# Blood Ammonia Content Assay Kit

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

**Operation Equipment:** Microplate Reader or Spectrophotometer

**Catalog Number:** BC4385

**Size:**100T/96S

**Components:**

Extract solution I: Liquid 20 mL×1.

Extract solution II: Liquid 20 mL×1.

Reagent I A: Liquid 4 mL×1. Storage at 4℃.

Reagent I B: Liquid 16 mL×1. Storage at 4℃. Reagent I working solution: make the solution as the volume ratio of Reagent I A: Reagent I B= 1:4, prepare the reagent when it will be use.

Reagent II: Liquid 15 mL×1. Storage at 4℃.

Standard: Liquid 1 mL×1, 100 μmol/mL ammonia standard solution. Storage at 4℃.

**Product Description**

Endogenous and exogenous ammonia are the main sources of blood ammonia. Ammonia maintains steady state in the blood, which means the source and consume of blood ammonia maintain dynamic balance. Ammonia is a poisonous and harmful substance and the metabolic detoxification mainly in the liver. Ammonia cannot be detoxified when liver function is severely impaired. Accumulation of ammonia in the central nervous system can lead to hepatic encephalopathy.

In this kit, the method is based on the principle of indophenol blue reaction of ammonia. First, the protein in the serum (plasma) is precipitated by a protein precipitating agent, and then the blood ammonia is measured by the direct colorimetric method of phenol-hypochlorite. The absorbance ratio of blue indophenol is in direct proportion to the contents of ammonia and has a special absorption peak at 630 nm.

**Reagents and Equipment Required but Not Provided:**

Microplate reader or spectrophotometer, desk centrifuge, transferpettor, water-bath/constant temperature incubator,， micro glass cuvette/96 well flat-bottom plate, EP tubes, and distilled water.

**Procedure:**

**I. Applicable range:**

This kit can be used to measure the content of blood ammonia in serum (plasma) of various animal and other samples.

**II. Determination procedure:**

1. Preheat the spectrophotometer/ microplate reader 30 minutes, adjust the wavelength to 630 nm and set zero with distilled water.
2. Standard solution: dilute the 100 μmol/mL ammonia standard solution with distilled water to 6、4、2、1、0.5、0.25、0.125 μmol/mL.

3. Add reagents with the following list:

| Reagent Name (μL) | Blank Tube (B) | Standard Tube (S) | Test Tube (T) |
|---|---|---|---|
| Serum(plasma) | - | - | 50 |
| Standard Solution | - | 50 | |
| Distilled water | 50 | - | - |
| Extract solution I | 125 | 125 | 125 |
| Extract solution II | 125 | 125 | 125 |
| Mix well, centrifuge at 3500 rpm for 10 minutes, take the supernatant for test. | | | |
| Supernatant | 100 | 100 | 100 |
| Reagent I | 100 | 100 | 100 |
| Reagent II | 100 | 100 | 100 |
| Mix well, place at 37℃ for 20 minutes. | | | |

Mix well, take 200 μL of reaction solution to the micro glass cuvette/96 well plate and measure the absorbance at 630 nm, which noted as $A_B$, $A_S$, $A_T$. $\Delta A=A_T-A_B$, $\Delta A_S=A_S-A_B$.

**III. Calculation:**

1. Standard curve

The concentration of standard solution as x-axis, $\Delta A_B$ as y-axis, obtain the equation y=kx+b. Take ΔA to the equation to acquire x value.

2. Calculation

Blood ammonia content (μmol/mL) = x×Vs÷Vs= x

Vs: Sample volume (mL), 0.05 mL.

**Note:**

1. Blank tube needs only to be tested once or twice.
2. Use as soon as possible after Reagent I is configured. Cannot be used if discoloration is found.
3. All equipment and blood collection devices should be free of ammonia. Measured immediately after blood collection, if cannot be measured immediately can be kept at 2-8℃ and for 2 hours. All sample should not be hemolyzed.

**Related products**：

BC2770/BC2775 Blood Potassium Content Assay Kit
BC2790/BC2795 Blood Magnesium Content Assay Kit
BC1650/BC1655 Blood Phosphate Content Assay Kit
BC2800/BC2805 Blood Sodion Content Assay Kit
BC1730/BC1735 Serum Ferri Ion Content Assay Kit

**Technical Specifications**：

Minimum Detection Limit：0.0251 μmol/mL
Linear Range：0.0625-8 μmol/mL