Urea-Nitrogen (Urea) Assay Kit

Note: Take two or three different samples for prediction before test.

Operation Equipment: Spectrophotometer

Catalog Number: BC1530

Size:50T/24S

Components:

Reagent I: Powder×2. Storage at 2-8℃. Add 5 mL of distilled water to per bottle before use, fully dissolved. Prepared when the solution will be used.

Reagent II: 15 mL×1. Storage at 2-8℃.

Reagent III A: 1mL×1.Storage at 2-8℃.

Reagent III B:4 mL×1. Storage at 2-8℃.

Reagent III: Pour liquid A into liquid B and mix before use, or mix according to the ratio (A:B=1:4).

Reagent IV: 4 mL×1. Storage at 2-8℃.

Standard: Powder×1. Storage at 2-8℃. 10 mg urea. Dissolve with 4.66 mL distilled water to form 1 mg/mL urea-nitrogen standard solution (equiv. 2.146mg/mL urea).

Product Description

Urea (BUN) is the main product of human protein metabolism. Urea constitutes the majority of non-protein nitrogen in blood. Blood urea nitrogen is one of the main indexes of renal function. This kit use indophenol blue colorimetric method to test NH_3-N product by urease hydrolysis. The concentration of indophenol is proportional to urea nitrogen concentration.

Reagents and Equipment Required but Not Provided:

Spectrophotometer, balance, cryogenic centrifuge, 1 mL glass cuvette, mortar/homogenizer, constant temperature water bath pot.

Procedure:

I. Sample preparation:

1. Tissue sample

According to the ratio of mass (g): volume of distilled water (1:5-10 (it is recommended to weigh about 0.1g, add 1mL of distilled water), homogenize on ice and then centrifuged at 4℃ 12000g for 15min, take the supernatant to be measured.

2. Cells

According to the number of cells (10^4): the volume of distilled water (mL) for the ratio of 500-1000:1 (recommended 5 million cells to add 1mL); then ice bath ultrasonic broken cells (power 300w, ultrasound 3s, interval 7s, total time 3min); then 4 ℃ 12000g centrifugation 15min take the supernatant on ice to be measured.

3. Serum (plasma) sample:

Detect sample directly.

II. Determination procedure:

1. Preheat the spectrophotometer 30 min, adjust the wavelength to 630 nm and set zero with distilled water.
2. Standard solution: dilute urea-nitrogen standard solution (1 mg/mL) with distilled water to 25 μg/mL (equiv. 53.65μg/mL urea).
3. Add reagents with the following list:

Reagent Name (μL)	Blank Tube (Ab)	Standard Tube (As)	Test Tube (At)	Control Tube (Ac)
Sample	-	-	60	60
Standard Solution	-	60	-	-
Distilled water	60	-	-	120
Reagent I	120	120	120	-
Reagent II	220	220	220	220
Mix well, place at 37℃ for 10 min.				
Reagent III	80	80	80	80
Reagent IV	60	60	60	60
Mix well, place at room temperature for 30 min.				
Distilled water	460	460	460	460
Mix well, detect absorbance at 630 nm. ΔAs=As-Ab, ΔAt=At-Ac. Standard tube and Blank tube only need to do 1-2 times				

III. Calculation:

1. Calculated by sample weight
   Urea Nitrogen content (μg/g)= ΔAt÷ΔAs×Cs×Ve÷W=25×ΔAt÷ΔAs÷W
   Urea concentration (μg/g)= ΔAt÷ΔAs×Cs'×Ve÷W=53.65×ΔAt÷ΔAs÷W
2. Calculated by protein concentration
   Urea Nitrogen content (μg/mg prot)=ΔAt÷ΔAs×Cs×Ve÷(Cpr×Ve)=25×ΔAt÷ΔAs÷Cpr
   Urea concentration (μg/mg prot)= ΔAt÷ΔAs×Cs'×Ve÷(Cpr×Ve)= 53.65×ΔAt÷ΔAs÷Cpr
3. Calculated by cell amount=
   Urea Nitrogen content (μg/10^4 cell)= ΔAt÷ΔAs×Cs×Ve÷n=25×ΔAt÷ΔAs÷n
   Urea concentration (μg/10^4 cell)= ΔAt÷ΔAs×Cs'×Ve÷n=53.65×ΔAt÷ΔAs÷n
4. Calculated by liquid volume
   Urea Nitrogen content (μg/mL)=ΔAt÷ΔAs×Cs=25×ΔAt÷ΔAs
   Urea concentration (μg/mL)= ΔAt÷ΔAs×Cs'=53.65×ΔAt÷ΔAs

Cs: concentration of urea nitrogen standard solution, 25 μg/mL;
Cs': concentration of urea standard solution, 53.65 μg/mL
Ve: extraction volume, 1 mL;

W: sample weight, g;
Cpr: sample protein concentration, mg/mL;
n: cell amount. 10^4.

Note:

1. Reagent I working solution can be stored at 2-8℃ for one week.
2. If measured value of ΔA or At exceed 1, it is suggested dilute sample with distilled water for determination.

Technical Specifications：

Minimum Detection Limit：

0.000086 μg/mL（urea nitrogen concentration）or 0.000185 μg/mL（urea concentration）

Linear Range：

0.390625-50 μg/mL（urea nitrogen concentration）or 0.838-107.3 μg/mL （urea concentration）

Recent Product citations：

[1] Xiaoguang Zhu,Jun Shi,Huicong li,et al. PVT1 knockdown alleviates vancomycin-induced acute kidney injury by targeting miR-124 via inactivation of NF-κB signaling. RSC advances. September 2018;(IF3.049)

Related products：

BC0080/BC0085 Nitrate Reductase(NR) Activity Assay Kit

BC1450/BC1455 Glutaminase (GLS) Assay Kit