

# Soil Urease (S-UE) Activity Assay Kit

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

**Operation Equipment:** Spectrophotometer

**Catalog Number:** BC0120

**Size:** 50T/24S

**Components:** 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 ×2	2-8°C
Reagent III	Liquid 65 mL×1	2-8°C
Reagent IV A	Liquid 2 mL×1	2-8°C
Reagent IV B	Liquid 8 mL×1	2-8°C
Reagent V	Liquid 0.5 mL×1	2-8°C
Standard	Liquid 1 mL×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. Reagent II: Dissolved one Reagent II with 10 mL of distilled water before use. The left reagent can be stored at 2-8°C for four weeks.
3. Reagent IV: Mix Reagent IV A: Reagent IV B=1: 4 according to sample number before use.
4. Reagent V: The liquid is placed in an EP tube inside the bottle and needs to be centrifuged before use. Before use, add 9.5 mL of distilled water, mix well and wait for use; The left reagent can be stored at 2-8°C for two weeks.
5. Standard: 1 mg/mL standard solution.

## Product Description

Urease is an enzyme that catalyzes the hydrolysis of urea into carbon dioxide and ammonia. The microbial quantity of soil, organic matter content, total nitrogen and available nitrogen content have positive correlation with soil urease activity. Soil nitrogen status is determined by soil urease activity.

The ammonia is determined by the indophenol blue method, resulting in blue indophenol produced is proportional to the concentration of ammonia.

## Reagents and Equipment Required but Not Provided.

Spectrophotometer, water bath, transferpeltor, 1 mL glass cuvette, ice, 30-50 mesh sieve (or smaller), methylbenzene (>98%, AR), distilled water.

## Procedure

### I. Sample processing:

The fresh soil sample shall be dried by naturally or in an oven at 37°C, and shall be screened through 30-50 mesh.

### II. Determination procedure:

1. Preheat the spectrophotometer for more than 30 minutes, adjust the wavelength to 630 nm, set zero with distilled water.

#### 2. Sample Preparation

Reagent	Test tube	Contract tube
Air drying soil sample (g)	0.25	0.25
Reagent I (μL)	125	125
Mix thoroughly, wetting all the soil, place at room temperature for 15 minutes.		
Reagent II (μL)	625	-
Distilled water (μL)	-	625
Reagent III (μL)	1250	1250
Mix thoroughly, culture for 24 hours in 37°C water-bath. Centrifuge at 10000 ×g for 10 minutes at room temperature. Take the supernatant for test.		

3. Dilute the supernatant 10 times. (Add 0.9 mL of distilled water to 0.1 mL of the supernatant). Dilute until the absorbance less than 1.

4. Prepare standard solution: Diluted the standard to 8, 6, 4, 2, 1, 0.5, 0.25, 0 μg/mL.

#### 5. Ammonia concentration test

Reagent	Test tube(T)	Contract tube(C)	Standard
Diluted supernatant solution/ Standard	360	360	360
Reagent IV (μL)	120	120	120
Reagent V (μL)	120	120	120
Mix thoroughly, incubate at room temperature for 20 minutes.			
Distilled water (μL)	400	400	400

Mix thoroughly, set the counter to zero with distilled water at 630 nm and measure the absorbance which noted as  $A_T$ ,  $A_C$ ,  $A_S$ ,  $A_B$ . Calculate  $\Delta A_T = A_T - A_C$ ,  $\Delta A_S = A_S - A_B$ . Set a contract cube for each test tube. Blank tube and standard curve only need to test once or twice.

### Calculation

1. According to the concentration (x, μg/mL) of the standard tube and the absorbance  $\Delta A_S$  (y,  $\Delta A_S$ ), establish a standard curve. According to the standard curve, bring  $\Delta A_T$  (y,  $\Delta A_T$ ) into the formula to calculate the sample concentration (x, μg/mL).

2. S-UE activity calculation

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the production of 1  $\mu\text{g}$  of  $\text{NH}_3\text{-N}$  in the reaction system per day every gram of soil sample.

$$\text{S-UE activity (U/g soil sample)} = x \times 10 \times \text{Vrv} \div \text{W} \div \text{T} = 80 \times x$$

10: Dilution factor;

T: Reaction time, 1 day;

Vrv: Total reaction volume, 2 mL;

W: Sample weight ,0.25 g.

### Recent Product Citations:

[1] Sun H, Xing R, Ye X, Yin K, Zhang Y, Chen Z, Zhou S. Reactive oxygen species accelerate humification process during iron mineral-amended sludge composting. *Bioresour Technol.* 2023 Feb; 370:128544. doi: 10.1016/j.biortech. 2022.128544. Epub 2022 Dec 27. PMID: 36584721.

[2] Ahsan T, Tian PC, Gao J, Wang C, Liu C, Huang YQ. Effects of microbial agent and microbial fertilizer input on soil microbial community structure and diversity in a peanut continuous cropping system. *J Adv Res.* 2023 Nov 28: S2090-1232(23)00367-3. doi: 10.1016/j.jare.2023.11.028. Epub ahead of print. PMID: 38030126.

[3] 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.

[4] Yang C, Xia L, Zeng Y, Chen Y, Zhang S. Hexaploid *Salix rehderiana* is more suitable for remediating lead contamination than diploids, especially male plants. *Chemosphere.* 2023 Aug; 333:138902. doi: 10.1016/j.chemosphere. 2023.138902. Epub 2023 May 12. PMID: 37182717.

[5] Bian X, Yang X, Zhang K, Zhai Y, Li Q, Zhang L, Sun X. Potential of *Medicago sativa* and *Perilla frutescens* for overcoming the soil sickness caused by ginseng cultivation. *Front Microbiol.* 2023 Apr 5;14:1134331. doi: 10.3389/fmicb.2023.1134331. PMID: 37089541; PMCID: PMC10113677.

### References:

[1] Kandeler E, Gerber H. Short-term assay of soil urease activity using colorimetric determination of ammonium[J]. *Biology and fertility of Soils*, 1988, 6(1): 68-72.

[2] Witte C P, Medina-Escobar N. In-gel detection of urease with nitroblue tetrazolium and quantification of the enzyme from different crop plants using the indophenol reaction[J]. *Analytical biochemistry*, 2001, 290(1): 102-107.

[3] Guo H, Yao J, Cai M, et al. Effects of petroleum contamination on soil microbial numbers, metabolic activity and urease activity[J]. *Chemosphere*, 2012, 87(11): 1273-1280.

### Related Products:

BC0110/BC0115 Soil Polyphenoloxidase (S-PPO) Activity Assay Kit  
BC0280/BC0285 Soil Alkaline Phosphatase(S-AKP/ALP) Activity Assay Kit  
BC4040/BC4045 Soil Neutral Invertase(S-NI) Activity Assay Ki