

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

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

Operation Equipment: Spectrophotometer/ Microplate Reader

Catalog Number: AK0565

Size: 100T/96S

Components:

Reagent I: Liquid 42 mL×1. Storage at 4°C in shadow.

Reagent II: Powder×1 . Storage at 4°C . Dissolve with 100 mL of distilled water before use.

Reagent III: Liquid 2.5 mL×1.Storage at 4°C .

Reagent IV: Powder×1. Storage at 4°C and protect from light. Dissolve with 576 μL of absolute ethyl alcohol (required but not provided) and 24 μL of distilled water before use. Do not use any more if it turns brown.

Standard: Liquid 1 mL×1. Storage at 4°C, 0.5 μmol/mL Phenol standard solution.

Product Description:

Soil phosphatase is an enzyme which catalyzes soil organic phosphate mineralization, the activity level influence the decomposition and transformation of organic phosphate and its bio-availability directly, which is the index 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, its divided into three types: acidic, neutral and alkaline according to the optimum pH.

In alkaline condition, soil alkaline phosphatase (S-AKP/ALP) can catalyzes disodium phenyl phosphate to form phenol and disodium hydrogen phosphate, the activity of S-AKP/ALP can be calculated by detecting the content of phenol.

Reagents and Equipment Required but Not Provided:

Spectrophotometer/microplate reader, micro glass cuvette/96 well flat-bottom plate, desk centrifuge, transferpettor, 37°C constant temperature incubator, analytical balance, mythylbenzene (express delivery not allowed), alcohol, ice and distilled water.

Determination Procedure:

I. Catalytic reaction

Crude enzyme preparation: Add 0.05 mL of mythylbenzene 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 constant temperature incubator for 24 hours. Then add 1 mL of Reagent II quickly to stop the catalysis. Centrifuge at 10000 rpm for 10 minutes at room temperature, take the supernatant on ice for test.

II. Chromogenic reaction

1. Preheat Spectrophotometer/ microplate reader for 30 minutes, adjust the wavelength to 660 nm, set

zero with distilled water.

2. Blank tube: Take 1 mL glass cuvette, add 10 μ L of Reagent I, 20 μ L of Reagent III, 4 μ L of Reagent IV, mix thoroughly, add 166 μ L of distilled water after color development. Mix thoroughly. Allow to stand for 30 minutes at room temperature. Determine the absorbance at 660 nm and record as A_B .

3. Standard tube: Take 1 mL glass cuvette, add 10 μ L of standard solution, 20 μ L of Reagent III, 4 μ L of Reagent IV, mix thoroughly, add 166 μ L of distilled water after color development. Mix thoroughly. Allow to stand for 30 minutes at room temperature. Determine the absorbance at 660 nm and record as A_S .

4. Test tube: Take 1 mL glass cuvette, add 10 μ L of supernatant, 20 μ L of Reagent III, 4 μ L of Reagent IV, mix thoroughly, add 166 μ L of distilled water after color development. Mix thoroughly. Allow to stand for 30 minutes at room temperature. Determine the absorbance at 660 nm and record as A_T .

Note: blank tube and standard tube just need to test once or twice.

III. S-AKP/ALP activity calculation:

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the generation of 1 nmol of phenol in the reaction system per day(24 hours) at 37 $^{\circ}$ C every gram soil sample.

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

C: Concentration of standard solution, 0.5 μ mol/mL;

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

W: Sample weight, g;

T: Reaction time, 24 h=1 day;

1000: Unit conversion factor, 1 μ mol=1000 nmol.

Note:

The linear range is 0.03125 μ mol/mL-2.5 μ mol/mL.

Recent Product 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] Shao T, Zhao J J, Liu A, et al. Effects of soil physicochemical properties on microbial communities in different ecological niches in coastal area[J]. *Applied Soil Ecology*, 2020: 103486.

References:

[1] 关松荫.土壤酶及其研究法[M].北京: 科学出版社, 1982

Related Products:

AK0592/AK0591	Soil Urease(UE) Activity Assay Kit
AK0594/AK0593	Soil Polyphenoloxidase Activity Assay Kit
AK0586/AK0585	Soil β -glucosidase(β - GC) Activity Assay Kit
AK0508/AK0507	Soil Peroxidase Activity Assay Kit