Tissue Total Phosphorus Content Assay Kit

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

Detection instrument: Spectrophotometer

Catalog Number: BC2850

Size: 50T/48S

Components:

Reagent I: Liquid 12 mL×1, store at 2-8℃. It is strong corrosive and strong oxidizing. Tighten the cover immediately after using.

Reagent II: Liquid 11 mL×1, store at 2-8℃.

Reagent IIIA: Powder×1, store at 2-8℃. Add 10 mL of distilled water to fully dissolve. Unused reagent is still stored at 2-8℃ for four weeks.

Reagent IIIB: Powder×1, store at 2-8℃. Add 10 mL of distilled water to fully dissolve. Unused reagent is still stored at 2-8℃ for four weeks.

Reagent III: Reagent IIIA, Reagent IIIB and Reagent II are mixed by the ratio of 1:1:1 to make Reagent III before use. Prepared Reagent III is light yellow. It is colorless if the reagent is invalid. It is blue if the reagent is contaminated with phosphorus. Prepared Reagent III could only be used the same day.

Standard: Liquid 1 mL×1, 10 mmol/L inorganic phosphorus standard, store at 2-8℃.

Product Description:

The form of phosphorus includes inorganic phosphorus and organic phosphorus. Inorganic phosphorus mainly refers to phosphate radical, which is involved in many kinds of metabolism, including energy metabolism, nucleic acid metabolism, protein phosphorylation, dephosphorylation, and so on. By measuring the content of total phosphorus and inorganic phosphorus, the utilization rate of phosphorus in crops can be understood, and the basis for rational fertilization can be provided.

After digestion, total phosphorus was converted into inorganic phosphorus. The molybdenum blue method is a classical method for determining the content of inorganic phosphorus. Under certain conditions, molybdenum blue and phosphate form a substance with a characteristic absorption peak at 660nm. By measuring the light absorption of 660nm, the inorganic phosphorus content can be calculated, and then the total phosphorus content in the tissue can be calculated.

Required but not Provided Material and Equipment:

Centrifuge, spectrophotometer, water bath, 1mL glass cuvette, transferpettor, distilled water and **concentrated sulfuric acid** (99%).

Procedure:

I. Sample Extraction:

0.1g of sample with 1 mL of concentrated sulfuric acid (Wrap the sealing film to prevent the lid from bursting) put in boiling water bath for 10 minutes. When the solution is black or brown, take it out. Add

200μL reagent I after cooling, mix well. Continue boiling (Wrap the sealing film to prevent the lid from bursting) until the solution is transparent, then cool at room temperature, add 3.8 mL of distilled water and mix well. centrifugated at 10000 rpm and room temperature for 10 minutes, supernatant is used for test.

II. Determination procedure:

1 Preheat the spectrophotometer for 30min, adjust wavelength to 660 nm, set zero with distilled water.

2 Set the temperature of water bath to 40℃.

3 Preparation of 1 mmol/L standard solution: 100μL 10 mmol/L phosphorus standard solution and 900μL distilled water mixed to prepare 1 mmol/L standard solution.

4 Add reagents with the following list:

Reagent (μL)	Blank tube (A_B)	Standard tube (A_T)	Test tube (A_S)
Standard	-	50	-
Supernatant	-	-	50
Distilled water	500	450	450
Reagent III	500	500	500

Mix well, 40℃ 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. Standard tube and blank tube only need to be measured once or twice.

III. Calculation:

Total phosphorus content(mmol/g weight)=$[C\times(A_T- A_B)\div(A_S - A_B)]\times V\div W$

$=0.005\times(A_T - A_B)\div(A_S - A_B)\div W$

C: standard concentration, 1 mmol/L;

V: supernatant volume, 5 mL=0.005 L;

W: Sample weight, g.

Note:

1. When the determination of A is greater than 0.8, it is recommended to dilute supernatant with distilled water before performing the measurement and multiply the dilution factor in the calculation formula.

Experimental example:

1. Take 0.1g kidney according to the extraction procedure, centrifugally take it up and clean it, and then follow the measurement procedure. Calculate: A_T= 0.273, A_B= 0.016, A_S= 0.429. Calculate the total phosphorus content according to the sample mass.

Total phosphorus content (mmol/g weight) = $0.005 \times (A-A_B) \div (A_S-A_B) \div W$=0.031 mmol/g weight.

2. Take 0.1g spleen according to the extraction procedure, centrifugally take the cleaning, and

then follow the measurement procedure. Calculate: A_T= 0.268, A_B= 0.016, A_S= 0.429, calculate the total phosphorus content according to the sample mass

Total phosphorus content (mmol/g weight) =0.005×(A-A_B) ÷(A_S-A_B) ÷W=0.031 mmol/g weight.

Related Products：

BC2860/BC2865	Serum Total Iron Binding Capacity (TIBC) Assay Kit
BC2810/BC2815	Blood Zinc Content Assay Kit
BC2820/BC2825	Water Mercury Ion (Hg^{2+}) Content Assay Kit
BC2840/BC2845	Phosphate Content Assay Kit

Technical Specifications:

The detection limit: 0.0585 mmol/L

Linear range: 0.0625-2 mmol/L