Tissue Inorganic Phosphorus Content Assay Kit

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

Detection instrument: Spectrophotometer/Microplate reader

Cat No: NA0421 **Size:** 100T/96S

Components:

Reagent I: 100 mL×1. Storage at 4°C. Reagent II: 5 mL×1. Storage at 4°C.

Reagent III: Powder×2. Storage at 4°C. Working solution: Fully dissolved with 5 mL of distilled water,

then add 2.5 mL of Regent II, mix well.

Standard: 1 mL×1, 1 mmol/L inorganic phosphorus standard. Storage at 4°C.

Product Description:

Inorganic phosphorus mainly refers to phosphate radical, which is involved in many kinds of metabolism, including energy metabolism, nucleic acid metabolism, protein phosphorylation and dephosphorylation, and so on. In addition, inorganic phosphorus promotes the synthesis, transformation and transport of carbohydrates.

Molybdenum blue can react with phosphate group, the reaction product can be detected by colorimetric assay at 660 nm and calculate the inorganic phosphorus content indirectly.

Required material

Spectrophotometer/microplate reader, centrifuge, water bath, transferpettor, micro glass cuvette/96 well flat-bottom plate, distilled water.

Procedure:

I. Sample Extraction:

Suggested 0.1g of sample with 1mL of Reagent I, fully grinding on ice, centrifuge at 10000 rpm and 4°C for 10 minutes, supernatant is used for test.

II. Determination procedure:

- 1 Preheat the spectrophotometer for 30 min, adjust wavelength to 660nm, set zero with distilled water.
- 2 Set the temperature of water bath to 40°C.
- 3 Add reagents with the following list:

Reagent name (µL)	Blank tube (A _B)	Standard tube (A _T)	Test tube (A _S)
Standard		10	
Supernatant			10
Distilled water	100	90	90
Reagent III	100	100	100

Mix well, 40°C water bath for 10 minutes, detect the absorbance at 660 nm after cooling at room temperature for 10 minutes. Record as A_B, A_S and A_T respectively.

Note: The colorimetry needs to be completed within 40 minutes.

III. Calculation:

Phosphate (mmol/g fresh weight)= $[C\times(A_T-A_B)\div(A_S-A_B)]\times V\div W=0.001\times(A_T-A_B)\div(A_S-A_B)\div W$

C: standard concentration, 1mmol/L;

V: supernatant volume, 1ml=0.001 L;

W: Sample weight, g

Note:

- 1. Reagent III should be prepared before use and only be used the same day.
- 2. Before the determination, 1-2 samples shall be used for pre-test. If the absorption value is greater than 1, distilled water shall be used for corresponding dilution.

Experimental example:

1. Take 0.1g kidney, add 1 mL of Reagent I, centrifugate the supernatant, and then operate according to the determination steps. Use 96 well plate to measure: $A_T = 0.696$, $A_B = 0.051$, $A_S = 0.282$.

Inorganic phosphorus content (mmol/g mass) = $0.001 \times (A_T - A_B) \div (A_S - A_B) \div W = 0.001 \times (0.696 - 0.051) \div (0.282 - 0.051) \div 0.1 = 0.028$ mmol/g mass.

2. 0.1g liver is added with 1 mL of Reagent I, and the supernatant is centrifuged. Then, the determination procedure is followed. The 96 well plate is used to determine. $A_T = 0.578$, $A_B = 0.051$, $A_S = 0.282$.

Inorganic phosphorus content (mmol/g mass) = $0.001 \times (A_T - A_B) \div (A_S - A_B) \div W = 0.001 \times (0.578 - 0.051) \div (0.282 - 0.051) \div 0.1 = 0.023$ mmol/g mass.

Related Products:

Bu J, Yu J, Wu Y, et al. Hyperlipidemia affects tight junctions and pump function in the corneal endothelium[J]. The American Journal of Pathology, 2020.

Related Products:

NA0661/NA0420 Serum Total Iron Binding Capacity(TIBC) Assay Kit

NA0662/NA0379 Total Phosphorus Content Assay Kit

NA0302/NA0301 Tissue Iron Content Assay Kit

NA0296/NA0295 Blood Ammonia Content Assay Kit

Technical Specifications:

The detection limit: 0.0302 mmol/L

Linear range: 0.625-8 mmol/L