

Soil Neutral Phosphatase (S-NP) Activity Assay Kit - Micro Method**Product Code**

67050

Product Introduction

Soil phosphatase catalyzes the mineralization of organic phosphorus in soil. Its activity directly affects the decomposition, transformation, and bioavailability of organic phosphorus, and it is used as an indicator for evaluating the direction and intensity of phosphorus biotransformation.

Phosphatase activity is significantly affected by soil carbon and nitrogen content, available phosphorus content, and pH. According to the optimum pH range, soil phosphatases are generally divided into neutral, acidic, and alkaline types.

In a neutral environment, S-NP catalyzes the hydrolysis of disodium phenyl phosphate to produce phenol and disodium hydrogen phosphate. S-NP activity is calculated by measuring the amount of phenol produced.

Actual readings may vary under different testing conditions due to differences in testing instruments. Performance data are for reference only.

Package Contents

Size	Code	Component	Quantity
100T	67050.1	Reagent I	1 bottle
100T	67050.2	Reagent II	1 bottle
100T	67050.3	Reagent III	1 bottle
100T	67050.4	Reagent IV	2 tubes
100T	67050.5	Standard	1 bottle
100T	67050.m	Instruction Manual	1 copy

Quality Standards and Safety Information

Raw Material or Packaging Name	Quality Standard	Main Toxicity
Reagent I	--	--
Reagent II	--	--
Reagent III	--	--
Reagent IV	--	--
Standard	--	--

Transportation and Storage

Transportation	Transport with ice packs.
Storage	Store at 2-8°C, protected from light. Shelf life: 180 days.

Instructions for Use**1. Reagent Preparation**

1. Before use, add 100 mL distilled water to Reagent II and dissolve thoroughly.
2. Immediately before use, add 576 μL absolute ethanol (self-prepared) and 24 μL distilled water to each vial of Reagent IV, then dissolve thoroughly. Do not use Reagent IV after it turns brown.

2. Catalytic Reaction

1. Weigh approximately 0.1 g of air-dried, well-mixed soil.
2. Add 0.05 mL toluene (self-prepared) and gently shake for 15 min.
3. Add 0.4 mL Reagent I and shake well.
4. Place the mixture in a 37°C constant-temperature incubator and start timing. Allow the catalytic reaction to proceed for 24 h.
5. When the reaction time is complete, quickly add 1 mL Reagent II and mix thoroughly to terminate the enzyme-catalyzed reaction.
6. Centrifuge at 10000 rpm at room temperature for 10 min.
7. Collect the supernatant and place it on ice for testing.

3. Color Development Reaction

1. Preheat the spectrophotometer or microplate reader for more than 30 min. Set the wavelength to 660 nm and zero with distilled water.
2. Blank tube: In a micro glass cuvette or microplate, add 10 μL distilled water, 20 μL Reagent III, and 4 μL Reagent IV. Mix thoroughly. After color development, add 166 μL distilled water, mix thoroughly, and let stand at room temperature for 30 min. Measure the absorbance at 660 nm and record as A_{blank} .
3. Standard tube: In a micro glass cuvette or microplate, add 10 μL standard solution, 20 μL Reagent III, and 4 μL Reagent IV. Mix thoroughly. After color development, add 166 μL distilled water, mix thoroughly, and let stand at room temperature for 30 min. Measure the absorbance at 660 nm and record as A_{standard} .
4. Measurement tube: In a micro glass cuvette or microplate, add 10 μL supernatant, 20 μL Reagent III, and 4 μL Reagent IV. Mix thoroughly. After color development, add 166 μL distilled water, mix thoroughly, and let stand at room temperature for 30 min. Measure the absorbance at 660 nm and record as $A_{\text{measurement}}$.

The blank tube and standard tube only need to be measured once.

4. S-NP Activity Calculation

Definition of activity unit: At 37°C, the release of 1 μmol phenol per gram of soil per day is defined as one enzyme activity unit.

$$\text{S-NP (U/g)} = [C_{\text{standard}} \times (A_{\text{measurement}} - A_{\text{blank}}) \div (A_{\text{standard}} - A_{\text{blank}})] \times V_{\text{total}} \div W \div T$$

$$\text{S-NP (U/g)} = 0.725 \times (A_{\text{assay}} - A_{\text{blank}}) \div (A_{\text{standard}} - A_{\text{blank}}) \div W$$

C_{standard}	0.5 $\mu\text{mol/mL}$
V_{total}	Total volume of the catalytic system, 1.45 mL
W	Soil sample mass, g
T	Catalytic reaction time, 24 h = 1 d

Precautions

1. This 100T kit can test 96 samples.
2. Absolute ethanol and toluene must be prepared separately.
3. Reagent IV cannot be used after it turns brown.

Visual Reference