PRINCIPLE OF THE METHOD
Uric acid in sample is oxidized to allantoin in presence of the enzyme uricase and H₂O₂. The H₂O₂ reacts with TOOS and 4-aminooantipyrine in the presence of peroxidase to form a violet dye. The intensity of color formed is proportional to the uric acid concentration and can be measured photometrically between 510 and 560 nm.

KIT COMPONENTS
For in vitro diagnostic use only.

The components of the kit are stable until expiration date on the label. Keep away from direct light sources.

Reagent A
F100: 4 x 20 ml (liquid) blue cap
F250: 4 x 50 ml (liquid) blue cap
F600: 4 x 120 ml (liquid) blue cap
100F: 1 x 200 ml (liquid) blue cap
Composition: Phosphate buffer pH 7.0, TOOS, 4-aminooantipyrine 1.5 mM, uricase 450 UI, POCD 1000 UI, surfactant.

Reagent B
F100: 1 x 20 ml (liquid) red cap
F250: 1 x 50 ml (liquid) red cap
F600: 1 x 120 ml (liquid) red cap
100F: 1 x 200 ml (liquid) red cap
Composition: Good buffer pH 7.7 50 mM, 4-aminooantipyrine 0.38 mM, ascorbate oxidase ≥ 1000 U/l, surfactant.

MATERIALS REQUIRED BUT NOT SUPPLIED

REAGENT PREPARATION
Use separate reagents ready to use.

STABILITY:
- Stability: up to expiration date on labels at 2-8°C.
- Stability since first opening of vials: preferably within 60 days at 2-8°C.

PRECAUTIONS
Reagent may contain some non-reactive and preservative components. It is suggested to handle carefully it, avoiding contact with skin and swallow. To perform the test according to the general “Good Laboratory Practice” (GLP) guidelines.

SPECIMEN
Serum, plasma heparinized. Oxalate, citrate and fluoride could yield a small decrease of uric acid. Urine. The reabsorption defect may be acquired because of exposure to toxic agents. Overtreatment of hyperuricemia with hemoglobin ≤ 100 g/dl bilirubin ≤ 29 mg/dl lipids ≤ 970 mg/dl ascobic acid ≤ 50 mg/dl

TEST PROCEDURE
Wavelength: 550 nm
Lightpath: 1 cm
Temperature: 37°C

<table>
<thead>
<tr>
<th>dispense</th>
<th>blank</th>
<th>calibrator</th>
<th>sample</th>
<th>reagent A</th>
<th>1 ml</th>
<th>1 ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>water</td>
<td>50 µl</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<td>-</td>
</tr>
<tr>
<td>calibrator</td>
<td>-</td>
<td>50 µl</td>
<td>-</td>
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<td>-</td>
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<tr>
<td>sample</td>
<td>-</td>
<td>-</td>
<td>50 µl</td>
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</tbody>
</table>

Mix, incubate at 37°C for 5 minutes. Read absorbances of calibrator (Ac) and samples (Ax) against reagent blank.

<table>
<thead>
<tr>
<th>dispense</th>
<th>blank</th>
<th>calibrator</th>
<th>sample</th>
<th>reagent B</th>
<th>250 µl</th>
<th>250 µl</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mix, incubate at 37°C for 5 minutes. Read absorbances of calibrator (Ac) and samples (Ax) against reagent blank.</td>
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</table>

RESULTS CALCULATION
Serum/plasma sample:

uric acid mg/dl = \( (A_{Ax} - A_{Ac}) / (A_{Ac} - A_{Ac}) \) x 5 (standard value)

Random urine sample:

uric acid mg/dl = \( (A_{Ax} - A_{Ac}) / (A_{Ac} - A_{Ac}) \) x 5 x 10 (standard value and dilution)

24 hours urine sample (uric acid mg/24h):

uric acid mg/24h = \( (A_{Ax} - A_{Ac}) / (A_{Ac} - A_{Ac}) \) x 5 x 10 x diuresis (dl)