

Instructions for Use

Version: 1.0.1

Revision date: 9-Oct-26



Uric Acid (UA) Assay Kit

Catalog No.: abx098465

Size: 96 tests

Detection Range: 2.95 mg/L – 100 mg/L

Sensitivity: 2.95 mg/L

Storage: Store all components at 4°C in the dark.

Application: For the detection and quantification of Uric Acid (UA) concentration in serum and plasma.

Principle of the Assay

Abbexa's Uric Acid (UA) Assay Kit is a quick, convenient, and sensitive method for measuring and calculating Uric Acid (UA) concentration. Uric Acid is catalyzed by uricase to generate allantoin, carbon dioxide (CO₂), and hydrogen peroxide (H₂O₂). The resulting H₂O₂ reacts with the chromogenic substrate in the presence of a color developer to form a red quinone imide dye, with an absorbance maximum at 510 nm. The intensity of the color is proportional to the Uric Acid (UA) concentration, which can then be calculated.

Kit components

1. 96-well microplate
2. Chromogenic Reagent A: 24 ml
3. Chromogenic Reagent B: 6 ml
4. Standard (1g/L): 1 ml
5. Plate sealer: 2

Materials required but not provided

1. Microplate reader (510 nm)
2. Double-distilled water
3. Pipette and pipette tips
4. Centrifuge and centrifuge tubes
5. Vortex mixer
6. Incubator

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Protocol

A. Preparation of samples and reagents

1. Reagents

- **Assay Working Solution:** Prepare enough Assay Working Solution for the wells to be tested. Mix Chromogenic Reagent A with Chromogenic Reagent B at a ratio of 4 : 1. For example, prepare 250 µl of Assay Working Solution by mixing 200 µl of Chromogenic Reagent A with 50 µl of Chromogenic Reagent B. Assay Working Solution should be prepared immediately before use and used within the same day.
- **100 mg/L Standard:** Dilute the stock Standard (1 g/L) 10-fold with Assay Buffer. For example, add 100 µl of Standard (1 g/L) to 900 µl of double-distilled water. Mix thoroughly. This solution can be stored at 4°C for up to 7 days.
- **Standards:** Label 7 tubes with 10 mg/L, 20 mg/L, 30 mg/L, 40 mg/L, 60 mg/L, 80 mg/L, and 100 mg/L. Prepare these dilutions according to the volumes in the following table:

Standard Dilution (mg/L)	10	20	30	40	60	80	100
100 mg/L Standard (µl)	20	40	60	80	120	160	200
Double-distilled water (µl)	180	160	140	120	80	40	0

For the blank, use pure double-distilled water. The volume of each standard dilution will be 200 µl.

Note:

- Allow all reagents to equilibrate to room temperature before use.

2. Samples

Isolate the test samples soon after collecting and analyze immediately or aliquot and store at -20°C or -80°C for long-term storage. Avoid multiple freeze-thaw cycles.

The following sample preparation methods are intended as a guide and may be adjusted as required depending on the specific samples used.

- **Serum and plasma:** Serum and plasma samples can be tested directly.

It is recommended to carry out a preliminary experiment to determine the optimal dilution factor of samples before carrying out the formal experiment. Dilute samples into different concentrations with double-distilled water, then carry out the assay procedure. The recommended dilution factor for different samples is as follows (for reference only):

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Sample Type	Dilution Factor
Human serum	1
Human plasma	1
Rabbit serum	1
Bovine serum	1
Rat plasma	1
Mouse plasma	1

Note:

- Fresh samples or recently obtained samples are recommended to prevent degradation and denaturalization that may lead to erroneous results.

B. Assay Procedure

- Mark positions on the 96-well microplate for each standard, blank, and sample. *Each sample requires a corresponding control. It is strongly recommended to test all wells in duplicate.*
- Add 5 µl of each standard dilution to the standard wells.
- Add 5 µl of double-distilled water to the blank wells.
- Add 5 µl of sample to the sample wells.
- Add 250 µl of Assay Working Solution to all wells.
- Mix the well contents by tapping the plate, or shaking with a microplate shaker, for approximately 5 seconds.
- Cover with a plate sealer, then incubate at 37°C for 10 minutes.
- Unseal the plate, then measure the OD of each well with a microplate reader at 510 nm.

C. Calculation of Results

Plot the standard curve, using the mean OD of the standard dilutions (adjusted for the blank) on the y-axis, and their respective concentrations on the x-axis. The curve should form a straight line, described by the formula $y = ax + b$.

Based on this curve, the concentration of Uric Acid (UA) in each sample well can be derived using the following formulae:

1. Serum and plasma samples:

$$\text{Uric Acid (mg/L)} = \frac{(\text{OD}_{\text{Sample}} - \text{OD}_{\text{Blank}} - b)}{a} \times F$$

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where:

OD_{Sample}	OD value of the sample.
OD_{Blank}	OD value of the blank
a	Gradient of the standard curve ($y = ax + b$)
b	Y-intercept of the standard curve ($y = ax + b$)
F	Dilution factor of the sample

Technical Support

For troubleshooting and technical assistance, please contact us at support@abbexa.com.

For Reference Only