

# Tissue Total Phosphorus Content Assay Kit

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

**Detection instrument:** Spectrophotometer /Microplate reader

**Cat No:** NA0379

**Size:** 100T/96S

## Components:

Reagent I: 20 mL×1. Storage at 4°C (strong corrosive, strong oxidizing).

Reagent II: 7 mL×1. Storage at 4°C.

Reagent III: Powder×2. Storage at 4°C and protected from light. 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:

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 material

Centrifuge, spectrophotometer/microplate reader, water bath, micro glass cuvette/96 well flat-bottom plate, transferpettor, distilled water, concentrated sulfuric acid.

## Procedure:

### I . Sample Extraction:

0.1g of sample with 1mL of concentrated sulfuric acid (cover tightly to prevent moisture loss) put in boiling water bath for 10 minutes. When the solution is black or brown, take it out. Add 200μL of Reagent I after cooling, mix well. Continue boiling (cover tightly) until the solution is transparent, then cool at room temperature, add 3.8mL of distilled water and mix well. centrifugated at 10000rpm and room temperature for 10 minutes, supernatant is used for test.

### II . Determination procedure:

1. Preheat the spectrophotometer/ microplate reader for 30min, 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 <sub>B</sub> )	Standard tube (A <sub>T</sub> )	Test tube (A <sub>S</sub> )
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<sub>B</sub>, A<sub>S</sub> and A<sub>T</sub> respectively.

#### Calculation:

$$\text{Total phosphate (mmol/g fresh weight)} = [C \times (A_T - A_B) \div (A_S - A_B)] \times V \div W \\ = 0.005 \times (A_T - A_B) \div (A_S - A_B) \div W$$

C: standard concentration, 1mmol/L;

V: supernatant volume, 5 mL=0.005 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 1.5, distilled water shall be used for corresponding dilution.

#### 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. Use 96 well plate to calculate: A<sub>T</sub>= 0.191, A<sub>B</sub>= 0.051, A<sub>S</sub>= 0.282. Calculate the total phosphorus content according to the sample mass

$$\text{Total phosphorus content (mmol/g mass)} = 0.005 \times (A - A_B) \div (A_S - A_B) \div W = 0.005 \times (0.191 - 0.051) \div (0.282 - 0.051) \div 0.1 = 0.030 \text{ mmol/g mass.}$$

2. Take 0.1g spleen according to the extraction procedure, centrifugally take the cleaning, and then follow the measurement procedure. Use 96 well plate to calculate: A<sub>T</sub>= 0.197, A<sub>B</sub>= 0.051, A<sub>S</sub>= 0.282, calculate the total phosphorus content according to the sample mass

$$\text{Total phosphorus content (mmol/g mass)} = 0.005 \times (A - A_B) \div (A_S - A_B) \div W = 0.005 \times (0.197 - 0.051) \div (0.282 - 0.051) \div 0.1 = 0.032 \text{ mmol/g mass.}$$

#### Related Products:

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

NA0666/NA0424 Blood Zinc Content Assay Kit

NA0665/NA0423 Water Mercury Ion(Hg<sup>2+</sup>) Content Assay Kit

NA0663/NA0421 Phosphate Content Assay Kit

#### Technical Specifications:

The detection limit: 0.0338 mmol/L

Linear range: 0.625-8 mmol/L