

Blood Ammonia Content Assay Kit

Note: The reagents have been changed, please be aware of and follow this instruction strictly.

Operation Equipment: Spectrophotometer

Catalog Number: BC4380

Size: 50T/48S

Components:

Extract solution: Liquid 45 mL×1. Storage at 2-8°C

Reagent I A: Liquid 5 mL×1. Storage at 2-8°C.

Reagent I B: Liquid 20 mL×1. Storage at 2-8°C.

Reagent I: make the solution as the volume ratio of Reagent I A: Reagent I B= 1:4, prepare the reagent when it will be used.

Reagent II: Liquid 25 mL×1. Storage at 2-8°C.

Standard: Liquid 1 mL×1, 100 μmol/mL ammonia standard solution. Storage at 2-8°C.

2 μmol/mL ammonia standard solution: Before use, 20 μL of 100 μmol/mL nitrogen standard solution and 980 μL of distilled water are mixed to form 2 μmol/mL nitrogen standard solution, which can be used as standard solution for standard tubes.

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:

Spectrophotometer, Desk Centrifuge, 1 mL Glass Cuvette, Transferpettor, Water-bath/Constant Temperature incubator, 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. Note that the sample is not hemolytic.

II. Determination procedure:

1. Preheat the spectrophotometer 30 minutes, adjust the wavelength to 630 nm and set zero with

distilled water.

2. Add reagents with the following list:

Reagent Name (μL)	Blank Tube (B)	Standard Tube (S)	Test Tube (T)
Serum(plasma)	-	-	120
Standard Solution	-	120	
Distilled water	120	-	-
Extract solution	600	600	600
Mix well, centrifuge at 8000 rpm for 10 minutes, take the supernatant for test.			
Supernatant	400	400	400
Reagent I	400	400	400
Reagent II	400	400	400
Mix well, place at 37°C for 20 minutes.			

Mix well, take 1 mL of reaction solution to the 1 mL glass cuvette and measure the absorbance at 630nm, noted as A_B , A_S , A_T . $\Delta A = A_T - A_B$, $\Delta A_S = A_S - A_B$. The standard tube and blank tube only need to be measured 1-2 times.

III. Calculation:

$$\text{Blood ammonia content } (\mu\text{mol/mL}) = C_s \times \Delta A \div \Delta A_s \times V_s \div V_t = 2 \times \Delta A \div \Delta A_s$$

V_s : Sample volume (mL), 0.12 mL. C_s : Standard solution concentration, 2μmol/mL。

Note:

- Use as soon as possible after Reagent I is configured. Cannot be used if discoloration is found.
- 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°C and for 2 hours. All sample should not be hemolyzed.
- If ΔA is less than 0.01, it is recommended to increase the volume while decreasing the volume of extract (e.g., 200 μL serum (plasma) + 520 μL extract); if ΔA is greater than 1.0, it is recommended to reduce the sample volume while increasing the volume of extract or diluting the supernatant for measurement, taking care to modify the calculation formula or multiply by the number of dilutions simultaneously.

Experimental examples:

1. Take horse serum and operate according to the measurement steps, use 1mL glass cuvette to measure and calculate $\Delta A = A_t - A_b = 0.311 - 0.005 = 0.306$, $\Delta A_s = A_s - A_b = 0.553 - 0.005 = 0.548$, and calculate the content to get:

$$\text{Blood ammonia content } (\mu\text{mol/mL}) = 2 \times \Delta A \div \Delta A_s = 1.117 \mu\text{mol/mL}$$

2. Fetal bovine serum was taken and operated in accordance with the measurement procedure, and 1mL glass cuvette was used to measure and calculate $\Delta A = A_t - A_b = 0.280 - 0.005 = 0.275$, $\Delta A_s = A_s - A_b = 0.553 - 0.005 = 0.548$, and the calculated content was:

Blood ammonia content ($\mu\text{mol/mL}$) = $2 \times \Delta A \div \Delta A_s = 1.004 \mu\text{mol/mL}$.

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 Sodium Content Assay Kit
- BC1730/BC1735 Serum Ferri Ion Content Assay Kit

Technical Specifications:

Minimum Detection Limit: $0.0258 \mu\text{mol/mL}$

Linear Range: $0.125\text{-}3 \mu\text{mol/mL}$