

## Soil Dehydrogenase(S-DHA) Activity Assay Kit

**Note:** Before the experiment, it is recommended to select 2-3 sample with large expected differences for pre-experiment.

**Operation Equipment:** Spectrophotometer

**Catalog Number:** BC0390

**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 A	Powder×2	2-8°C
Reagent I B	Powder×2	2-8°C
Reagent II	Liquid 40 mL×1	2-8°C
Reagent III	Liquid 60 mL×1 (Required but not provided)	2-8°C

### Solution Preparation:

- Reagent I:** Before use, take a bottle of reagent I-B and add it to a bottle of reagent I-A, dissolve it with 8mL distilled water (about 20S), ready for use. After preparation, it should be stored at 2-8°C away from light. It is best to use it within a week. If it appears red, it should not be used.
- Reagent III:** Ethyl Acetate, self-provided.

### Product Description:

The activity of Soil Dehydrogenase (S-DHA) can reflect the active microbial biomass and its degradation activity towards organic matter in the soil system, and can be used as an indicator of soil microbial degradation performance.

The hydrogen receptor 2,3,5-Triphenyl Tetrazolium Chloride (TTC) is reduced to Triphenyl Formazine (TF) during cellular respiration after receiving hydrogen. The TF appears red and has a maximum absorption peak at a wavelength of 485 nm. Its absorbance value is measured at 485 nm to obtain soil dehydrogenase activity.

### Required reagents and equipment:

Visible spectrophotometer, 1mL glass cuvette, sieve (30-50 meshes), balance, constant temperature incubator/water bath, low-temperature centrifuge, ice, distilled water, ethyl acetate (>98%, AR).

### Procedure:

#### I. Sample preparation

- Soil sample: Take 0.1 g of fresh soil sample which passed through 30-50 meshes sieve (To ensure that TTC is in full contact with soil particles).

2. Mud sample: Wash mud with distilled water, centrifuge at 12000 rpm for 10 minutes at 25°C, discard supernatant, repeat 3- 4 times.

## II. Determination operation

1. Preheat Spectrophotometer/microplate reader for 30 minutes, adjust the wavelength to 485 nm, and zero with distilled water.
2. Add the following reagents (add the following reagents in sequence to a 2 mL EP tube):

Reagent	Control tube (A <sub>C</sub> )	Test tube (A <sub>T</sub> )
Sample (g)	0.1	0.1
Reagent I (mL)	-	0.4
Reagent II (mL)	0.8	0.4
Mix thoroughly, incubate at 37°C dark place for 24 hours, then keep on ice for 5 minutes immediately.		
Reagent III (mL)	1.2	1.2
<p><b>Vigorously shake</b> for 10 min, 15000 rpm, 4 °C and centrifuge for 10 min. Take 1 mL of supernatant into a 1 mL glass cuvette, measure the absorbance of the control tube and the measurement tube at 485 nm, and record them as A<sub>C</sub> and A<sub>T</sub>, respectively. Calculate <math>\Delta A = A_T - A_C</math>.</p> <p>Note: One test tube is required for one control tube.</p>		

## III. Calculation:

Definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the increasing absorbance of every 0.01 for per hour in per milliliter reaction system at 37°C every gram of sample.

$$s\text{-DHA (U/g)} = \Delta A \div 0.01 \div T \div W \times V_{rv} = 83.33 \times \Delta A$$

T: Incubation time, 24 hours

W: Sample weight, 0.1g;

V<sub>rv</sub>: Total reaction volume, 2 mL.

### Note:

1. Reagent III is volatile and toxic, please wear lab clothes, masks and latex gloves for your health.
2. If the absorbance is larger, detect again after decreasing the sample. If the absorbance value is too small, the culture time can be appropriately prolonged.
3. If the supernatant to be tested remains turbid after centrifugation, try increasing the centrifugation speed or extending the time, such as 15000rpm, 4 °C, centrifugation for 20 minutes.

## Recent Product Citations:

[1] Han Z, Osman R, Liu Y, Wei Z, Wang L, Xu M. Analyzing the impacts of cadmium alone and in co-existence with polypropylene microplastics on wheat growth. *Front Plant Sci.* 2023 Aug 10; 14:1240472. doi: 10.3389/fpls.2023. 1240472. PMID: 37636097; PMCID: PMC10449543.

[2] Ali M, Song X, Wang Q, Zhang Z, Zhang M, Chen X, Tang Z, Liu X. Thermally enhanced

biodegradation of benzo[a]pyrene and benzene co-contaminated soil: Bioavailability and generation of ROS. *J Hazard Mater.* 2023 Aug 5; 455:131494. doi: 10.1016/j.jhazmat.2023.131494. Epub 2023 Apr 25.

PMID: 37172381.

[3] Li X, Lu H, Yang K, Zhu L. Attenuation of tetracyclines and related resistance genes in soil when exposed to nanoscale zero-valent iron. *J Hazard Mater.* 2023 Apr 15; 448:130867. doi: 10.1016/j.jhazmat.2023.130867. Epub 2023 Jan 31. PMID: 36758429.

[4] 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.

[5] Zhou H, Liu Q, Jiang L, Shen Q, Chen C, Zhang C, Tang J. Enhanced remediation of oil-contaminated intertidal sediment by bacterial consortium of petroleum degraders and biosurfactant producers. *Chemosphere.* 2023 Jul; 330:138763. doi: 10.1016/j.chemosphere.2023.138763. Epub 2023 Apr 22. PMID: 37094722.

[6] Li N, Chen J, Liu C, Yang J, Zhu C, Li H. Cu and Zn exert a greater influence on antibiotic resistance and its transfer than doxycycline in agricultural soils. *J Hazard Mater.* 2022 Feb 5; 423(Pt B):127042. doi: 10.1016/j.jhazmat.2021.127042. Epub 2021 Aug 26. PMID: 34536850.

#### References :

[1] Kumar S, Chaudhuri S, Maiti S K. Soil dehydrogenase enzyme activity in natural and mine soil-a review[J]. *Middle-East Journal of Scientific Research*, 2013, 13(7): 898-906.

[2] Friedel J K, Mölter K, Fischer W R. Comparison and improvement of methods for determining soil dehydrogenase activity by using triphenyltetrazolium chloride and iodinitrotetrazolium chloride[J]. *Biology and fertility of soils*, 1994, 18(4): 291-296.

#### Related Products :

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|---------------|---|
| BC0280/BC0285 | Soil Alkaline Phosphatase(S-AKP/ALP) Activity Assay Kit |
| BC0110/BC0115 | Soil Polyphenoloxidase Activity Assay Kit               |
| BC4030/BC4035 | Soil $\beta$ -1,4-Glucanase Activity Assay Kit          |