Soil Lignin Peroxidase (S-Lip) Activity Assay Kit

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

Operation Equipment: Microplate reader/Spectrophotometer

Catalog Number: BC1975

Size:100T/48S

Components:

Reagent I: 25 mL×1, stored at 4℃.

Reagent II: Powder×1. storage at 4℃ and protect from light. Before use, add 6 mL of ethanol into bottle, fully dissolve it for standby.

Reagent III: 10 μL×1. storage at 4℃. Before use, add 5 mL of distilled water into bottle, fully dissolve it for standby.

Product Description

Lignin peroxidase (EC1.11.1.14) is a kind of peroxidase containing heme, which belongs to lignin degradation enzyme system. It has great application potential in lignin biodegradation, papermaking industry, textile industry, aromatics transformation and degradation， and environmental pollution control. Resveratrol is oxidized by lignin peroxidase to form resveraldehyde, which has a characteristic absorption peak at 310 nm.

Reagents and Equipment Required but Not Provided.

Scales, low temperature centrifuge, ultraviolet spectrophotometer/microplate reader, micro quartz cuvette/96 well plate(UV), oscillating instrument, toluene, ethanol, 30-50 mesh sieve (or smaller), distilled water.

Procedure

I. Sample processing:

The fresh soil samples are dried naturally and screened with 30-50 mesh.

II. Determination steps:

1. Preheat ultraviolet spectrophotometer/microplate reader for 30 minutes, adjust the wavelength to 310 nm, set zero with the distilled water.

2. The Reagent II is dilute 10 times by ethanol for standby. Prepare as much as you need. Add reagents in turn according to the following table:

Reagent name	Test tube (T)	Contrast tube (C)
Soil sample (g)	0.03	0.03
Toluene (μL)	15	15
Allow to stand for 15 minutes at room temperature.		
Reagent I (μL)	240	240

Reagent II (μL)	30	-
Reagent III (μL)	15	-
After reaction in water bath at 30℃ for 1 hour, boil immediately for 5 minutes.		
Reagent II (μL)	-	30
Reagent III (μL)	-	15
Centrifugate at 14000 ×g for 10 minutes at room temperature, take 200 μL of supernatant to micro quartz cuvette/96 well plate(UV) and measure the absorbance value at 310 nm, record it as A_T and A_C respectively, calculate $\Delta A= A_T-A_C$.		

III. Calculate activity of S-LiP

1. Calculated by micro glass cuvette

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the generation of 1 nmol of resveratrol in the reaction system per minute every g soil sample.

S-LiP (U/g weight) = $\Delta A \div (\varepsilon \times d) \times 10^9 \times V_{RT} \div W \div T = 0.538 \times \Delta A \div W$

ε: Molar extinction coefficient of resveratrol: 9300 L/mol/cm;

d: Light diameter of cuvette, 1 cm;

V_{RT}: The total volume of reaction, 300 μL = 3×10^{-4} L;

W: Mass of soil sample, g;

T: Reaction time, 60 min;

10^9: Unit conversion coefficient, 1mol = 10^9 nmol.

2. Calculated by 96 well plate(UV)

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the generation of 1 nmol of resveratrol in the reaction system per minute every g soil sample.

T-LiP (U/g weight) = $\Delta A \div (\varepsilon \times d) \times 10^9 \times V_{RT} \div W \div T = 0.896 \times \Delta A \div W$

ε: Molar extinction coefficient of resveratrol: 9300 L/mol/cm;

d: Light diameter of cuvette, 0.6 cm;

V_{RT}: The total volume of reaction, 300 μL = 3×10^{-4} L;

W: Mass of soil sample, g;

T: Reaction time, 60 min;

10^9: Unit conversion coefficient, 1mol = 10^9 nmol.

Experimental example:

1. Take 2 pieces 0.03g grass to 1.5ml EP tube, one is test tube and the other is contract tube, operate as the procedure, $\Delta A=A_T-A_C=1.399-0.9924=0.4066$, calculate content by sample weight: S-Lip Activity （U/g weight)= $0.538\times\Delta A\div W=0.538\times0.4066\div0.03=7.292$ U/g weight.
2. Take 2 pieces 0.1g soil to 1.5ml EP tube, one is test tube and the other is contract tube, operate as the procedure, $\Delta A=A_T-A_C=1.3831-0.9924=0.3907$, calculate content by sample weight: S-Lip Activity

（U/g weight)= 0.538×ΔA÷W=0.538×0.3907÷0.03=7.007 U/g weight.

Related products：

BC0110/BC0115 Soil Polyphenoloxidase Activity Assay Kit

BC0120/BC0125 Soil Urease(UE) Activity Assay Kit

BC0240/BC0245 Soil Saccharase(S-SC) Activity Assay Kit