

Soil Cellulase (S-CL) Activity Assay Kit

Note: It is necessary to predict 2-3 large difference samples before the formal determination.

Operation Equipment: Spectrophotometer

Catalog Number: BC0150

Size:50T/24S

Product Composition: Before use, please carefully check whether the volume of the reagent is consistent with the volume in the bottle. If you have any questions, please contact Solarbio staff in time.

Reagent name	Size	Preservation Condition
Reagent I	Self-Provided Reagent	-
Reagent II	Liquid 15 mL×1	2-8°C
Reagent III	Liquid 50 mL×1	2-8°C
Reagent IV	Liquid 15 mL×1	2-8°C
Standard	Powder ×1	2-8°C

Solution Preparation:

1. Reagent I: About 10mL methylbenzene (Required but not provided), store at RT. A 30mL brown reagent bottle is provided in the kit. Please label the reagent name yourself.
2. Standard: Contain 10 mg of anhydrous glucose (dry weight loss < 0.2%). Dissolve the standard with 1 mL of distilled water to generate a 10 mg/mL glucose solution standard, store at 2-8°C and use within two weeks or dissolve the standard with saturated benzoic acid solution stored for a longer time.

Product Description:

Soil Cellulase (S-CL) mainly comes from soil microorganisms. Glucose produced by S-CL is the main carbon source nutrients of soil microorganisms. In this kit, this product uses the 3,5-dinitrosalicylic acid method to determine the content of reducing sugars produced by S-CL catalyzing cellulose degradation.

Reagents and Equipment Required but Not Provided:

Spectrophotometer, water-bath, adjustable pipette, 1 mL glass cuvette, 30-50 mesh sieve, mortar, methylbenzene (>98%, AR), ice and distilled water.

Procedure:

I. Sample processing

Fresh soil samples are naturally air-dried or oven to dry at 37°C, then sieved by 30-50 mesh sieve.

II. Determination procedure

1. Preheat the spectrophotometer for more than 30 minutes, adjust the wavelength to 540 nm, and set zero with distilled water.
2. Standard preparation: Dilute the standard to 1, 0.8, 0.6, 0.4, 0.2, 0.1 mg/mL with distilled water.
3. Add reagents as the following table.

Reagent	Contrast Tube (C)	Test Tube (T)	Standard Tube (S)	Blank Tube (B)
Water-free soil (g)	0.25	0.25	-	-
Reagent I (μL)	125	125	-	-
-	Boil for 15 minutes (Wrap the sealing film to prevent bursting)	Shack to mix thoroughly, place at RT for 15 minutes	-	-
Reagent II (μL)	250	250	-	-
Reagent III (μL)	1000	1000	-	-
Distilled water (μL)	250	250	-	-
Shake to mix thoroughly, then saccharification in water bath at 40°C for 1 hour. After the saccharification, boil for 15 minutes (Wrap the sealing film to prevent bursting), 10000rpm for 10min at RT and take the supernatant (saccharification liquid).				
Saccharified liquid (μL)	50	50	-	-
Standard solution (μL)	-	-	50	-
Distilled water (μL)	-	-	-	50
Reagent IV (μL)	150	150	150	150
Mix thoroughly, boil in boiling water bath for 15 minutes (Wrap the sealing film to prevent bursting), then leave the tube to cool.				
Distilled water (μL)	1050	1050	1050	1050
Mix thoroughly, take 1000μL solution into 1 mL glass cuvette, then detect the absorbance at 540 nm and noted as A _C , A _T , A _S , and A _B . $\Delta A_T = A_T - A_C$. $\Delta A_S = A_S - A_B$. The blank tube and standard curve only need to be measured 1-2 times. A contrast tube is required for each test tube.				

III. Calculation

1. Standard curve

The concentration of standard solution as x-axis, ΔA_S as y-axis, obtain the equation $y=kx+b$. Take ΔA_T to the equation to acquire x value (mg/mL).

2. Calculation

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the production of 1 milligram of glucose in the reaction system per day every gram soil sample.

$$S\text{-CL activity (U/g weight)} = x \times V_r \div W \div T = 156 \times x$$

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

V_r: Total reaction volume, 1.625 mL;

W: Sample weight, 0.25 g.

Note:

Prolong the reaction time at 40°C (24h or more) if the absorbance of sample tube is too small (0.01) or increase the adding volume of saccharification liquid in the coloration step while decrease the volume of distilled water correspondingly. And modify the calculation formula.

Recent Product Citations:

- [1] Huang J, Ye J, Gao W, Liu C, Price GW, Li Y, Wang Y. Tea biochar-immobilized *Ralstonia Bcul-1* increases nitrate nitrogen content and reduces the bioavailability of cadmium and chromium in a fertilized vegetable soil. *Sci Total Environ.* 2023 Mar 25; 866:161381. doi: 10.1016/j.scitotenv.2022.161381. Epub 2023 Jan 5. PMID: 36621509.
- [2] Guan TK, Wang QY, Li JS, Yan HW, Chen QJ, Sun J, Liu CJ, Han YY, Zou YJ, Zhang GQ. Biochar immobilized plant growth-promoting rhizobacteria enhanced the physicochemical properties, agronomic characters and microbial communities during lettuce seedling. *Front Microbiol.* 2023 Jul 5;14:1218205. doi: 10.3389/fmicb.2023.1218205. PMID: 37476665; PMCID: PMC10354297.
- [3] Pu Q, Zhang K, Poulain AJ, Liu J, Zhang R, Abdelhafiz MA, Meng B, Feng X. Mercury drives microbial community assembly and ecosystem multifunctionality across a Hg contamination gradient in rice paddies. *J Hazard Mater.* 2022 Aug 5;435:129055. doi: 10.1016/j.jhazmat.2022.129055. Epub 2022 May 4. PMID: 35650726.
- [4] Yang L, Muhammad I, Chi YX, Wang D, Zhou XB. Straw Return and Nitrogen Fertilization to Maize Regulate Soil Properties, Microbial Community, and Enzyme Activities Under a Dual Cropping System. *Front Microbiol.* 2022 Mar 15;13:823963. doi: 10.3389/fmicb.2022.823963. PMID: 35369510; PMCID: PMC8965350.
- [5] Song K, Zhou Z, Leng J, Fang S, Zhou C, Ni G, Kang L, Yin X. Effects of rumen microorganisms on the decomposition of recycled straw residue. *J Zhejiang Univ Sci B.* 2023 Apr 15;24(4):336-344. doi: 10.1631/jzus.B2200504. PMID: 37056209; PMCID: PMC10106401.

References:

- [1] Deng S P, Tabatabai M A. Cellulase activity of soils[J]. *Soil Biology and Biochemistry*, 1994, 26(10): 1347-1354.
- [2] Sinegani A A S, Sinegani M S. The effects of carbonates removal on adsorption, immobilization and activity of cellulase in a calcareous soil[J]. *Geoderma*, 2012, 173: 145-151.

Related Protects:

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| BC0280/BC0285 | Soil Alkaline Phosphatase (S-AKP/ALP) Activity Assay Kit |
| BC0110/BC0115 | Soil Polyphenoloxidase (S-PPO) Activity Assay Kit |
| BC0125/BC0125 | Soil Urease (S-UE) Activity Assay Kit |
| BC0140/BC0145 | Soil Acid Phosphatase (S-ACP) Activity Assay Kit |