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# Soil β-1,4-Glucanase/ Cellobiosidase (S-C1) Activity Assay Kit

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

**Operation Equipment:** Spectrophotometer/ Microplate reader

Catalog Number: AK0117

**Size:** 100T/48S

# **Components:**

Reagent I: Toluene 2mL×1. Storage at 4C. (self-provided reagent)

Reagent II: Powder×2. Storage at 4C. Add 7.5 mL of Reagent III to fully dissolve when the solution

will be used. Store unused reagents at 4°C.

Reagent III:  $40 \text{ mL} \times 1$ . Storage at 4C.

Reagent IV: 30 mL×1. Storage at 4C.

Standard solution: 1mL×1, 5mmol/L p-nitrophenol solution. The standard is diluted 50 times with reagent

III to obtain a 100µmol/L standard solution before use.

# **Product Description**

β- 1,4-glucanase/cellobiosidase (C1, EC3.2. 1.91) exists in bacteria, fungi and animals, and is a component of the cellulase system. The end of the linear molecule hydrolyzes the β-glucosidic bond and cuts out one cellobiose molecule every time.

S-C1 can catalyze p-nitrobenzene cellobiose (PNPC) to p-nitrophenol, which has a characteristic light absorption at 400nm.

#### Reagents and Equipment Required but Not Provided

Spectrophotometer/ Microplate reader, centrifuge, water-bath, transferpettor, micro glass cuvette/ 96-well flat-bottom plate, mortar, **toluene**, sieve (30-50 mesh) and distilled water.

#### **Procedure**

#### 1. Sample preparation:

Fresh soil samples are naturally air-dried or oven dried at 37°C and passed through a 30-50 mesh sieve.

## 2. Determination steps and sample adding table:

- a. Preheat spectrophotometer/microplate reader more than 30 min, adjust wavelength to 400 nm and set zero with distilled water.
- b. Operate according to the following table:

Reagent Name	Test tube (T)	Control tube (C)	Standard tube (S)	Blank tube (B)
Soil sample (g)	0.03	0.03		
Reagent I	15	15		
Mix by shaking to make the soil sample wet and leave it for				





15min at room temperate	ure			
Reagent II (μL)	120			
Reagent III (μL)	150	150		
Mix well. After react	ing for 1 h at 370			
immediately boil in a v	water bath for 5 m			
prevent water loss) a	and cool in runnin			
Reagent II		120		
Centrifuge at 10,000 rp	om and 25°C for 1			
supernatant				
Supernatant	100	100		
Standard solution (µL)	-	_	100	
Distilled water				100
Reagent IV (μL)	200	200	200	200

Mix well, react for 2 minutes at RT. record the absorption value a of each tube at 400 nm, calculate  $\Delta A = A_T - A_C$ ,  $\Delta A_S = A_S - A_B$ 

## Calculation of S-C1 activity:

1. Calculation of S-C1activity:

Unit definition: Unit definition: one unit is defined as the amount of enzyme that catalyzes the production of 1  $\mu$ mol ofp-nitrophenol per day every gram of soil catalyzes at 37C.

S-C1 activity (U/mg) =
$$\Delta A \div (\Delta A_S \div C_S) \times V1 \div W \div T = 0.684 \times \Delta A \div \Delta A_S \div W$$

 $C_S$ : concentration of standard solution, 100  $\mu$  mol/L

V1: the volume of reaction system, 2.85×10-4 L;

W: sample fresh weight, g;

T: reaction time: 1/24d.

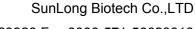
# Note

1. If the absorbance value is greater than 1.5, it is recommended to dilute the supernatant with reagent III and determine with decrease the quality of soil samples.

#### **Experimental Examples:**

- 1. Take two tubes of  $0.03\,g$  soil sample, which are the measuring tube and the control tube. Follow the measuring steps and mark them as At and Ac. Measure with 96-well plate and calculate alculate  $\Delta$ At=At-A=0.444-0.244=0.2,  $\Delta$ As=As-A b=0.413-0.048=0.365, calculate the enzyme activity:
  - S-C1 activity (U/g soil)  $=0.684 \times \Delta At \div \Delta As \div W = 0.684 \times 0.2 \div 0.365 \div 0.03 = 12.493$  U/g soil.
- 2. Take two tubes of 0.03g forest soil samples, which are the measuring tube and the control tube. Follow the measuring steps and mark them as At and Ac. Measure with 96-well plate and calculate  $\Delta$ At =At-Ac=0.33-0. 195=0. 135,  $\Delta$ A s=As-Ab=0.413-0.048=0.365, calculate the enzyme activity:

S-C1 activity (U/g soil ) =  $0.684 \times \Delta At \div \Delta As \div W = 0.684 \times 0.1135 \div 0.365 \div 0.03 = 8.4329$  U/g soil





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# **Related Products:**

AK0122/AK0121 Soil  $\beta$ -Xylosidase(S- $\beta$ -XYS) Activity Assay Kit AK0155/AK0154 Soil  $\alpha$ -glucosidase(S- $\alpha$ -GC) Activity Assay Kit Soil Saccharase(S-SC) Activity Assay Kit