

Uric acid FS*

TBHBA

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

Order Information

Cat. No.	Kit size					
1 3021 99 10 021	R1 4 x 20 mL + R2 1 x 20 mL + 1 x 3 mL Standard					
1 3021 99 10 026	R1 5 x 80 mL + R2 1 x 100 mL					
1 3021 99 10 023	R1 1 x 800 mL + R2 1 x 200 mL					
1 3021 99 10 704	R1 8 x 50 mL + R2 8 x 12.5 mL					
1 3021 99 90 314	R1 10 x 20 mL + R2 2 x 30 mL					
1 3000 99 10 030	6 x 3 mL Standard					

Summary [1,2]

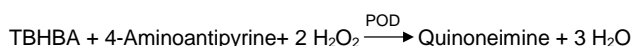
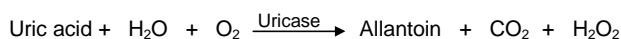
Uric acid and its salts are end products of the purine metabolism. In gout, the most common complication of hyperuricemia, increased serum levels of uric acid lead to formation of monosodium urate crystals around the joints. Further causes of elevated blood concentrations of uric acid are renal diseases with decreased excretion of waste products, starvation, drug abuse and increased alcohol consume as well as use of certain medications. High uric acid levels also constitute a indirect risk factor for coronary heart disease. Hypouricemia is seldom observed and associated with rare hereditary metabolic disorders.

Method

Enzymatic photometric test using TBHBA (2,4,6-Tribromo-3-hydroxybenzoic acid)

Principle

Uric acid is oxidized to allantoin by uricase. The generated hydrogen peroxide reacts with 4-aminoantipyrine and 2,4,6-tribromo-3-hydroxybenzoic acid (TBHBA) to quinoneimine.



Reagents

Components and Concentrations

R1:	Phosphate buffer	pH 7.0	100 mmol/L
	TBHBA (2,4,6-Tribromo-3-hydroxybenzoic acid)		1.25 mmol/L
R2:	Phosphate buffer	pH 7.0	100 mmol/L
	4-Aminoantipyrine		1.5 mmol/L
	K ₄ [Fe(CN) ₆]		50 μmol/L
	Peroxidase (POD)		≥ 10 kU/L
	Uricase		≥ 150 U/L

Standard: 6 mg/dL (357 μmol/L)

Storage Instructions and Reagent Stability

The reagents and the standard are stable up to the end of the indicated month of expiry, if stored at 2 – 8°C, protected from light and contamination is avoided. Do not freeze the reagents!

Note: It has to be mentioned, that the measurement is not influenced by occasionally occurring color changes, as long as the absorbance of the monoreagent is < 0.5 at 546 nm.

Warnings and Precautions

- Reagent 2 contains biological material. Handle the product as potentially infectious according to universal precautions and good laboratory practice.
- In very rare cases, samples of patients with gammopathy might give falsified results [8].

- N-acetylcysteine (NAC), acetaminophen and metamizole medication leads to falsely low results in patient samples.
- Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
- For professional use only!

Waste Management

Please refer to local legal requirements.

Reagent Preparation

The standard is ready to use.

Substrate Start

The reagents are ready to use.

Sample start

Mix 4 parts of R1 with 1 part of R2
(e.g. 20 mL R1 + 5 mL R2) = monoreagent
Stability: 3 months at 2 – 8°C
2 weeks at 15 – 25°C

Protect the monoreagent from light!

Materials required but not provided

NaCl solution 9 g/L
General laboratory equipment

Specimen

Serum, heparin plasma or EDTA plasma, urine

Stability in serum/plasma [3]:

6 months at –20°C
7 days at 4 – 8°C
3 days at 20 – 25°C

Freeze only once.

Discard contaminated specimens.

Stability in urine [4]:

4 days at 20 – 25°C

Dilute urine 1 +10 with dist. water and multiply the results by 11.

Discard contaminated specimens.

Assay Procedure

Application sheets for automated systems are available on request.

Wavelength	520 nm, Hg 546 nm, 500 - 550 nm
Optical path	1 cm
Temperature	20 – 25°C/ 37°C
Measurement	Against reagent blank

Substrate start

	Blank	Sample or standard
Sample or standard	-	20 μL
Dist. water	20 μL	-
Reagent 1	1000 μL	1000 μL
Mix, incubate 5 min., then add:		
Reagent 2	250 μL	250 μL
Mix, incubate 30 min. at 20 – 25°C or 10 min. at 37 °C.		
Read the absorbance against the reagent blank within 60 min.		

Sample start

	Blank	Sample or standard
Sample or standard	-	20 µL
Dist. water	20 µL	-
Mono reagent	1000 µL	1000 µL
Mix, incubate 30 min. at 20 – 25°C or 10 min. at 37 °C.		
Read the absorbance against the reagent blank within 60 min.		

Calculation

With standard or calibrator

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

Conversion factor

Uric acid [mg/dL] x 59.48 = Uric acid [µmol/L]

Calibrators and Controls

For the calibration of automated photometric systems, DiaSys TruCal U calibrator is recommended. The assigned values of the calibrator have been made traceable to the reference method gas chromatography-isotope dilution mass spectrometry (GC-IDMS). DiaSys TruLab N and P controls should be assayed for internal quality control. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat. No.	Kit size
TruCal U	5 9100 99 10 063	20 x 3 mL
	5 9100 99 10 064	6 x 3 mL
TruLab N	5 9000 99 10 062	20 x 5 mL
	5 9000 99 10 061	6 x 5 mL
TruLab P	5 9050 99 10 062	20 x 5 mL
	5 9050 99 10 061	6 x 5 mL
TruLab Urine Level 1	5 9170 99 10 062	20 x 5 mL
	5 9170 99 10 061	6 x 5 mL
TruLab Urine Level 2	5 9180 99 10 062	20 x 5 mL
	5 9180 99 10 061	6 x 5 mL

Performance Characteristics**Measuring range**

The test has been developed to determine uric acid concentrations within a measuring range from 0.07 – 20 mg/dL (4.2 – 1190 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.

Specificity/Interferences

No interference was observed by bilirubin up to 10 mg/dL and lipemia up to 2000 mg/dL triglycerides. Hemoglobin interferes starting with a concentration of 100 mg/dL. Ascorbic acid interferes even in minimal concentrations.

For measurement without interference by ascorbic acid DiaSys Uric acid FS TOOS is recommended. For further information on interfering substances refer to Young DS [7].

Sensitivity/Limit of Detection

The lower limit of detection is 0.07 mg/dL.

Precision (at 37°C)

Intra-assay precision n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	2.75	0.04	1.55
Sample 2	5.35	0.04	0.74
Sample 3	10.1	0.08	0.77

Inter-assay precision n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	2.68	0.04	1.52
Sample 2	5.23	0.09	1.63
Sample 3	9.98	0.11	1.06

Method Comparison

A comparison of DiaSys Uric acid FS TBHBA (y) with a commercially available test (x) using 70 samples gave the following results: $y = 1.02x - 0.44$ mg/dL; $r = 0.997$

Reference Range**Serum/Plasma**

	Female mg/dL (µmol/L)	Male mg/dL (µmol/L)
Adults [5]	2.6 – 6.0 (155 – 357)	3.5 – 7.2 (208 – 428)
Children [6]		
1 – 30 days	1.0 – 4.6 (59 – 271)	1.2 – 3.9 (71 – 230)
31 – 365 days	1.1 – 5.4 (65 – 319)	1.2 – 5.6 (71 – 330)
1 – 3 year(s)	1.8 – 5.0 (106 – 295)	2.1 – 5.6 (124 – 330)
4 – 6 years	2.0 – 5.1 (118 – 301)	1.8 – 5.5 (106 – 325)
7 – 9 years	1.8 – 5.5 (106 – 325)	1.8 – 5.4 (106 – 319)
10 – 12 years	2.5 – 5.9 (148 – 348)	2.2 – 5.8 (130 – 342)
13 – 15 years	2.2 – 6.4 (130 – 378)	3.1 – 7.0 (183 – 413)
16 – 18 years	2.4 – 6.6 (142 – 389)	2.1 – 7.6 (124 – 448)

Urine [1]

≤ 800 mg/24h (4.76 mmol/24h) assuming normal diet

≤ 600 mg/24h (3.57 mmol/24h) assuming low purine diet

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

Literature

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8. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007; 45(9):1240–1243.

Manufacturer

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