmol/L and within ± 10 % for samples > 8.3 mmol/L.

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[9]

Serum/plasma

Icterus:[10] 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:[10] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[10] No significant interference up to an L index of 1000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Ammonium ions may cause erroneously elevated results.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[11,12]

Urine

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.[12]

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

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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

0.5-40 mmol/L (3.0-240 mg/dL urea, 1.4-112 mg/dL urea nitrogen)

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

Urine

1-2000 mmol/L (6-12000 mg/dL urea, 2.8-5600 mg/dL urea nitrogen)

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:1.8 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 1.8.

Determine samples having concentrations lower than the technical limit of 40 mmol/L (240 mg/dL urea and 112 mg/dL urea nitrogen) via the rerun function. Samples are measured undiluted.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Serum/plasma

Limit of Blank	= 0.5 mmol/L
Limit of Detection	= 0.5 mmol/L
Limit of Quantitation	= 0.5 mmol/L

Urine

Limit of Blank	= 1.0 mmol/L
Limit of Detection	= 1.0 mmol/L
Limit of Quantitation	= 1.0 mmol/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 urea/urea nitrogen samples.

Expected values

mmol/L

Urea:

Serum/plasma[13]

Adults	2.76-8.07 mmol/L

Urine

24-hour urine[14]	428-714 mmol/24 h, corresponding to 286-595 mmol/L[a)]

a) Based on average urine output of 1.2-1.5 L/24 h

Urea nitrogen (BUN):

Serum/plasma[14]

Adults (18-60 years)	2.14-7.14 mmol/L
Adults (60-90 years)	2.86-8.21 mmol/L
Infants (< 1 year)	1.43-6.78 mmol/L
Infants/children	1.79-6.43 mmol/L

Urine

24-hour urine[14]	428-714 mmol/24 h, corresponding to 286-595 mmol/L[b)]

b) Based on average urine output of 1.2-1.5 L/24 h

mg/dL

Urea:

Serum/plasma[13]

Adults	16.6-48.5 mg/dL

Urine

24-hour urine[14]	25.7-42.9 g/24 h, corresponding to 1.71-3.57 g/dL[a)]

a) Based on average urine output of 1.2-1.5 L/24 h

Urea nitrogen (BUN):

Serum/plasma[14]

Adults (18-60 years)	6-20 mg/dL
Adults (60-90 years)	8-23 mg/dL
Infants (< 1 year)	4-19 mg/dL
Infants/children	5-18 mg/dL

Urine

24-hour urine[14]	12-20 g/24 h, corresponding to 801-1666 mg/dL[b)]

b) Based on average urine output of 1.2-1.5 L/24 h

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[c)]	6.53	0.0408	0.6
PCCC2[d)]	18.3	0.0690	0.4
Human serum 1	1.31	0.0416	3.2
Human serum 2	5.12	0.0441	0.9
Human serum 3	7.67	0.0451	0.6
Human serum 4	18.7	0.101	0.5

Human serum 5	31.0	0.124	0.4
Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
PCCC1[c)]	6.50	0.0745	1.1
PCCC2[d)]	18.4	0.198	1.1
Human serum 1	1.31	0.0459	3.5
Human serum 2	5.12	0.0659	1.3
Human serum 3	7.67	0.0931	1.2
Human serum 4	18.7	0.226	1.2
Human serum 5	31.0	0.350	1.1

c) PreciControl ClinChem Multi 1

d) PreciControl ClinChem Multi 2

Urine

Repeatability	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
Control 1[e)]	143	2.86	2.0
Control 2[e)]	239	3.68	1.5
Human urine 1	3.22	0.0435	1.4
Human urine 2	73.2	2.50	3.4
Human urine 3	407	3.28	0.8
Human urine 4	922	5.03	0.5
Human urine 5	1583	10.1	0.6
Intermediate precision	*Mean*	*SD*	*CV*
	mmol/L	*mmol/L*	*%*
Control 1[e)]	143	3.17	2.2
Control 2[e)]	239	4.32	1.8
Human urine 1	3.22	0.0547	1.7
Human urine 2	73.2	2.78	3.8
Human urine 3	411	4.93	1.2
Human urine 4	919	11.5	1.2
Human urine 5	1583	19.7	1.2

e) 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

Urea 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) = 94

Passing/Bablok[15]	Linear regression
y = 1.009x + 0.0202 mmol/L	y = 1.006x + 0.0265 mmol/L
τ = 0.986	r = 1.000

The sample concentrations were between 0.600 and 38.1 mmol/L.

Urine

Sample size (n) = 91

Passing/Bablok[15]	Linear regression
y = 0.962x - 0.432 mmol/L	y = 0.960x + 0.586 mmol/L
τ = 0.982	r = 1.000

The sample concentrations were between 71.0 and 1964 mmol/L.

Urea 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) = 89

Passing/Bablok[15]	Linear regression
y = 1.017x + 0.0905 mmol/L	y = 1.015x + 0.148 mmol/L
τ = 0.986	r = 1.000

The sample concentrations were between 0.700 and 35.4 mmol/L.

Urine

Sample size (n) = 73

Passing/Bablok[15]	Linear regression
y = 0.981x + 0.901 mmol/L	y = 0.973x + 4.74 mmol/L
τ = 0.960	r = 0.999

The sample concentrations were between 41.0 and 1875 mmol/L.

Urea 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) = 74

Passing/Bablok[15]	Linear regression
y = 1.000x + 0.0800 mmol/L	y = 1.000x + 0.0683 mmol/L
τ = 0.983	r = 1.000

The sample concentrations were between 0.821 and 39.5 mmol/L.

Urine

Sample size (n) = 74

Passing/Bablok[15]	Linear regression
y = 0.948x - 1.68 mmol/L	y = 0.942x + 0.489 mmol/L
τ = 0.980	r = 1.000

The sample concentrations were between 46.4 and 1912 mmol/L.

References

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

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

Symbols

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

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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