CheKine™ Micro Soil Available Sulfur Content Assay Kit

Cat #: KTB4059 Size: 48 T/48 S 96 T/96 S

	Micro Soil Available Sulfur Content Assay Kit		
REF	**Cat #:** KTB4059	LOT	**Lot #:** Refer to product label
	Detection range: 5-160 mg/L		**Sensitivity:** 5 mg/L
	Applicable samples: Soli		
	Storage: Stored at 4℃ for 6 months, protected from light		

Assay Principle

Sulfur is an essential component of sulfur-containing amino acids and proteins, which directly participate in the growth and development of crops and affect the quality of crops. Soil sulfur mainly comes from parent material, irrigation water, dry and wet settlement in the atmosphere, and fertilization, etc., which plays an important role in agriculture, forestry and animal husbandry. Therefore, by determining the effective sulfur content of the soil and rational application of sulfur fertilizers plays a key role in improving the yield and quality of crops. CheKine™ Micro Soil Available Sulfur Content Assay Kit can be used to detect biological samples such as soli. In the kit, The sulfur proposed from the soil is basically in the form of SO_4^{2-}. In the acidic medium, SO_4^{2-} reacts with Ba^{2+} to form a $BaSO_4$ white precipitate with very little solubility. The barium sulfate turbidity method is used to determine the effective sulfur content in the soil.

Materials Supplied and Storage Conditions

Kit components	Size		Storage conditions
	48 T	96 T	
Extraction Buffer Ⅰ	80 mL	80 mL×2	4℃
Extraction Buffer Ⅱ	80 mL	80 mL×2	4℃
Reagent Ⅰ	0.5 mL	1 mL	4℃
Reagent Ⅱ	2.5 mL	5 mL	4℃
Reagent Ⅲ	5 mL	10 mL	4℃, protected from light
Reagent Ⅳ	2.5 mL	5 mL	4℃
Standard	Powder×2 vials	Powder×2 vials	4℃

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 440 nm
- 96-well plate or microglass cuvette, precision pipettes, disposable pipette tips, 1.5 mL EP tube
- Incubator, oscillator, centrifuge, 30-50 mesh sieve
- Deionized water

Reagent Preparation

Extraction Buffer Ⅰ: Ready to use as supplied; Equilibrate to room temperature before use; Store at 4℃. Used to extract acidic and neutral soils (pH<7.5).

Note: Extraction Buffer Ⅰ has certain irritation, so personal protection is recommended during use.

Extraction Buffer Ⅱ: Ready to use as supplied; Equilibrate to room temperature before use; Store at 4℃. Used to extract alkaline soil (pH≥7.5).

Reagent Ⅰ: Ready to use as supplied; Equilibrate to room temperature before use; Store at 4℃.

Reagent Ⅱ: Ready to use as supplied; Equilibrate to room temperature before use; Store at 4℃.

Reagent Ⅲ: Ready to use as supplied; Equilibrate to room temperature before use; Store at 4℃, protected from light.

Reagent Ⅳ: Ready to use as supplied; Equilibrate to room temperature before use; Store at 4℃.

Standard: Prepared before use. Add 1 mL Extraction Buffer corresponding to the pH of the sample for each bottle to fully dissolve, that is 3,200 mg/L sodium sulfur Standard. Equilibrate to room temperature before use; Store at 4℃ for 1 month. Using 3,200 mg/L sodium pyruvate Standard, prepare standard curve dilution as described in the table:

Num.	Standard Volume (μL)	Corresponding Extraction Buffer (μL)	Concentration (mg/L)
Std.1	50 μL of 3,200 mg/L Standard	950	160
Std.2	200 μL of Std.1 (160 mg/L)	200	80
Std.3	200 μL of Std.2 (80 mg/L)	200	40
Std.4	200 μL of Std.3 (40 mg/L)	200	20
Std.5	200 μL of Std.4 (20 mg/L)	200	10
Std.6	200 μL of Std.5 (10 mg/L)	200	5
Blank	0	200	0

Note: Always prepare fresh standards per use; Diluted Standard Solution is unstable and must be used within 4 h.

Sample Preparation

Note: It is recommended to use fresh soil samples. According to the acidity and alkalinity of the soil, select Extraction Buffer Ⅰ or Extraction Buffer Ⅱ.

Fresh soil samples naturally air dried or air dried in an oven at 37℃ and sieved through 30-50 mesh sieve. Weigh 0.2 g air-dried soil sample, add 1 mL Extraction Buffer, extract with vibration at 25℃ for 1 h. Centrifuge at 10,000 g for 10 min at 25℃. Use supernatant for assay.

Note: Gas may be generated when extracting with ExtractionBuffer Ⅰ. Be sure to tighten the cover when oscillating.

Assay Procedure

1. Preheat the microplate reader or visible spectrophotometer for more than 30 min, and adjust the wavelength to 440 nm. Visible spectrophotometer was returned to zero with deionized water.

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

Reagent	Test Tube (μL)	Standard Tube (μL)	Blank Tube (μL)
Sample	140	0	0
Standard	0	140	0
Corresponding Extraction Buffer	0	0	140
Reagent Ⅰ	7	7	7
Open the lid at 90℃ and cook for 5 min, remove it and cool it naturally. **The lid must be opened to allow Reagent Ⅰ to completely dissipate. If you use a water bath pot to suck it, you should prevent the water in the water bath pot from splashing into the EP tube, which will affect the detection data.**			
Reagent Ⅱ	35	35	35
Reagent Ⅲ	70	70	70
Reagent Ⅳ	35	35	35

3. Mix well, oscillating for 20 min at 25℃, add 200 μL to micro glass cuvette/96-well plate, detect the absorbance at 440 nm. The Blank Well is recorded as A_{Blank}, the Standard Well is marked as $A_{Standard}$, and the Test Well is marked as A_{Test}. Finally calculate $\Delta A_{Test}=A_{Test}-A_{Blank}$, $\Delta A_{Standard}=A_{Standard}-A_{Blank}$.

Note: (1) The Standard Well and Blank Well only need to be done once or twice. (2) In order to guarantee the accuracy of experimental results, need to do a pre-experiment with 2-3 samples. Testing should be done immediately after the oscillation is over. If the detection is not promptly found that precipitation has fallen to the bottom of the EP tube, it is recommended to oscillate again and wait for mixing and check. (3) If ΔA_{Test} is less than 0.01, increase the sample quantity appropriately. If ΔA_{Test} is greater than $\Delta A_{Standard}$ of 160 mg/L, the sample can be appropriately diluted with corresponding Extraction Buffer, the calculated result multiplied by the dilution factor, or decrease the sample quantity appropriately.

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.

1. Drawing of standard curve

With the concentration of the standard solution as the x-axis and the $\Delta A_{Standard}$ as the y-axis, draw the standard curve and obtain the standard equation. The determination of ΔA_{Test} is substituted into the equation to get x (mg/L).

2. Calculation of the soil available sulfur content

Soil available sulfur content (mg/g soli)=$V_{Total\ sample}\times x\div W\times 10^{-3}$**=x÷W**

$V_{Total\ sample}$: Added the corresponding Extraction Buffer volume, 1 mL; W: Sample weight, g; 10^{-3}: Unit conversion coefficient, 1 g=10^{-3} kg.

Typical Data

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

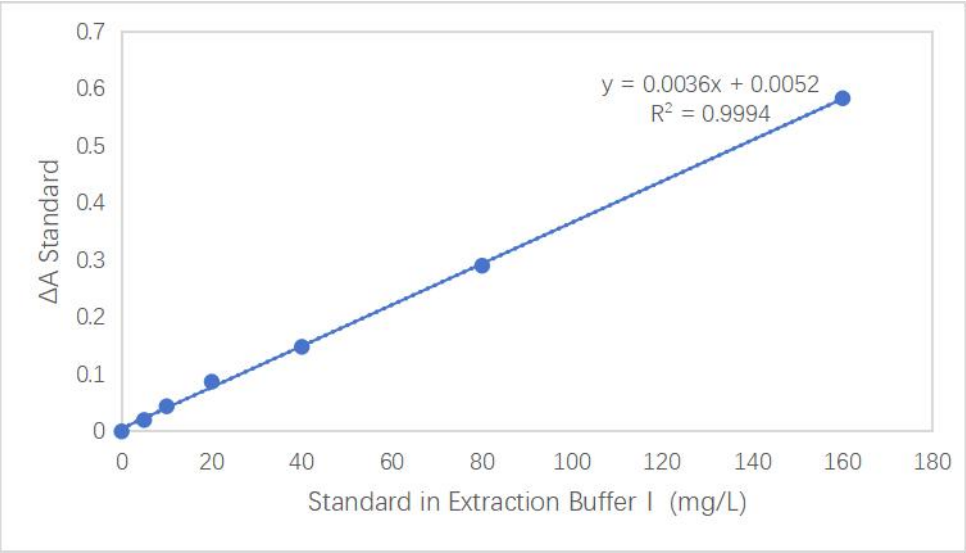

Figure 1. Standard curve of soil available sulfur content in Extraction Buffer Ⅰ.

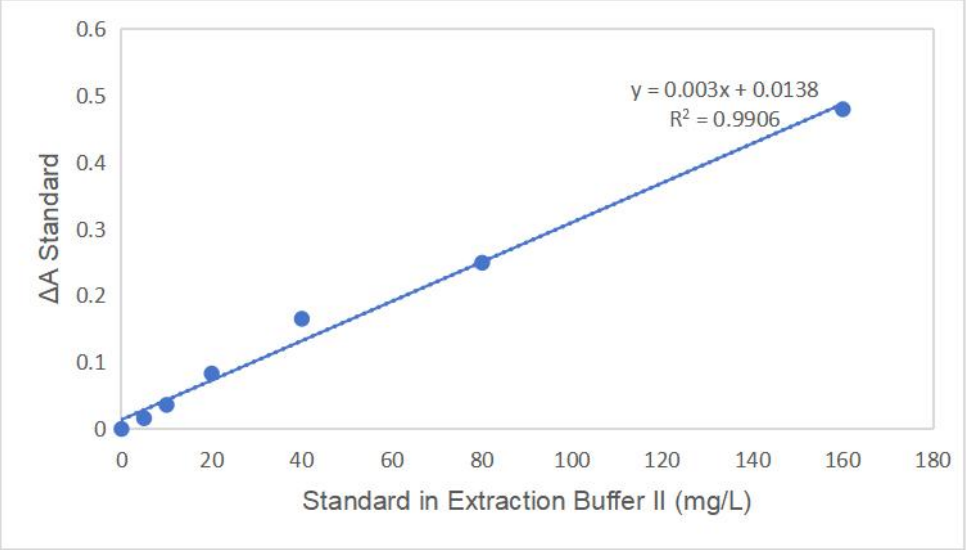

Figure 2. Standard curve of soil available sulfur content in Extraction Buffer Ⅱ.

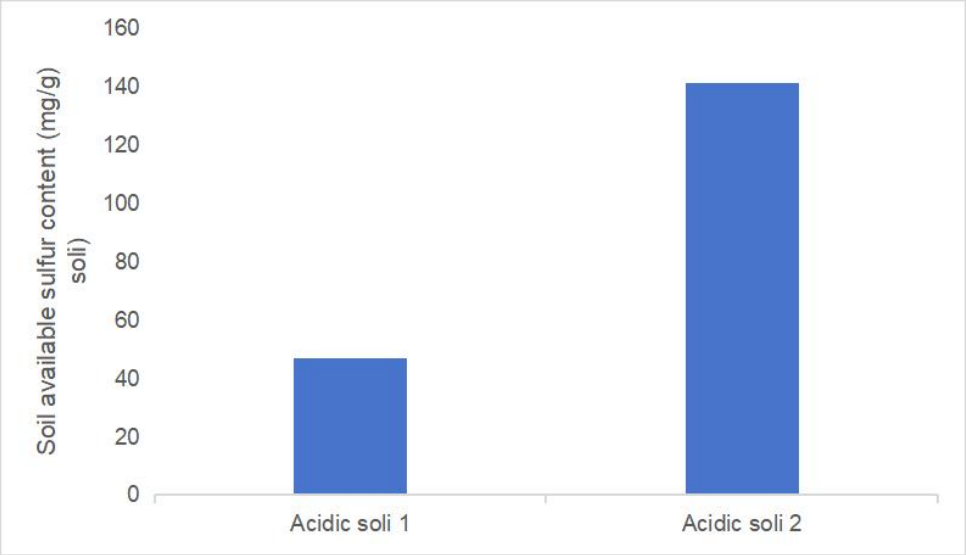

Figure 3. Determination of soil available sulfur content in acidic soli by this kit.

Recommended Products

Catalog No.	Product Name
KTB4012	CheKine™ Micro Soil Nitrate Nitrogen Assay Kit
KTB4014	CheKine™ Micro Acid Soil Available Phosphorous Assay Kit
KTB4041	CheKine™ Micro Soil Alkaline Phosphatase(S-AKP/ALP) 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.