# Uric Acid Microplate Assay Kit

**Instructions for Use | Cat. No.: ARS6665**

Detection and Quantification of Uric Acid( UA)Content in Urine, Serum, Plasma, Other biological fluidsSamples.

**For research use only . Not for diagnostic or therapeutic procedures .**

## 1. Introduction

In humans and primates, uric acid is the end product of purine metabolism. It is produced by the oxidation of xanthine and hypoxanthine by xanthine oxidase and excreted in urine. High serum levels of uric acid, hyperuricemia, are associated with insulin resistance, cardiovascular disease, and gout. The mechanisms leading to hyperuricemia are typically either increased uric acid production or decreased urine excretion. Increased serum uric acid may be a marker of renal disease.

Uric Acid Colorimetric Microplate Assay Kit provides a simple and direct procedure for measuring uric acid content in a variety of samples.The assay is initiated with the enzymatic catalysis of the uric acid by uricase. The enzyme catalysed reaction products quinone can be measured at a colorimetric readout at 550 nm.

## 2. Kit Components

| Component | Volume | Storage |
|---|---|---|
| 96-Well Microplate | 1 plate | - |
| Diluent | 20mlx 1 | 4 °C |
| Enzyme | Powderx 1 | -20 °C, keep in dark |
| Dye Reagent | Powder x 1 | -20 °C, keep in dark |
| Standard (4 mmol/L) | 1 mL x 1 | 4 °C |
| Plate Adhesive Strips | 3 strips | - |
| Technical Manual | 1 Manual | - |

## 3. Materials Required But Not Provided

1. Microplate reader to read absorbance at 550 nm
2. Distilled water
3. Pipettor , multi-channel pipettor
4. Pipette tips
5. Mortar
6. Centrifuge
7. Timer
8. Ice

## 4. Reagent Preparation

**Standard:** Briefly centrifuge prior to opening. Perform 2 -fold serial dilutions of the top standardsolution using distilled water to make the standard curve.The concentration of standard curve could be 4/2/1/0.5/0.25/0.125/0.0625 mmol/L.

**Enzyme:** Add 10 mL Diluentto dissolve before use.Keep in dark and store at 4°C for 1 week after reconstitution.

**Dye Reagent:** Add 10 mL Diluentto dissolve before use.Keep in dark and store at 4°C for 2-3 weeks or -20°C for 2 months.

## 5. Sample Preparation

**1. Forurine , serum or plasma samples** Detect directly.

## 6. Assay Procedure

Warm all reagents to room temperature before use . Add following reagents in to the microplate :

| Reagent* | Standard | Blank | Sample* |
|---|---|---|---|
| Enzyme | 100 µl | 100 µl | 100 µl |
| Standard | 10 µl | -- | -- |
| Distilled water | -- | 10 µl | -- |
| Sample | -- | -- | 10 µl |
| Dye Reagent | 100 µl | 100 µl | 100 µl |
| Mix, put it in the oven,37 °C for 15 minutes, measured at 550 nm and recordthe absorbance. | | | |

**Note:**

*Reagents must be added sequentially and should not be premixed prior to addition.

**The concentrations can vary over a wide range depending on the different samples. For unknown samples, we recommend doing a pilot experiment & testing several doses to ensure the readings are within the standard curve range.

## 7. Calculation

**1. Calculate the sample concentration in ASSAY PROCEDURE according to the slope of**

the standard curve

$$C = \frac{(OD_{Sample} - OD_{Blank}) - Intercept}{Slope} \times \frac{V_{Standard}}{V_{Sample}} \times n \ (mmol/L)$$

Calculate the initial concentration according to sample preparation procedure .

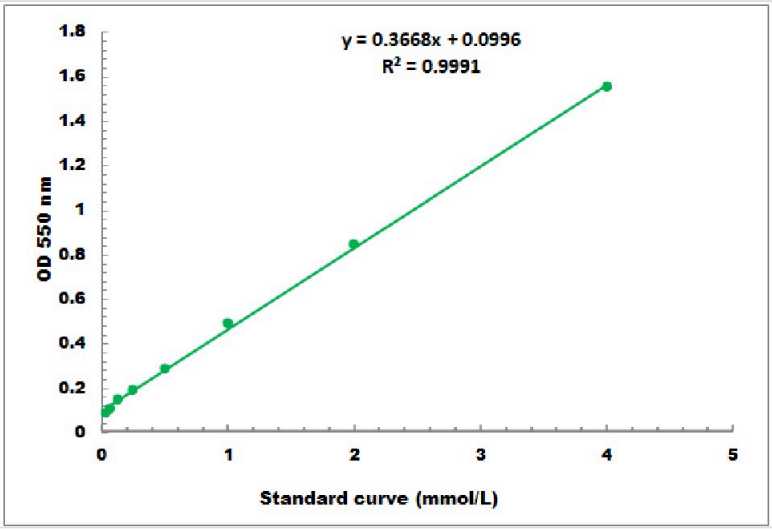
**2. According to one point of the standard OD and concentration**

$$C = \frac{(C_{Standard} \times V_{Standard}) \times (OD_{Sample} - OD_{Blank})}{(OD_{Standard} - OD_{Blank}) \times V_{Sample}} \ (\mu mol/ml)$$

| Item | Definition |
|---|---|
| **Slope** | the absorbance slope of standard curve |
| **n** | the dilution factor |
| **$C_{Standard}$** | the standard concentration, mmol/L = μmol/ml |
| **$V_{Standard}$** | the volume of standard in assay procedure, μl |
| **$V_{Sample}$** | the volume of sample in assay procedure, μl |

## 8. Typical Data

The standard curve is for demonstration only. A standard curve must be run with each assay.




Detection Range : 0.04 mmol/L - 4 mmol/L