URIC ACID

(Uricase / PAP method)

CHEM Count

INTRODUCTION

Uric acid is a product of purine catabolism. It is produced in the liver and excreted in the urine. Both, the amount of uric acid production and the efficiency of renal excretion, affect serum urate level. Elevated serum uric acid level is caused usually by gout, leukemia, diabetes mellitus, hyperfunction of parathyroid and thyroid, renal failure, renal calculosis. Urate concentration in serum depends on glomerular filtration, thus is useful for renal function monitoring.

METHOD PRINCIPLE

Enzymatic, colorimetric method with uricase and peroxidase.

uric acid + 2 H_2O + O_2 uricase > allantoine + CO_2 + H_2O_2

ADPS + 4-aminoantipyrine + 2 H_2O_2 POD> quinoneimine dye +4 H_2O

(coloured compound)

The colour intensity is proportional to the uric acid concentration.

KIT CONTENTS

Reagent Name	Pack Size
R1 - Uric Acid Reagent	2 X 50 ml
R2 - Standard	2 ml

Refer standard **Concentration** on the vial.

WORKING REAGENT PREPARATION AND STABILITY

The reagents are to be stored at 2-8ºC. Do not freeze the reagents.

CONCENTRATIONS IN THE TEST

Buffer PIPES (pH 7.8)	> 150 mmol/l
Chromogen	1.0 mmol/l
Ascorbate oxidase	> 100 mmol/l
Peroxidase (POD)	> 100 mmol/l
Uricase	> 100 mmol/l

Activators & stabilizers.

ADDITIONAL EQUIPMENT

Automatic analyzer or photometer able to read at 546 nm (Hg 530- 550 nm), Thermostat at 25ºC or 37ºC, General laboratory equipment.

SPECIMEN

Serum, heparinized plasma free from hemolysis.

Do not use EDTA and fluoride as anticoagulants

Specimen can be stored 3-5 days at 2-8ºC or 6 months 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 **on request.**

Wavelength	505 nm
Temperature	25ºC / 37ºC
Cuvette	1 cm

Pipette into the cuvettes:

Reagent	Blank (B)	Standard (S)	Test (T)
R1 Uric Acid Reagent	1000 µl	1000 µl	1000 µl
Bring up the temperature of determination. Then add,			
Distilled water	20 µl		
R2 - Standard		20 µl	
Sample			20 µl

Mix well, incubate for 15 min. at 20-25ºC or 10 min. at 37ºC. Read the absorbance of test A(T) and standard A(S) against blank (B).

CALCULATION

Uric acid concentration = A (T) / A(S) x standard concentration

REFERENCE VALUES

Female	2.5 - 6.8 mg/dl
Male	3.6 - 7.7 mg/dl

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: 0.2 mg/dl (11.9 µmol/l)

Linearity: up to 25 mg/dl

Specificity / Interferences

Haemoglobin up to 7.5 g/dl, ascorbate up to 62 mg/l, bilirubin up to 20 mg/dl and triglycerides up to 500 mg/dl do not interfere with the test.

LITERATURE

1. Thefeld C. et al.: Dtsch. Med. Wschr. 98, 380-384 (1973).
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5. Kaplan L.A., Pesce A.J., ed. Chemistry Theory, Analysis, and Correlation, 3rd ed. St Louis, MO: Mosby, 501-2 (1996).
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SYSTEM PARAMETER

Method	End Point
Wavelength	505 nm
Zero Setting	Reagent Blank
Temperature Setting	37° C
Incubation Temperature	37° C
Incubation Time	10 mins.
Delay Time	----
Read Time	----
No. of Reading	----
Interval Time	----
Sample Volume	0.020 ml (20 ul)
Reagent Volume	1.0 ml (1000 ul)
Standard Concentration	Refer Standard vial
Units	mg/dl
Factor	----
Reaction Slope	Increasing
Linearity	25 mg/dl

IVD

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