Tribioscience

Tribo™ Urea (BUN) Colorimetric Assay (Catalog# TBS2201, 200 Assays, Store at 4°C)

DESCRIPTION

Urea (carbamide, carbonyl diamide) is a predominant final metabolite of nitrogenous compounds in mammals, accounting for 80-90% of nitrogen excretion in human and animals. Formed exclusively in the liver, urea is mostly transported by the bloodstream to the kidneys, where it is excreted into the urine. It is an indicator of liver and kidney functions. Determination of urea concentration is an essential task in clinical laboratories and research.

The Tribo™ Urea Colorimetric Assay provides a rapid, sensitive method to measure urea concentration in biological samples. The intensity of the color is directly proportional to the urea concentration in the sample.

Synonyms: Carbamide, Carbonyl diamide, Carbonyldiamine, Diaminomethanal, Diaminomethanone, BUN.

APPLICATION

Direct Assay: urea in serum, plasma, urine, milk, cell/tissue culture, animal products and wine.
Drug Discovery and screen: effects of drugs on urea metabolism.

KIT CONTENTS FOR 200 ASSAYS

Component	Unit Size
Urea Standard 500 mg/dL)	200 µL
Urea reagent Component A	22 mL
Urea reagent Component B	22 mL
Component C: Assay Buffer	30 mL

Storage conditions: Store the kit at 4°C. For long-term storage, keep the standard at -20 °C. Shelf-life: 12 months.

FEATURES

- Sensitive and accurate: The Kit detects as low as 0.1 mg/dL in solution.
- Resistant to ammonia interference, a common problem in urease-based assays.
- Works reliable in complex matrices such as phenol red-containing media (e.g., cell culture media).
- Simple and high-throughput: The procedure is easily adapted to automation with no separation required as a high-throughput assay.

PROCEDURES

Note: Bring all reagents to room temperature (18-25˚C) before using it. If reagent A appeared cloudy, warm it in water bath at 37°C until it clears.

1. **Sample Preparation:** Serum and plasma samples can be directly analyzed (dilute n=1). Urine samples should be diluted in distilled water prior to assay (dilute n=50). For tissue /cell samples, homogenize samples in 120 µL assay supernatant for assay.
2. **AB Mixture:** Mix equal volumes of Reagent A and B shortly prior to assay.

Standard Preparation:

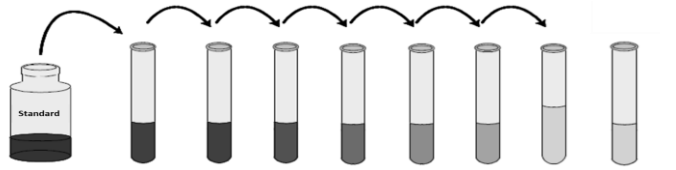
1. Label test tubes as #1 through #8. Pipet 270 µL assay buffer to #1, and 150 µL assay buffer from #2 to #8.

2. Pipet 30 µL of 500mg/dL standard into tube #1, then make 2x serial dilutions of the standard using the Tube#1(50 mg/dL standard solution) from Tube #2 through #7 with sequential transfer of 150 µL to the next concentration. Mix each tube thoroughly before the next transfer. as shown in **Fig1.** diagram**.**

The standard concentration in tube 1 through 7 of Urea standard concentrations: 50, 25, 12.5, 6.25, 3.13, 1.56 and 0.78mg/dL. Tube# 8 is Standard 8 (0 mg/dL) as **Blank**.

Fig.1 diagram

Addition:30 µL 150 µL 150 µL 150 µL 150 µL 150 µL 150 µL



	Std1	Std2	Std3	Std4	Std5	Std6	Std7	Std8
Assay Buffer (µL)	270	150	150	150	150	150	150	150
Addition	Stock	Std1	Std2	Std3	Std4	Std5	Std6	
Addition Vol. (µL)	30	150	150	150	150	150	150	0
Conc (mg/dL)	50	25	12.5	6.25	3.13	1.56	0.78	0

3. Add 50 µL of urea standards, and samples and blank with duplicate into separate wells.

4. Add 200 µL urea reagent A and B mix to each well. Mix well gently and carefully, do not create foaming in the well.

5. Incubate 1-2 hours at room temperature with gently shaking.

6. Read the plate at 430 nm and record data.

CACULATION OF DATA

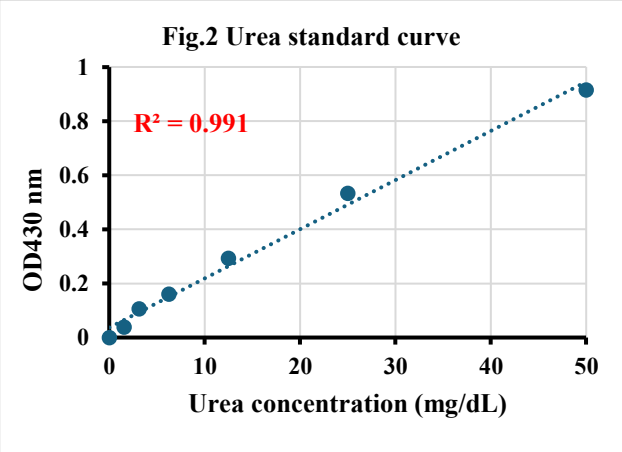
Urea concentration (mg/dL) of the sample is calculated as the formula below**.**

The OD values of samples and standards should subtract the OD values of **Blank, after substrate blank then get ΔOD of samples and standards**. **ΔOD** of standards is used to generate the standard curve. Determine the slope and calculate the urea concentration of the samples using the equation obtained from the linear regression of the standard curve. The equation is **Y=aX+b.**

X is the concentration of urea (mg/dL), **Y** is the ΔOD value, **a** is slope, and **b** is a Y-intercept.
Calculate sample urea concentration (x) using the equation:
X (mg/dL) = N* (Y-b)/a.

buffer, then centrifuge at 10,000rpm for 5min, and collect the **N** is dilution Factor.
Conversions: 1mg/dL =167 μM =0.001% =10 ppm
BUN (Blood Urea Nitrogen) (mg/dL) = concentration of sample urea (mg/dL) /2.14

Typical Standard Curve
The typical standard curve shown in Fig. 2. is only for reference. It cannot be used for urea analysis.



RELATED PRODUCTS:

Resazurin Cell Viability Kit (TBS2001)
Cytotoxicity Colorimetric Assay (TBS2002)
CCK-8 Cell Viability Assay (TBS2022)
ATP Colorimetric/Fluorometric Assay Kit (TBS2010)
ADP/ATP Ratio Assay Kit (Bioluminescent (BS2015)
ADP Colorimetric/Fluorometric Assay Kit (TBS2020)
NAD/NADH Colorimetric Assay (TBS2029)
Caspase-3 Colorimetric Assay kit (TBS2030)
Mitochondria Isolation (TBS2116)
Mitochondria Complex 1 Activity Assay (TBS2017)
Mitochondria Oxidase Activity (TBS2105)
Mitochondrial Membrane Potential Assay (TBS2049)
Cytochrome C Oxidase Activity Assay (TBS2115)