Application Sheet

Laboratory Name

Test Name: Creatinine Jaffé Gen.2

System information

For **cobas c** 311/501 analyzers:
CREJ2: ACN 690 (Rate blanked, compensated, serum and plasma)
CRJ2U: ACN 691 (Rate blanked, urine)
SCRE2: ACN 773 (STAT, compensated, serum and plasma, reaction time: 4)
SCR2U: ACN 774 (STAT, urine, reaction time: 4)

Intended use

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

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

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 glomerular filtration rate (GFR) less than 60 mL/min per 1.73 m^2 for three months or more, regardless of cause.
The assay of creatinine in serum or plasma is the most commonly used test to assess renal function. 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 re-absorbed by the tubules to any appreciable extent. A small but significant amount is also actively secreted.
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 based only on the creatinine concentration in serum or plasma have been made. Among the various approaches suggested, two have found wide recognition: that of Cockroft and Gault and that based on the results of the MDRD trial. While the first equation was derived from data obtained with the conventional Jaffé method, a newer version of the second is usable for IDMS-traceable creatinine methods. Both are applicable for adults. In children, the Bedside Schwartz formula should be used.[6,7,8,9]
In addition to the diagnosis and treatment of renal disease, 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.

Test principle[10,11,12]

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).

Creatinine + picric acid $\xrightarrow{\textit{Alkaline pH}}$ yellow-orange complex

Reagents - working solutions

R1	Potassium hydroxide: 900 mmol/L; phosphate: 135 mmol/L; pH ≥ 13.5; preservative; stabilizer
R3 (STAT R2)	Picric acid: 38 mmol/L; pH 6.5; non reactive buffer

R1 is in position B and R3 (STAT R2) is in position C.

Precautions and warnings

For in vitro diagnostic use.
Exercise the normal precautions required for handling all laboratory reagents.
Disposal of all waste material should be in accordance with local guidelines.
Safety data sheet available for professional user on request.
For USA: For prescription use only.
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.
H412	Harmful to aquatic life with long lasting effects.
EUH 001	Explosive when dry

Prevention:

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

Product safety labeling primarily follows EU GHS guidance.
Contact phone: 1-800-428-2336

Reagent handling

Ready for use

Storage and stability

CREJ2

Shelf life at 15-25 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	8 weeks
Diluent NaCl 9 %	
Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
On-board in use and refrigerated on the analyzer:	12 weeks

Specimen collection and preparation[13]

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.

Stability in *serum/plasma*:[14]	7 days at 15-25 °C
	7 days at 2-8 °C
	3 months at (-15)-(-25) °C
Stability in *urine* (without preservative):[14]	2 days at 15-25 °C
	6 days at 2-8 °C
	6 months at (-15)-(-25) °C

Centrifuge samples containing precipitates before performing the assay.

Materials provided

See “Reagents – working solutions” section for reagents.

Materials required (but not provided)

- See “Order information” section
- General laboratory equipment

In addition, other suitable control material can be used**.**

Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator’s manual for analyzer-specific assay instructions.
The performance of applications not validated by Roche is not warranted and must be defined by the user.

Application for serum and plasma

cobas c 501/502 **test definition**

Assay type	Rate A		
Reaction time / Assay points	10 / 42-52 - 24-34 (STAT 4 / 17-27)		
Wavelength (sub/main)	570/505 nm		
Reaction direction	Increase		
Units	µmol/L (mg/dL, mmol/L)		
Reagent pipetting		Diluent (H_2O)	
R1	13 µL	77 µL	
R3	17 µL	30 µL	

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	10 µL	–	–
Decreased	10 µL	20 µL	80 µL
Increased	10 µL	–	–

Enter the correction value for the non-specific protein reaction as the instrument factor **y = ax + b** for mg/dL or for µmol/L, where **a = 1.0** and **b = -0.3** (mg/dL) or **a = 1.0** and **b = -26** (µmol/L).

Application for urine

cobas c 501 **test definition**

Assay type	Rate A		
Reaction time / Assay points	10 / 42-52 - 24-34 (STAT 4 / 17-27)		
Wavelength (sub/main)	570/505 nm		
Reaction direction	Increase		
Units	µmol/L (mg/dL, mmol/L)		
Reagent pipetting		Diluent (H_2O)	
R1	13 µL	77 µL	
R3	17 µL	30 µL	

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	10 µL	6 µL	144 µL
Decreased	10 µL	2 µL	180 µL
Increased	10 µL	6 µL	144 µL

Calibration

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

Traceability: This method has been standardized against ID/MS.
For the USA, this method has been standardized against a primary reference material (SRM 914 and SRM 967 (ID/MS)).

Quality control

At least once daily run solutions at two levels of a quality control material with known concentrations.

Refer to Brown Clinic Quality Control Requirements, Rules and Reviews Policy

Refer to Brown Clinic Quality Control Specialty and Subspecialty Policy

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Calculation

Roche/Hitachi **cobas c** systems automatically calculate the analyte concentration of each sample.
Conversion factors: µmol/L x 0.0113 = mg/dL
µmol/L x 0.001 = mmol/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 2500 µmol/L (28.3 mg/dL) in urine.
Serum/plasma
Icterus (*CREJ2*):[16] 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).
Icterus (*SCRE2*):[16] No significant interference up to an I index of 2 for conjugated bilirubin and 3 for unconjugated bilirubin (approximate conjugated bilirubin concentration: 34 µmol/L or 2 mg/dL; approximate unconjugated bilirubin concentration: 51 µmol/L or 3 mg/dL).
Hemolysis:[16] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).
Lipemia (Intralipid):[16] No significant interference up to an L index of 800. 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.[17,18]
Exception: Antibiotics containing cephalosporin lead to significant false-positive values.[19,20]
Exception: Cefoxitin causes artificially high creatinine results.
Exception: 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 (≥ 30 mg/dL for *SCRE2* applications).[21] 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.[22]
In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[23]

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 a hemoglobin concentration of 621 µmol/L or 1000 mg/dL.
Glucose < 120 mmol/L (< 2162 mg/dL) and urobilinogen < 676 µmol/L (< 40 mg/dL) do not interfere.
Drugs: No interference was found at therapeutic concentrations using common drug panels.[18]
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 Roche/Hitachi **cobas c** systems. The latest version of the carry-over evasion list can be found with the NaOHD-SMS-SmpCln1+2-SCCS Method Sheets. For further instructions refer to the operator's manual. **cobas c** 502 analyzer: All special wash programming necessary for avoiding carry-over is available via the **cobas** link, manual input is not required.

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

Limits and ranges

Measuring range

Serum/plasma

15-2200 µmol/L (0.17-24.9 mg/dL)
The technical limit in the instrument setting is defined as 41-2226 µmol/L (0.463-25.2 mg/dL) due to the compensation factor of 26.
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

375-55000 µmol/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 and Limit of Detection
Serum/plasma (CREJ2)

Limit of Blank = 15 µmol/L (0.17 mg/dL)
Limit of Detection = 15 µmol/L (0.17 mg/dL)
The Limit of Blank and Limit of Detection were determined in accordance with the CLSI (Clinical and Laboratory Standards Institute) EP17-A 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 %).
Lower detection limit of the test

Serum/plasma (SCRE2)

15 µmol/L (0.17 mg/dL)
The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).
Urine (CRJ2U/SCR2U)
375 µmol/L (4.2 mg/dL)
The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Expected values

Serum/plasma

Adults[24]

Females	44-80 µmol/L	(0.50-0.90 mg/dL)
Males	62-106 µmol/L	(0.70-1.20 mg/dL)

Children[25]

Neonates (premature)	25-91 µmol/L	(0.29-1.04 mg/dL)
Neonates (full term)	21-75 µmol/L	(0.24-0.85 mg/dL)
2-12 m	15-37 µmol/L	(0.17-0.42 mg/dL)
1- < 3 y	21-36 µmol/L	(0.24-0.41 mg/dL)
3- < 5 y	27-42 µmol/L	(0.31-0.47 mg/dL)
5- < 7 y	28-52 µmol/L	(0.32-0.59 mg/dL)
7- < 9 y	35-53 µmol/L	(0.40-0.60 mg/dL)
9- < 11 y	34-65 µmol/L	(0.39-0.73 mg/dL)
11- < 13 y	46-70 µmol/L	(0.53-0.79 mg/dL)
13- < 15 y	50-77 µmol/L	(0.57-0.87 mg/dL)

Urine

1st morning urine[24]

Females	2470-19200 µmol/L	(28-217 mg/dL)
Males	3450-22900 µmol/L	(39-259 mg/dL)

24-hour urine[26]

Females	7000-14000 µmol/24 h	(740-1570 mg/24 h)
Males	9000-21000 µmol/24 h	(1040-2350 mg/24 h)

Creatinine clearance[26,27] 71-151 mL/min

Refer to reference for a prospective study on creatinine clearance in children.[28]
Roche has not evaluated reference ranges in a pediatric population.
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

For Known Interfering Substances section refer to package insert.
For Known Non-Interfering Substance refer to package insert.

For Additional Technical Information refer to package insert.

References

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Alternative method

Refer to Brown Clinic Back-up Testing Policy

Source document

Reagent Name: CREJ2
Method Sheet Version: V18.0 English

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
04810716 190	Creatinine Jaffé Gen.2 700 tests	System ID 07 6928 2	Roche/Hitachi **cobas c** 311, **cobas c** 501/502
10759350 190	Calibrator f.a.s. (12 x 3 mL)	Code 401	
10759350 360	Calibrator f.a.s. (12 x 3 mL, for USA)	Code 401	
12149435 122	Precinorm U plus (10 x 3 mL)	Code 300	
12149435 160	Precinorm U plus (10 x 3 mL, for USA)	Code 300	
12149443 122	Precipath U plus (10 x 3 mL)	Code 301	
12149443 160	Precipath U plus (10 x 3 mL, for USA)	Code 301	
10171743 122	Precinorm U (20 x 5 mL)	Code 300	
10171778 122	Precipath U (20 x 5 mL)	Code 301	
03121313 122	Precinorm PUC (4 × 3 mL)	Code 240	
03121291 122	Precipath PUC (4 × 3 mL)	Code 241	
04489357 190	Diluent NaCl 9 % (50 mL)	System-ID 07 6869 3	

Effective date

Effective date for this procedure:

Author

Source documentation compiled by Roche Diagnostics

Revised by: Heather J Hall, MBA, MT(ASCP), CG(ASCP)[cm] Date: 4/9/2018

Approved by: Aaron Shives MD (Signature on file Date: 4/11/2018

REVIEW – REVISION SUMMARY DOCUMENTATION

Date:	By:	Revision Summary: