(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS!)

Urea (BUN) Assay Kit (Urease Method)

Catalog No.: BC00051

Size: 100T

Please read the instructions carefully before use. If you have any questions or need further help during experiment, please don't hesitate to contact us through the following methods:

✉ Email (Sale) order@enkilife.com

✉ Email (Techsupport) techsupport@enkilife.com

☎Tel: 0086-27-87002838

Website: www.enkilife.com

Shelf life: Please refer to the label on the outer package.

Techsupport: In order to provide you with better service, please inform us the lot number on the label of the outer package.

Basic Information

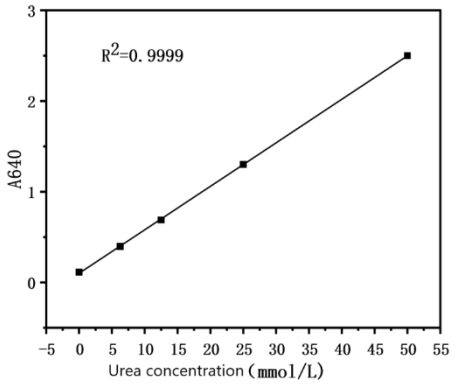
Product Name	Urea (BUN) Assay Kit (Urease Method)
Detection Method	Colorimetric
Sample Type	Serum, plasma, saliva, urine and other samples
Assay Type	Quantitative
Detection Instrument	Microplate reader (640 nm)
Range	6.25-50mM
Sensitivity	2.174mM

Product Introduction

Urea is the main end product of protein metabolism in the human body, and it constitutes the vast majority of non-protein nitrogen in the blood. Urea nitrogen in the blood originates from the liver and is excreted from the body through the kidneys with urine. Kidney failure, nephritis, urinary tract obstruction, etc. can increase the blood urea nitrogen content.

Principle

Urea is hydrolyzed by urease to produce ammonia ions and carbon dioxide. The ammonia ions react with the phenol color developer in an alkaline medium to produce a blue-green substance. The figure below shows the standard curve of urea determination by this kit.



Components

No.	Components	Size (100T)	Storage

Reagent 1	100mmol/L Urea Standard	200 μL	-20℃, store at 2-8°C after opening.
Reagent 2	Enzyme Diluent	7.5 mL	-20℃, store at 2-8°C after opening.
Reagent 3	Enzyme Solution	25 μL	-20℃, protect from light, store at 2-8°C after opening.
Reagent 4	Phenol Color Developer	15 mL	-20℃, protect from light, store at 2-8°C after opening.
Reagent 5	Alkaline Sodium Hypochlorite Solution	15 mL	-20℃, protect from light, store at 2-8°C after opening.
Consumable 1	Microplate	1 plate	RT
Consumable 2	Plate Sealer	2 pieces	RT

Storage

The unopened kit can be stored at -20℃ for 12 months, and after opening, it can be stored at 2-8℃ for 6 months.

Preparation

- **Sample handling**

Before the formal test, 2-3 samples with large expected differences should be selected and diluted to different concentrations for preliminary experiments, so that the concentration of the diluted samples is within the linear range. Serum, plasma, and saliva can be undiluted, and urine can be diluted 50 times. The above dilution ratios are for reference only. The diluent is physiological saline (0.9% NaCl) or PBS (0.01 M, pH 7.4).

- **Preparation of the kit**

1. Before testing, equilibrate the reagents in the kit to room temperature.
2. Prepare the enzyme working solution by mixing Reagent 2 and Reagent 3 in a volume ratio of 300:1, prepare fresh and use immediately.

3. Prepare different concentrations of standards by diluting the standard solution with water using the halving dilution method, to concentrations such as 50, 25, 12.5, 6.25, 3.125, 0 (blank well) mmol/L.

Operation process

1. Standard wells: Take 4 μL of different concentrations of standard solution and add to the corresponding standard wells. Sample wells: Take 4 μL of sample and add to the corresponding sample wells. Control wells: Add 4 μL of sample to the corresponding control wells.
2. Add 50 μL of enzyme working solution to the standard and sample wells from step (1). Add 50 μL of Reagent 2 to the control wells.
3. Stand at 37°C for 10 minutes.
4. Add 125 μL of Reagent 4 and Reagent 5 to the standard, sample, and control wells from step (1), respectively.
5. Stand at 37°C for 10 minutes, and measure the OD values at 640 nm in each well.

	Standard well	**Measurement well**	**Control well**
Different concentrations of standard solutions (μL)	4	--	--
Sample to be tested (μL)	--	4	4
Enzyme working solution (μL)	50	50	--
Reagent 2 (μL)	--	--	50
Stand at 37°C for 10 minutes.			
Reagent 4 (μL)	125	125	125
Reagent 5 (μL)	125	125	125
Stand at 37°C for 10 minutes, and measure OD values at 640 nm.			

Calculation

Standard fitting curve: $y = ax + b$

Urea content calculation formula: Urea content (mmol/L) = $(\Delta A_{640} - b) \div a \times f$

Annotation:

y: Standard OD value - blank OD value (OD value when the standard concentration is 0)

x: concentration corresponding to the absorbance

ΔA_{640}: Sample OD value - control OD value

f: dilution factor of the sample before adding it to the reaction system

Notes

1. When the color is too dark, dilute the sample appropriately and multiply the result by the dilution factor.
2. The enzyme working solution should be prepared immediately before use and should not be kept for a long time.
3. The enzyme stock solution has high viscosity and should be pipetted slowly.
4. After adding the enzyme working solution, the sample should be placed in a water bath for 10 minutes. If the sample volume is large, the operation should be performed in batches; the number of operations in the same batch should be controlled within 15.
5. The normal range of urea concentration in human serum is: 2.9–8.2 mmol/L.
6. This product is intended for scientific research use only by professionals and must not be used for clinical diagnosis or treatment, in food or drugs, or stored in ordinary residences.