CREATININE (Kin)

(Mod. Jaffe's Kinetic method)

CHEMCount

INTRODUCTION

Creatinine is a product of creatine nonenzymatic dehydration in skeletal muscle. The amount of creatinine generated and excreted by kidney is proportional to muscle mass and usually is higher in men than women. Daily creatinine generation remains fairly constant, with the exception of crushing injury or degenerative diseases that cause massive damage to muscle. Creatinine blood and urine level depends on glomelural filtration so creatinine clearance is excellent index of renal function.

METHOD PRINCIPLE

Modified Jaffe's method, without deproteinization. In alkaline solution picrate reacts with creatinine to form a yellow-red 2,4,6-trinitrocyclohexadienate. The colour intensity is proportional to the creatinine concentration.

KIT CONTENTS

Reagent Name	2 x 50 ml	2 x 100 ml
R1 Buffer Reagent	1 x 50 ml	1 x 100 ml
R2 Picric Acid Reagent	1 x 50 ml	1 x 100 ml
R3 Standard	2 ml	3 ml

Plese refer standard value mentioned in the vial.

The reagents when stored at R.T are stable up to expiry date printed on the package. The reagents are stable for 1 week on board the analyser at 2-10°C. Protect from light and avoid contamination.

WORKING REAGENT PREPARATION AND STABILITY

Assay can be performed with use of separate R1- **Buffer** and R2-**Picric Acid** reagents or with use of working reagent.

Stability of working reagent : 1 week at 2-8°C
5 days at 15-25°C

Working reagent should be stored in tightly closed vials! If stored in open vial, it is stable for 1 day at 15-25°C! Protect from light and avoid contamination.

CONCENTRATIONS IN THE TEST

Sodium hydroxide 500 mmol/l
Sodium picrate 7.7 mmol/l

WARNINGS AND NOTES

Product for in vitro diagnostic use only.

The reagents are usable when the absorbance of the working reagent is less than 0.750 (read against distilled water, wavelength λ=500 nm, cuvette l=1 cm, at temp. 25°C).

ADDITIONAL EQUIPMENT

- Automatic analyzer or photometer able to read at 500 nm (492 nm)
- Thermostat at 25ºc
- General laboratory equipment

SPECIMEN

Serum, EDTA or heparinized plasma free from hemolysis, 24-hours urine.

Urine Creatinine should be usually carried out in 24 hours urine collection. Thymol should be used as a preservative for collection. The urine sample should be mixed and diluted with 1: 25 with distilled water Specimen can be stored up to 7 days at 2-8ºC. For longer storage samples should be frozen at -20ºC. Nevertheless it is recommended to perform the assay with freshly collected samples.

PROCEDURE

These reagents may be used both for manual assay (Sample Start and Reagent Start method) and in several automatic analyzers. **Programme Sheets** are available

Wavelength 505 nm
Temperature 25°C
Cuvette 1 cm

Pipette into the cuvettes:

Reagent	Standard (S)	Test (T)
R1 BufferReagent	500 µl	500 µl
R2 Picric Acid Reagent	500 µl	500 µl
R3 Standard	100 µl	-
Sample	-	100 µl

Mix well and after exactly 30 sec read absorbance A1 of the test (T) and standard (S) against air. After next 60 sec repeat absorbance reading (A2) and calculate ΔA (A2 – A1) for the test and standard.

Calculation

Creatinine concentration = ΔA(T) / ΔA(S) x standard concentration

REFERENCE VALUES

Serum / Plasma	mg / dl
Female	0.6 to 1.1
Male	0.7 to 1.3
2 hour urine	mg / 24 hours
Female	11 to 20
Male	14 to 26

It is recommended for each laboratory to establish its own reference ranges for local population.

QUALITY CONTROL

To ensure adequate quality control, each run should include assayed normal and abnormal controls. If commercial controls are not available it is recommended that known value samples be aliquoted, frozen and used as controls

PERFORMANCE CHARACTERISTICS

- Sensitivity / Limit of Quantitation: 0.05 mg/dl
- Linearity: up to 30 mg/d, if creatinine concentration exceeds 30 mg/dl, dilute the sample with distilled water in the ratio of 1:9 and repeat the assay. Multiply the result by 10
- Specificity / Interferences
  Haemoglobin up to 12.5 g/dl, triglycerides up to 500 mg/dl, ascorbate up to 62 mg/l and bilirubin up to 20 mg/dl do not Interfere with the test.

WASTE MANAGEMENT

Please refer to local legal requirements.

LITERATURE

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SYSTEM PARAMETERS

Method	Fixed Time (2-Point)
Wavelength	505 nm
Zero Setting	Distilled Water
Temperature Setting	25° C
Incubation Temperature	25° C
Incubation Time	----
Delay Time	30 secs
Read Time	60 secs
No. of Reading	2
Interval Time	----
Sample Volume	0.1ml (100 ul)
Reagent Volume	1.0 ml (1000 ul)
Standard Concentration	Refer Standard vial
Units	mg/dl
Factor	----
Reaction Slope	Increasing
Linearity	30 mg/dl

IVD

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