SunLong Biotech Co.,LTD
Tel: 0086-571-56623320 Fax:0086-571-56623318
E-mail:sales@sunlongbiotech.com
www.sunlongbiotech.com

Cysteine (Cys) Content Assay Kit

Note: Take two or three different samples for prediction before test.

Detection equipment: Spectrophotometer

Cat No: AK0582

Size: 50T/48S

Components:

Extract solution: Liquid 25 mL×1, store at 4℃ .

Reagent I: Liquid 45 mL×1, store at 4℃ .

Reagent II: Powder×2, store at 4℃ . The day before it is to be used, add 5 mL of distilled water to Reagent II ， fully stir for dissolving, then add 1.25 mL phosphoric acid, mix thoroughly. Incubate at boi ling water for 2 hours and add 20 mL distilled water after cooling. The reagent can be stored for two weeks at 4℃ . **Standard:** Powder×1, 10 mg of cysteine, store at 4℃ . Dissolve in 4.13 mL of distilled water prepare as 20 μmol/mL standard solution before use. The solution could be stored at 4℃ for 4 weeks.

Description:

Protein contains three kinds of sulfur-containing amino acids: methionine, cystine and cysteine (Cys). Cys is the only sulfur-containing amino acid containing sulfhydryl groups, which derived from methionine and could be transformed with cystine. Cys participates in the formation of protein disulfide bonds, which usually is a component of active centers of protein and can provide mercapto groups for other physiological and biochemical reactions. Besides, a large amount of Cys accumulates on skin and mucosal surfaces to maintain elasticity and texture of skin and keep the activity of thiolase in the process of keratin production. It has the functions of whitening, detoxification, inflammation improvement and so on.

The phosphotungstic acid is reduced to tungsten blue by Cys, and the tungsten blue has absorption peak at 600 nm. In this kit, the content of Cys is calculated by measuring the absorbance at 600 nm.

Required but not provided:

Spectrophotometer, refrigerated centrifuge, transferpettor, water bath, 1 mL glass cuvette, mortar/ homogenizer, phosphoric acid and distilled water.

Protocol:

I. Sample preparation

1. Liquid sample:

Add 0.3 mL of Extract solution to 0.2 mL of liquid sample, mix thoroughly. Centrifuge at 11000 rpm for 10 minutes at 4℃ to remove insoluble materials, and take the supernatant on ice for testing.

2. Tissue sample:

Add 0.5 mL of Extract solution to 0.2 g of tissue, mix thoroughly on ice. Centrifuge at 11000 rpm for 10 minutes at 4℃ to remove insoluble materials, and take the supernatant on ice for testing.

II. Determination procedure

1. Preheat spectrophotometer for 30 minutes, adjust wavelength to 600 nm, set zero with distilled water.

2. Standard: Dilute the standard solution with 4.13 mL of distilled water to generate a 20 μmol/mL standard, then dilute to 2, 1, 0.5, 0.25, 0.125， 0.0625 μmol/mL standard with distilled water.

3. Add reagents as the following table.

Reagent (μL)	Test tube (T)	Standard tube (S)	Blank tube (B)
Sample	200	-	-
Standard	-	200	-
Distilled water	-	-	200
Reagent I	500	500	500
Reagent II	300	300	300

Mix and keep at room temperature for 15 minutes and detect the absorbance at 600 nm.

III. Calculation

1. Standard curve.

The concentration of standard solution as x-axis, $\Delta A(A_S-A_B)$ as y-axis, obtain the equation $y=kx+b$. Take (A_T-A_B) to the equation to acquire x value.

2. Calculate

1) Liquid sample

Cys (μmol/mL) = $x\times V_{ST}\div V=2.5x$

2) Sample weight

Cys (μmol/g FW) = $x\times V_{ST}\div W=0.5x\div W$

V_S: Liquid volume, 0.2 mL;

V_{ST}: Extract solution volume, 0.5 mL;

W: Sample weight, g.

Note:

If the absorbance value exceeds the linear range, the sample size can be increased or diluted before the determination.

Recent Product Citations:

[1] Huang Q, Wang M, Xia Z. The SULTR gene family in maize (Zea mays L.): gene cloning and expression analyses under sulfate starvation and abiotic stress[J]. Journal of plant physiology, 2018, 220:

24-33.

[2] Yansha Han, Mengyang Wu, Lihong Hao, et al. Sulfur dioxide derivatives alleviate cadmium toxicity by enhancing antioxidant defence and reducing Cd^{2+} uptake and translocation in foxtail millet seedlings. Ecotoxicology and Environmental Safety. August 2018;(IF4.527)

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Related Products:

AK0423/AK0422	Glutamic-pyruvic Transaminase(GPT) Activity Assay Kit
AK0421/AK0420	Glutamic- oxalacetic Transaminase(GOT) Activity Assay Kit
AK0564/AK0563	Proline(PRO) Content Assay Kit
AK0419/AK0418	Amino Acid(AA) Content Assay Kit
AK0417/AK0416	Glutamic Acid(Glu) Content Assay Kit

Technical Specification:

Detection Limit: 0.0053 μmol/mL

Linear range: 0.03125-3 μmol/mL