



Urea Microplate Assay Kit

User Manual

Catalog # ASK1160

Detection and Quantification of Urea Content in Urine, Serum,
Tissue extracts, Cell lysate, Cell culture, Other biological fluids
media Samples.

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

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I. INTRODUCTION.....2

II. KIT COMPONENTS.....3

III. MATERIALS REQUIRED BUT NOT PROVIDED.....3

IV. SAMPLE PREPARATION.....4

V. ASSAY PROCEDURE.....5

VI. CALCULATION.....6

VII. TYPICAL DATA.....7



I. INTRODUCTION

Urea is primarily produced in the liver and secreted by the kidneys. Urea is the major end product of protein catabolism in animals. It is the primary vehicle for removal of toxic ammonia from the body. Urea determination is very useful for the medical clinician to assess kidney function of patients. In general, increased urea levels are associated with nephritis, renal ischemia, urinary tract obstruction, and certain extrarenal diseases, e.g., congestive heart failure, liver diseases and diabetes.

Decreased levels indicate acute hepatic insufficiency or may result from overvigorous parenteral fluid therapy.

Urea Microplate Assay Kit is designed to measure urea directly in biological samples without any pretreatment. The intensity of the color, measured at 620nm, is directly proportional to the urea concentration in the sample.



II. KIT COMPONENTS

Component	Volume	Storage
96-Well Microplate	1 plate	
Enzyme	Powder x 1	-20 °C
Enzyme Diluent	1.2 ml x 1	4 °C
Dye Reagent I	Powder x 1	4 °C
Dye Reagent II	3 ml x 1	4 °C
Standard (50 mg/L)	1 ml x 1	4 °C
Plate Adhesive Strips	3 Strips	
Technical Manual	1 Manual	

Note:

Enzyme: add 1 ml Enzyme Diluent into Enzyme tube to dissolve before use.

Dye Reagent I: add 6 ml distilled water to dissolve before use.

III. MATERIALS REQUIRED BUT NOT PROVIDED

1. Microplate reader to read absorbance at 620 nm
2. Distilled water
3. Pipettor
4. Pipette tips
5. Mortar
6. Centrifuge
7. Timer



IV. SAMPLE PREPARATION

1. For urine, serum or other biological fluids samples

Detect directly.

**V. ASSAY PROCEDURE**

Add following reagents in the microplate:

Reagent	Sample	Standard	Blank
Sample	100 μ l	--	--
Standard	--	100 μ l	--
Distilled water	--	--	100 μ l
Enzyme	10 μ l	10 μ l	10 μ l
Shake and mix, put it into the oven, 37 °C for 10 minutes.			
Dye Reagent I	60 μ l	60 μ l	60 μ l
Dye Reagent II	30 μ l	30 μ l	30 μ l
Shake and mix, put it into the oven, 37 °C for 15 minutes. Then record absorbance measured at 620 nm.			



VI. CALCULATION

1. According to the volume of sample

$$\begin{aligned} \text{Urea (mg/L)} &= C_{\text{Standard}} \times V_{\text{Standard}} \times (\text{OD}_{\text{Sample}} - \text{OD}_{\text{Control}}) / (\text{OD}_{\text{Standard}} - \text{OD}_{\text{Blank}}) / V_{\text{Sample}} \\ &= 50 \times (\text{OD}_{\text{Sample}} - \text{OD}_{\text{Control}}) / (\text{OD}_{\text{Standard}} - \text{OD}_{\text{Blank}}) \end{aligned}$$

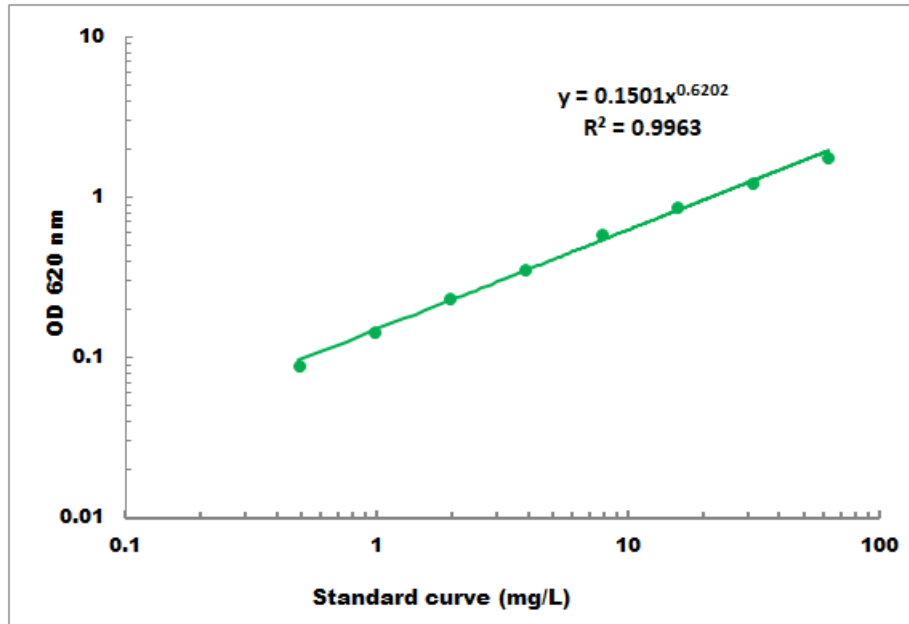
C_{Standard} : the standard concentration, 50 mg/L;

V_{Standard} : the volume of standard, 0.1 ml;

V_{Sample} : the volume of sample, 0.1 ml.

VII. TYPICAL DATA

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



Detection Range: 0.5 mg/L - 50 mg/L