

## Soil $\beta$ -glucosidase (S- $\beta$ -GC) Activity Assay Kit

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

**Operation Equipment:** Spectrophotometer/ Microplate reader

**Catalog Number:** BC0165

**Size:** 100T/48S

**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	Powder $\times 2$	-20°C
Reagent III	Liquid 30 mL $\times 1$	2-8°C
Reagent IV	Liquid 20 mL $\times 1$	2-8°C
Standard	Liquid 1 mL $\times 1$	2-8°C

### Solution Preparation:

1. Reagent I: About 1.5mL Toluene (Required but not provided). A 30mL brown reagent bottle is provided in the kit. Please label the reagent name yourself.
2. Reagent II: Add 7.5 mL of distilled water to one bottle before use and dissolve it fully. The left reagent could be stored at -20°C for 4 weeks.
3. Standard: 5 mmol/L p-nitrophenol solution.
4. Preparation of standard solution: Take 100  $\mu$ L of standard and add it to 400  $\mu$ L of Reagent III to get 1 mmol/L standard solution. Dilute the 1 mmol/L standard solution for 10 times to 100  $\mu$ mol/L.

### Product Description

Soil  $\beta$ -glucosidase (S- $\beta$ -GC) can catalyze the hydrolysis of glycoside bonds between aryl or hydroxyl groups and glycosylated atomic groups to generate glucose. It is an important component of cellulose decomposition enzyme system and has important physiological functions in the carbohydrate metabolism of soil microorganisms.

S- $\beta$ -GC can catalyze the p-nitrophenyl- $\beta$ -D-glucopyranoside to p-nitrophenol. The product is slightly yellow and has characteristic of absorption at 400 nm.

### Reagents and Equipment Required but Not Provided.

Spectrophotometer/microplate reader, table centrifuge, water bath/constant temperature incubator, transferpettor, micro cuvette/96 well flat-bottom plate, 30-50 mesh sieve, toluene (>98%, AR), ice and distilled water.

### Preparation

## I. Sample processing:

Air dry the fresh soil sample naturally or in an oven at 37°C and sieve it through 30~50 meshes.

## II. Determination procedure:

1. Preheat the spectrophotometer for more than 30 minutes, adjust the wavelength to 400 nm, set spectrophotometer zero with distilled water.
2. **Preparation of standard samples:** Dilute it to 50, 25, 12.5, 6.25 μmol/L with the distilled water. Detect the standard solutions of 100, 50, 25, 12.5 and 6.25 μmol/L.
3. Add reagents with the following list:

Reagent	Test Tube (T)	Contrast Tube	Standard Tube (S)	Blank Tube (B)
Air-dried soil sample	0.02	0.02	-	-
Reagent I (μL)	10	10	-	-
The soil samples are all wetted by oscillating mixing and store at room temperature for 15				
Reagent II (μL)	130	-	-	-
Reagent III (μL)	160	160	-	-
Mix thoroughly and incubate the reaction for 1 hour at 37°C, then take the reaction solution in a boiling water bath for 5 minutes immediately (Wrap the sealing film to prevent bursting), flowing water to cool.				
Reagent II (μL)	-	130	-	-
Mix thoroughly, centrifuge at 10000 ×g for 10 minutes at room temperature and take the				
Supernatant (μL)	70	70	-	-
Standard (μL)	-	-	70	-
Distilled water (μL)	-	-	-	70
Reagent IV (μL)	130	130	130	130

Mix thoroughly and stand at room temperature for 2 min. Detect the absorbance of each tube at 400 nm and noted as  $A_T$ ,  $A_C$ ,  $A_S$  and  $A_B$ . Calculate  $\Delta A_T = A_T - A_C$ ,  $\Delta A_S = A_S - A_B$ . 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 (y, μmol/L) as y-axis,  $\Delta A_S$  (x,  $\Delta A_S$ ) as x-axis, obtain the equation  $y=kx+b$ . Take  $\Delta A_T$  (x,  $\Delta A_T$ ) to the equation to acquire y value (μmol/L).

### 2. Calculation

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

$$\beta\text{-GC activity (U/g weight)} = y \times V_{ra} \div W \div T = 0.36 \times y$$

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

$V_{ra}$ : Total volume of the reaction system,  $3 \times 10^{-4}$  L;

W: Sample weight, 0.02 g.

### Recent Product Citations:

[1] Shu X, Hu Y, Liu W, Xia L, Zhang Y, Zhou W, Liu W, Zhang Y. Linking between soil properties, bacterial communities, enzyme activities, and soil organic carbon mineralization under ecological restoration in an alpine degraded grassland. *Front Microbiol.* 2023 Apr 6; 14:1131836. doi: 10.3389/fmicb.2023.1131836. PMID: 37180269; PMCID: PMC10167489.

[2] Chen W, Guo X, Guo Q, Tan X, Wang Z. Long-Term Chili Monoculture Alters Environmental Variables Affecting the Dominant Microbial Community in Rhizosphere Soil. *Front Microbiol.* 2021 Jul 1; 12:681953. doi: 10.3389/fmicb.2021.681953. PMID: 34276615; PMCID: PMC8281244.

[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] Kou X, Liu H, Chen H, Xu Z, Yu X, Cao X, Liu D, Wen L, Zhuo Y, Wang L. Multifunctionality and maintenance mechanism of wetland ecosystems in the littoral zone of the northern semi-arid region lake driven by environmental factors. *Sci Total Environ.* 2023 Apr 20;870:161956. doi: 10.1016/j.scitotenv.2023.161956. Epub 2023 Feb 1. PMID: 36737024.

[5] Wang YF, Chen P, Wang FH, Han WX, Qiao M, Dong WX, Hu CS, Zhu D, Chu HY, Zhu YG. The ecological clusters of soil organisms drive the ecosystem multifunctionality under long-term fertilization. *Environ Int.* 2022 Mar;161:107133. doi: 10.1016/j.envint.2022.107133. Epub 2022 Feb 8. PMID: 35149447

### References:

[1] Dick W A, Thavamani B, Conley S, et al. Prediction of  $\beta$ -glucosidase and  $\beta$ -glucosaminidase activities, soil organic C, and amino sugar N in reflectance spectroscopy[J]. *Soil Biology & Biochemistry*, 2013, 56(9): 9e104.

[2] Sestelo A B F, Poza M, Villa T G.  $\beta$ -Glucosidase activity in a *Lactobacillus plantarum* wine strain[J]. *World Journal of Microbiology and Biotechnology*, 2004, 20(6): 633.

### Related Products:

BC0280/BC0285	Soil Alkaline Phosphatase(S-AKP/ALP) Activity Assay Kit
BC0110/BC0115	Soil Polyphenoloxidase Activity Assay Kit
BC4040/BC4045	Soil Neutral Invertase(S-NI) Activity Assay Kit
BC4030/BC4035	Soil $\beta$ -1,4-Glucanase Activity Assay Kit
BC4010/BC4015	Soil $\beta$ -Xylosidase(S- $\beta$ -XYS) Activity Assay Kit