

Soil β -Xylosidase (S- β -XYS) Activity Assay Kit

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

Operation Equipment: Spectrophotometer

Catalog Number: AK0122

Size:50T/24S

Components:

Reagent I: Methylbenzene 3 mL \times 1, Storage at 4°C (**self-provided reagent**).

Reagent II : Liquid 30 mL \times 1. Storage at 4°C .

Reagent III : Powder \times 2. Storage at -20°C . Add 10 mL of distilled water to each bottle before use. The left reagent store at -20°C .

Reagent IV: Liquid 60 mL \times 1. Storage at 4°C .

Standard: Liquid 1 mL \times 1. Storage at 4°C . 5 mmol/L Phenol standard solution. Dilute the standard solution for 50 times to 100 μ mol/L with the Reagent I before use.

Product Description:

Soil β -xylosidase (S- β -XYS) exists in organisms such as plants, bacteria and fungi, which is a key enzyme that catalyzes the degradation of xylan hemicellulose. The product xylose can be used as a carbon source in microbial fermentation. In addition, β -XYS can also be used as a biological bleaching agent in the paper industry, which is more environment-friendly than traditional bleaching methods and has a widespread application value.

S- β -XYS can catalyze the p-nitrophenyl beta-xylopyranoside to p-nitrophenol. The product has characteristic of absorption at 400 nm. In this kit, the S- β -XYS activity is quantified by measuring the increase in the color development at 400 nm.

Reagents and Equipment Required but Not Provided:

Spectrophotometer, water-bath, desk centrifuge, transferpettor, 1 mL glass cuvette, analytical balance, mortar, 30-50 mesh sieve, **methylbenzene**, ice and distilled water.

Procedure:

I. Preparation of samples

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

II. Determination procedure:

- Preheat Spectrophotometer for 30 minutes, adjust the wavelength to 400 nm, set zero with distilled water.
- Add reagents in 1 mL glass cuvette as the following:

Reagent	Test tube (T)	Contrast Tube (C)	Standard tube (S)	Blank tube (B)
Air-dried soil (g)	0.1	0.1	-	-

Reagent I (μL)	20	20	-	-
The soil samples are all wetted by oscillating mixing, and store at 37°C for 15 minutes.				
Reagent II (μL)	500	500	-	-
Reagent III	400	-	-	-
Mix thoroughly and incubate the reaction for 1 hour at 50°C water bath, then take the reaction solution in a boiling water bath for 5 minutes immediately (tightly close to prevent moisture loss), flowing water to cool.				
Reagent III (μL)	-	400	-	-
Mix thoroughly, centrifuge at 10000 rpm for 10 minutes at 25°C and take the supernatant.				
Supernatant (μL)	500	500	-	-
Standard solution (μL)	-	-	500	-
Distilled water (μL)	-	-	-	500
Reagent IV (μL)	1000	1000	1000	1000

Mix thoroughly and stand at room temperature for 2 minutes, centrifuge at 10000×g for 5 minutes. Take the supernatant and detect the absorbance of each tube at 400 nm and noted as A_T , A_C , A_S and A_B . Calculate $\Delta A_T = A_T - A_C$, $\Delta A_S = A_S - A_B$. Each test tube should be provided with one contrast tube.

III. S-NAG activity calculation:

Unit definition: One unit of enzyme activity is defined as the amount of enzyme that catalyzes the generation 1μmol ofp-nitrophenol every gram of soil sample in the reaction system per day.

$$S\text{-}\beta\text{-XYS (U/g soil sample)} = \Delta A_T \div (\Delta A_S \div C) \times V_{rv} \div W \div T = 2.21 \times \Delta A_T \div \Delta A_S \div W$$

C: Concentration of standard solution, 100 μmol/L;

V_{rv} : Total volume in catalyze system, 9.2×10^{-4} L;

W: Soil sample weight, g;

T: React time, 1 hour = 1/24 day;

Experimental Examples:

1. Take two tubes of 0.1g soil, which are the measuring tube and the control tube. Follow the measuring steps and mark them as A_t and A_c . Calculate $\Delta A_t = A_t - A_c = 0.46 - 0.221 = 0.239$, $\Delta A_s = A_s - A_b = 0.568 - 0.002 = 0.566$, calculate the enzyme activity:

$$S\text{-NAG activity (U/g soil)} = 2.21 \times \Delta A_t \div \Delta A_s \div W = 2.21 \times 0.239 \div 0.566 \div 0.1 = 9.3312 \text{ U/g soil.}$$

2. Take two tubes of 0.1g forest soil samples, which are the measuring tube and the control tube. Follow the measuring steps and mark them as A_t and A_c . Calculate $\Delta A_t = A_t - A_c = 0.356 - 0.109 = 0.247$, $\Delta A_s = A_s - A_b = 0.568 - 0.002 = 0.566$, calculate enzyme activity:

$$S\text{-NAG activity (U/g soil)} = 2.21 \times \Delta A_t \div \Delta A_s \div W = 2.21 \times 0.247 \div 0.566 \div 0.1 = 9.6443 \text{ U/g soil}$$

Related Products:

AK0155/AK0154 Soil α-glucosidase(S-α-GC) Activity Assay Kit

AK0574/AK0573 Soil Saccharase(S-SC) Activity Assay Kit