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Creatinine

mod. Jaffe

Diagnostic reagent for quantitative in vitro determination of creatinine in human serum, plasma or urine on photometric systems

REF	Kit Size	Configuration
D03111B	1 x 1.25 mL	1 x 1 L R1 + 1 x 0.25 L R2
D95595	5 x 100 mL	4 x 100 mL R1 + 1 x 100 mL R2
D00616	5 x 50 mL	4 x 50 mL R1 + 1 x 50 mL R2
D00617	5 x 25 mL	4 x 25 mL R1 + 1 x 25 mL R2
D00618	5 x 10 mL	4 x 10 mL R1 + 1 x 10 mL R2
D66911	10 x 50 mL	10 x 40 mL R1 + 4 x 25 mL R2
D0422917	5 x 62.5 mL	4 x 62.5mL R1 + 1 x 62.5mL R2
DA0823	5 x 50 mL	5 x 40 mL R1 + 5 x 10 mL R2
DT1023	4 x 62.5 mL	4 x 50 mL R1 + 4 x 12.5mL R2
DK0722	5 x 50 mL	4 x 50 mL R1 + 1 x 50 mL R2
DE1823	4 x 62.5 mL	4 x 50 mL R1 + 4 x 12.5 mL R2
DB20317	4 x 62.5 mL	4 x 50 mL R1 + 4 x 12.5 mL R2

Additionally available:

D94592	1 x 3 mL	Creatinine Standard	
D98485	5 x 3 mL	Calibrator	Diacal Auto
D98485SV	1 x 3 mL	Calibrator	Diacal Auto
D98481	12 x 5 mL	Control normal	Diacon N
D14481	5 x 5 mL	Control normal	Diacon N
D98481SV	1 x 5 mL	Control normal	Diacon N
D98482	12 x 5 mL	Control abnormal	Diacon P
D14482	5 x 5 mL	Control abnormal	Diacon P
D98482SV	1 x 5 mL	Control abnormal	Diacon P
D08581	12 x 5 mL	Urine Ctrl. normal	Diacon Urine Level 1
D08581SV	1 x 5 mL	Urine Ctrl. normal	Diacon Urine Level 1
D08582	12 x 5 mL	Urine Ctrl. abnormal	Diacon Urine Level 2
D08582SV	1 x 5 mL	Urine Ctrl. abnormal	Diacon Urine Level 2

For professional in vitro diagnostic use only.

GENERAL INFORMATION

Method	Colorimetric, "mod. Jaffe", 2 point kinetic, increasing reaction
Shelf life	24 months
Storage	2 – 25 °C
Wavelength	Hg 492 nm (490 nm - 510 nm)
Temperature	20 – 25 °C / 37 °C
Sample	Serum, heparin plasma, urine

INTENDED USE

Diagnostic reagent for quantitative in vitro determination of creatinine in human serum, plasma or urine on photometric systems.

DIAGNOSTIC SIGNIFICANCE [1, 2]

Creatinine is a waste product excreted by the kidneys mainly by glomerular filtration. The concentration of creatinine in plasma of a healthy individual is fairly constant, independent from water intake, exercise and rate of urine production. Therefore, increased plasma creatinine values always indicate decreased excretion, i.e. impaired kidney function. The creatinine clearance enables a quite good estimation of the glomerular filtration rate (GFR) which allows better detection of kidney diseases and monitoring of renal function. For this purpose creatinine is measured simultaneously in serum and urine collected over a defined time period.

TEST PRINCIPLE

Creatinine forms a coloured orange-red complex in an alkaline picrate solution. The difference in absorbance at fixed times during conversion is proportional to the concentration of creatinine in the sample.

Creatinine + Picric acid ⟶ Creatinine picrate complex

REAGENT COMPOSITION

COMPONENTS	CONCENTRATION
Reagent 1	
Sodium Hydroxide	0.2 mol/L
Reagent 2	
Picric Acid	20 mmol/L

MATERIAL REQUIRED BUT NOT PROVIDED

- NaCl solution (9 g/L).
- Clinical chemistry analyser.

REAGENT PREPARATION

Substrate Start:
Reagents are ready to use.

Sample Start:
Mix 4 parts of Reagent 1 with 1 part of Reagent 2.
(= working reagent)

STORAGE AND STABILITY

Conditions: Protect from direct light.
Close immediately after use.
Do not freeze the reagent.
Avoid contamination.

Substrate Start:
Storage: at 2 – 25 °C
Stability: up to the expiration date
Sample Start (Working Reagent):
Stability: at 15 – 25 °C 5 hours

WARNINGS AND PRECAUTIONS

1. Reagent 1: Warning

H290: May be corrosive to metals.
H315: Causes skin irritation.
H319: Causes serious eye irritation.
P234: Keep only in original container.
P264: Wash hands and face thoroughly after handling.
P280: Wear protective gloves/protective clothing/eye protection.
P302+P352: IF ON SKIN: Wash with plenty of water/soap.
P332+P313: If skin irritation occurs: Get medical advice/attention.
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.
P390: Absorb spillage to prevent material damage.

2. Reagent 2: Warning

H290: May be corrosive to metals.
P234: Keep only in original container.
P280: Wear protective gloves/protective clothing/eye protection.
P390: Absorb spillage to prevent material damage.

3. High homogentisic acid concentrations in urine samples lead to false results.
4. In very rare cases, samples of patients with gammopathy might give falsified results [11].
5. Eltrombopag medication leads to falsely low or high results in patient samples.
6. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents.
7. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
8. For professional use only!

SPECIMEN COLLECTION AND STORAGE

Sample preparation (Urine): Dilute urine 1 + 49 with dist. water. Multiply result by 50. (The urine controls Diacon Urine must be prediluted in the same way as patient samples).

Stability [5]:

In serum/plasma	at 4 – 25 °C	7 days
	at - 20 °C	at least 3 months
In urine:	at 20 – 25 °C	2 days
	at 4 – 8 °C	6 days
	at - 20 °C	6 months

Freeze only once! Discard contaminated specimens.

STANDARD

(not included in the kit; has to be ordered separately)
Concentration 2 mg/dL (177 µmol/L)
Storage: 2 – 25 °C
Stability: up to the indicated expiration date
Protect from light! Close immediately after use! Do not freeze!

TEST PROCEDURE

Bring reagents and samples to room temperature.

Substrate Start

Pipette into test tubes	Blank	Std/Cal.	Sample
Reagent 1	1000 µl	1000 µL	1000 µL
Sample	-	-	50 µL
Std./Cal.	-	50 µL	-
Dist. water	50 µL	-	-
Mix. Incubate 0 - 5 min., then add:			
Reagent 2	250 µL	250 µL	250 µL
Mix. Incubate for exactly 1 min. and read A1 against reagent blank. Incubate for exactly 2 min. and read A2 against reagent blank. Calculate: ΔA = (A2 - A1) sample or standard/calibrator			

Sample Start

Pipette into test tubes	Blank	Std/Cal.	Sample
Working Reagent	1000 µL	1000 µL	1000 µL
Sample	-	-	50 µL
Std./Cal.	-	50 µL	-
Dist. water	50 µL	-	-
Mix. Incubate for exactly 1 min. and read A1 against reagent blank. Incubate for exactly 2 min. and read A2 against reagent blank. Calculate: ΔA = (A2 - A1) sample or standard/calibrator			

Automation

Special adaptations for automated analysers can be made on request.

INTERPRETATION OF RESULTS

Calculation
Serum/Plasma:

$$\text{Creatinine [mg/dL]} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ Std/Cal}} \times \text{Conc. Std/Cal [mg/dL]}$$

Urine:

$$\text{Creatinine [mg/dL]} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ Std/Cal}} \times \text{Conc. Std/Cal [mg/dl]} \times 50$$

Creatinine Clearance [7]

$$[mL/min/1.73\ m^2] = \frac{mg\ Creatinine/100\ mL\ Urine \times mL\ Urine}{mg\ Creatinine/100\ mL\ Serum \times min\ Urine\ collection\ time}$$

The calculated creatinine clearance refers to the average body surface of an adult (1.73 m^2).

Unit Conversion
Creatinine [mg/dL] x 88.4 = Creatinine [µmol/L]

COMPENSATED METHOD [3,4]

Picric acid which forms the coloured complex reacts unspecifically with interfering serum components, so-called pseudo-creatinines. This leads to falsely elevated creatinine values in serum and plasma samples especially in the low measuring range. To compensate these interferences the calibrator value for the compensated method indicated in the value sheet of Diacal Auto has to be used for calculation. Additionally 0.3 mg/dL (27 µmol/L) has to be subtracted from the calculated creatinine value. For use of the compensated method calibration with the calibrator Diacal Auto is strictly recommended. The method is applicable only for serum and plasma samples.

QUALITY CONTROL AND CALIBRATION

All controls with Creatinine values determined by this method can be used.
We recommend the Dialab serum controls **Diacon N** (control serum with values in the normal range) and **Diacon P** (control serum with values in the abnormal range) as well as the Dialab urine controls **Diacon Urine Level 1** (control urine normal) and **Level 2** (control urine abnormal).
Each laboratory should establish corrective action in case of deviations in control recovery.

Calibration
The assay requires the use of a creatinine standard or calibrator.
We recommend the Dialab **Creatinine Standard** and the Dialab multi calibration serum **Diacal Auto**

PERFORMANCE CHARACTERISTICS

LINEARITY, MEASURING RANGE

The test has been developed to determine creatinine concentrations within a measuring range from 0.2 – 15 mg/dL (18 – 1330 µmol/L). When values exceed this range, samples should be diluted 1 + 1 with NaCl solution (9 g/L) and the result multiplied by 2.

SENSITIVITY/LIMIT OF DETECTION

The lower limit of detection is 0.2 mg/dL (17.7 µmol/L).

PRECISION (at 37 °C)

Intra-assay n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	0.56	0.01	1.30
Sample 2	1.24	0.01	0.83
Sample 3	6.73	0.06	0.93

Inter-assay n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	0.81	0.03	3.63
Sample 2	1.60	0.01	0.87
Sample 3	5.73	0.05	0.85

SPECIFICITY/INTERFERENCES

no interference up to:

Ascorbic acid	30 mg/dL
Bilirubin	4 mg/dL
Hemoglobin	500 mg/dL
Triglycerides	2000 mg/dL

For further information on interfering substances refer to Young DS [10].

METHOD COMPARISON

A comparison of Dialab Creatinine (y) with a commercially available Jaffé method (x) using 68 human sera samples within a range of 0.6 – 10 mg/dL (53.0 – 884 µmol/L) gave following results:
y = 1.014 x – 0.031 mg/dL; r = 1.000.
A comparison of Dialab Creatinine compensated (y) with an enzymatic method (x) using 65 human sera samples within a range of 0.5 – 4.3 mg/dL (44.2 – 380 µmol/L) gave following results:
y = 0.986 x + 0.043 mg/dL; r = 0.998.

TRACEABILITY

The values of Creatinine Jaffé-method compensated and not compensated in the calibrator Diacal Auto have been made traceable to NIST (National Institute for Standardization) Standard Reference Material SRM 967 using level 1 and 2 and therefore to GC-IDMS (gas chromatography – isotope dilution mass spectrometry).

EXPECTED VALUES

Serum/plasma, Jaffé-method not compensated:

Adults [1]	mg/dL	µmol/L
Women	0.6 – 1.1	53 – 97
Men	0.7 – 1.3	62 – 115
Children [2,8]		
Neonate	0.5 – 1.2	44 – 106
Infant	0.4 – 0.7	35 – 62
Child	0.5 – 1.2	44 – 106

Serum/plasma, Jaffé-method compensated:

Adults [3]	mg/dL	µmol/L
Women	0.5 – 0.9	44 – 80
Men	0.7 – 1.2	62– 106
Children [9]		
Neonate	0.24 – 1.04	21 – 92
Infant	0.17 – 0.42	15 – 37
Child	0.24 – 0.87	21 – 77

24h Urine [1]:

Women	11 – 20 mg/kg/24h	97 – 177 µmol/kg/24h
Men	14 – 26 mg/kg/24h	124 – 230 µmol/kg/24h

Creatinine clearance [2]:

Women	95 - 160 mL/min/1.73 m^2
Men	98 - 156 mL/min/1.73 m^2

* Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

LIMITATIONS

- Eventual Creatinine (mod. Jaffe) carry-over to reagents Phosphorus Inorganic (Molybdate), Iron (Ferene), LDH-L (IFCC) and LDH-P (opt. DGKC). The actual carry-over depends on the analyser.

WASTE MANAGEMENT

Please refer to local legal requirements.

LITERATURE

1. Newman DJ, Price CP. Renal function and nitrogen metabolites. In: Burtis CA, Ashwood ER, editors. Tietz Textbook of Clinical Chemistry. 3rd ed. Philadelphia: W.B Saunders Company; 1999. p. 1204-1270.
2. Thomas L. Clinical Laboratory Diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft; 1998. p. 366-74.
3. Mazzachi BC, Peake MJ, Ehrhardt V. Reference Range and Method Comparison Studies for Enzymatic and Jaffé Creatinine Assays in Plasma and Serum and Early Morning Urine. Clin. Lab. 2000; 46: 53-55
4. Swanson AF, Swartzentruber M, Nolen PA et al. Multicenter Evaluation of the Boehringer Mannheim Compensated, Rate-Blanked Creatinine/Jaffe Application on BM/Hitachi Systems. Advances in Clinical Diagnostics. 1993. Boehringer Mannheim Corporation
5. Guder WG, Zawta B. Recommendations of the Working group on Preanalytical Quality of the German Society for Clinical Chemistry and the German Society for Laboratory Medicine: The quality of Diagnostic Samples. 1st ed Darmstadt: GIT Verlag 2001; p. 24-5,50-1
6. Levey AS, Coresh J, Greene T, Marsh J et al: Expressing the Modification of Diet in Renal Disease Study Equation for Estimating Glomerular Filtration Rate with Standardized Serum Creatinine Values. Clin Chem 2007; 53 (4): 766-72.
7. Junge W, Wilke B, Halabi A, Klein G. Determination of reference intervals for serum creatinine, creatinine excretion and creatinine clearance with an enzymatic and a modified Jaffé method. Clin Chim Acta 2004; 344: 137-148
8. Soldin SJ, Brugnara C, Wong EC, eds. Pediatric Reference Intervals. 6th ed. AACC Press, 2007: p.77-78
9. Schlebusch H, Liappis N, Klein G. Ultrasensitive CRP and Creatinine: Reference intervals from infancy to childhood. Clin Chem Lab Med. 2001; 39 Special supplement pp S1-S448; May 2001. PO-T042
10. Young DS. Effects of Drugs on Clinical Laboratory Tests. 5th ed. Vol. 1 and 2. Washington, CD: The American Association for Clinical Chemistry Press 2000.
11. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. ClinChemLabMed 2007;45(9):1240-1243.

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