

Soil Acid Phosphatase (S-ACP) Activity Assay Kit

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

Operation Equipment: Spectrophotometer

Catalog Number: BC0140

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 μL of Reagent I, 200 μL of Reagent III, 20 μL of Reagent IV, mix thoroughly. Then add 730 μL 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 μL of standard solution, 200 μL of Reagent III, 20 μL of Reagent IV, mix thoroughly. Then add 730 μL 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 μL of supernatant, 200 μL of Reagent III, 20 μL of Reagent IV, mix thoroughly. Then add 730 μL 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.

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

C: Standard concentration, 0.5 $\mu\text{mol/mL}$;

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

W: Soil sample weight, g;

T: Reaction time, 24 hours=1 day;

1000: 1 μmol =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:

BC0280/BC0285 Soil Alkaline Phosphatase(S-AKP/ALP) Activity Assay Kit

BC0110/BC0115 Soil Polyphenoloxidase Activity Assay Kit

BC0120/BC0125 Soil Urease(UE) Activity Assay Kit