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# CheKine™ Micro Soli Dehydrogenase (S-DHA) Activity Assay Kit

Cat #: KTB4033

Size: 48 T/48 S 96 T/96 S

[ <u>;</u> ]	Micro Soli Dehydrogenase (S-DHA) Activity Assay Kit		
REF	Cat #: KTB4033	LOT	Lot #: Refer to product label
	Applicable samples: Soli		
X	Storage: Stored at 4°C for 6 months, protected from light		

## **Assay Principle**

The activity of soil dehydrogenase (S-DHA) can reflect the amount of active microorganisms in the soil system and its degradation activity of organic matter, and can be used as an indicator of the degradation performance of soil microorganisms. CheKine<sup>™</sup> Micro Soli Dehydrogenase (S-DHA) Activity Assay Kit can be used to detect biological samples such as soli. In this kit, The hydrogen acceptor 2,3,5-triphenyl tetrazolium chloride (TTC) generates red triphenyl formazone (TFF) after receiving hydrogen during cell respiration. TFF has a characteristic absorption peak at 485 nm, the S-DHA activity can quantified by measuring the absorbance at 485 nm.

## **Materials Supplied and Storage Conditions**

Kit componente	Si	Storage conditions		
Kit components	48 T	96 T	Storage conditions	
Reagent	Powder×1 vial	Powder×2 vials	4°C, protected from light	
Reagent II	25 mL	50 mL	4°C	

Note: Before formal testing, it is recommended to select 2-3 samples with large expected differences for pre-experiment.

## **Materials Required but Not Supplied**

- · Microplate reader or visible spectrophotometer capable of measuring absorbance at 485 nm
- 96-well quartz plate/lass plate plate (non-polystyrene) or microglass cuvette, precision pipettes, disposable pipette tips, 1.5 mL EP tube
- Water bath, cryogenic centrifuge, 30-50 mesh sieve
- Deionized water, ethyl acetate

## **Reagent Preparation**

**Working Reagent I:** Prepared before use. Add 12 ml Reagent II to each bottle to fully dissolve, The remaining reagent store at 4°C for 1 week, protected from light. If it turns red, it cannot be used.



**Reagent II:** Ready to use as supplied; Equilibrate to room temperature before use; Store at 4°C. **Note: Reagent I has certain irritation, so personal protection is recommended during use.** 

# **Sample Preparation**

Note: Note: It is recommended to use fresh soil samples.

Fresh soil samples naturally air dried or air dried in an oven at 37°C and sieved through 30-50 mesh sieve.

## **Assay Procedure**

1. Preheat the microplate reader or visible spectrophotometer for more than 30 min, and adjust the wavelength to 485 nm. Visible spectrophotometer was returned to zero with **ethyl acetate**.

2. Sample measurement. (The following operations are operated in the 1.5 mL EP tube)

Reagent	Test Tube	Control Tube
Sample (g)	0.1	0.1
Reagent ⊨ (µL)	200	0
Reagent II (µL)	0	200

Mix thoroughly, put it in an incubator at 37°C for dark reaction for 24 h, during which it oscillates for 5-6 times to make the soil sample fully contact with the reaction solution. After taking it out, take an ice bath for 5 min **immediately**.

Ethyl acetate (μL)	800	800

3. After uniformly mixing and shaking for 10 min, Centrifuge at 8,000 g for 10 min at 25°C, take 200  $\mu$ L of supernatant in a **96-well quartz plate/lass plate plate (non-polystyrene)** or micro glass cuvette to test the absorbance at 485 nm. The Test Well is marked as A<sub>Test</sub>, and the Control Well is marked as A<sub>control</sub>. Finally calculate  $\Delta$ A<sub>Test</sub>=A<sub>Test</sub>-A<sub>control</sub>.

Note: In order to guarantee the accuracy of experimental results, need to do a pre-experiment with 2-3 samples. If  $\Delta A$  is greater than 1.0, decrease the sample quantity appropriately. If  $\Delta A$  is less than 0.05, the sample volume can be appropriately increased, or the dark reaction time can be prolonged, and the calculation formula should be modified simultaneously.

# Data Analysis

Note: We provide you with calculation formulae, including the derivation process and final formula. The two are exactly equal. It is suggested that the concise calculation formula in bold is final formula.

Calculation of the S-DHA activity

Active unit definition: One unit of enzyme activity is defined as the amount of 1 g soli increases the absorbance of every 0.005 for per hour in the reaction system at 37°C.

S-DHA (U/g soli)=∆A÷0.005÷T÷W**=8.33×∆A÷W** 

T: reaction time, 1 d=24 h; W: sample weight, g.

# **Typical Data**

The following data are for reference only. And the experimenters need to test the samples according to their own experiments.





Figure 1. Determination of S-DHA activity in soli sample by this kit.

# **Recommended Products**

Catalog No.	Product Name
KTB1127	CheKine™ Micro Acetokinase (ACK) Activity Assay Kit
KTB1023	CheKine™ Micro Citrate Synthase (CS) Activity Assay Kit

# **Disclaimer**

The reagent is only used in the field of scientific research, not suitable for clinical diagnosis or other purposes. For your safety and health, please wear a lab coat and disposable gloves.

