

Soil Acid Phosphatase (S-ACP) Activity Assay Kit

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

Operation Equipment: Spectrophotometer

Catalog Number: AK0590

Size: 50T/48S

Components:

Reagent I: Liquid 21 mL×1 , storage at 4°C . Protect from light.

Reagent II: powder×1 bottle, storage at 4°C . Dissolve with 50 mL of distilled water before use.

Reagent III: Liquid 11 mL×1, storage at 4°C .

Reagent IV: Powder×1, storage at 4°C . Dissolve with 1152 μL of absolute ethyl alcohol (required but not provided) and 48 μL of distilled water before use. Do not use any more if it turns brown.

Standard: Liquid 1 mL×1 bottle, storage at 4°C, 0.5 μmol/mL phenol standard solution, storage at 4°C .

Product Description:

Soil phosphatase is an enzyme which catalyze soil organic phosphate mineralization, the activity influence directly the decomposition and transformation of organic phosphate and its bio-availability. The activity is the indicator of evaluating the direction and intensity of soil phosphorus bio-transformation. Soil phosphatase is influenced by the content of carbon, nitrogen, available phosphorus in the soil and pH. Soil phosphatase is divided into three types: acidic, neutral and alkaline phosphatase according to the optimum pH.

In acidic condition, soil acid phosphatase (S-ACP) can hydrolyze disodium phenyl phosphate to phenol and disodium hydrogen phosphate. The activity of S-ACP can be calculated by measuring the amount of phenol produced.

Reagents and Equipments Required but Not Provided:

Spectrophotometer, 37°C incubator, centrifuge, transferpettor, table centrifuge, 1 mL glass cuvette, analytical balance, toluene, alcohol, ice and distilled water.

Procedure:

I. Crude enzyme preparation:

Add 0.05 mL of methylbenzene to 0.1 g of dry soil sample, shake slightly for 15 minutes. Add 0.4 mL of Reagent I, mix thoroughly and keep in 37°C incubator for 24 hours. Then add 1 mL of Reagent II immediately and mix thoroughly to stop the catalysis. Centrifuge at 8000 rpm for 10 minutes at room temperature, take the supernatant on ice for testing.

II. Determination procedure:

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

2. Blank tube: Take a 1 mL glass cuvette, add 50 μ L of Reagent I, 200 μ L of Reagent III, 20 μ L of Reagent IV, mix thoroughly. Then add 730 μ L of distilled water after color development. Mix thoroughly and place for 30 minutes at room temperature. Determine the absorbance at 660 nm and record as A_B .
3. Standard tube: Take a 1 mL glass cuvette, add 50 μ L of standard solution, 200 μ L of Reagent III, 20 μ L of Reagent IV, mix thoroughly. Then add 730 μ L of distilled water after color development. Mix thoroughly and place for 30 minutes at room temperature. Determine the absorbance at 660 nm and record as A_S .
4. Test tube: Take a 1 mL glass cuvette, add 50 μ L of supernatant, 200 μ L of Reagent III, 20 μ L of Reagent IV, mix thoroughly. Then add 730 μ L of distilled water after color development. Mix thoroughly and place for 30 minutes at room temperature. Determine the absorbance at 660 nm and record as A_T .

Note: Blank tubes only need to be tested 1-2 times.

III. S-ACP activity calculation:

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the production of 1 nmol of phenol in the reaction system per day every gram soil sample.

$$\begin{aligned} \text{S-ACP(nmol/d/g)} &= [C \times (A_T - A_B) \div (A_S - A_B)] \times V_{rv} \times 1000 \div W \div T \\ &= 725 \times (A_T - A_B) \div (A_S - A_B) \div W \end{aligned}$$

C: Standard concentration, 0.5 μ mol/mL;

V_{rv} : Total volume in catalyze system, 1.45 mL;

W: Soil sample weight, g;

T: Reaction time, 24 hours=1 day;

1000: 1 μ mol=1000 nmol.

Recent Protect Citations:

[1] Liu B, Wang S, Wang J, et al. The great potential for phytoremediation of abandoned tailings pond using ectomycorrhizal *Pinus sylvestris*[J]. *Science of The Total Environment*, 2020, 719: 137475.

[2] Hou Q, Wang W, Yang Y, et al. Rhizosphere microbial diversity and community dynamics during potato cultivation[J]. *European Journal of Soil Biology*, 2020, 98: 103176.

References:

[1] 关松荫.土壤酶及其研究法[M].北京: 科学出版社, 1982.

Related Products:

- AK0566/AK0565 Soil Alkaline Phosphatase(S-AKP/ALP) Activity Assay Kit
- AK0594/AK0593 Soil Polyphenoloxidase Activity Assay Kit
- AK0592/AK0591 Soil Urease(UE) Activity Assay Kit