Cysteine (Cys) Content Assay Kit

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

Detection equipment: Spectrophotometer

Cat No: BC0180

Size: 50T/48S

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
Extract solution	Liquid 25 mL×1	2-8℃
Reagent I	Liquid 45 mL×1	2-8℃
Reagent II	Powder ×2	2-8℃
Standard	Powder ×1	2-8℃

Solution Preparation:

1. Reagent II: The day before it is to be used, add 5 mL of distilled water to one Reagent II, fully stir for dissolving, then add 1.25 mL phosphoric acid, mix thoroughly. Incubate at boiling water for 2 hours (Wrap the sealing film to prevent bursting) and add 20 mL distilled water after cooling. The reagent can be stored for two weeks at 2-8℃. (One bottle of reagent is enough for 50T, another is to prolong the stable storage time of the reagent)
2. Standard: 10 mg of cysteine. Dissolve in 4.13 mL of distilled water to prepare as 20 μmol/mL standard solution before use. The solution could be stored at 2-8℃ 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, 1 mL glass cuvette, mortar/ homogenizer, concentrated phosphoric acid (85%, AR) 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 20 μmol/mL standard with distilled water to 2, 1, 0.5, 0.25, 0.125， 0.0625 μmol/mL standard.
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. Note as A_T, A_S, A_B. calculate $\Delta A_T = A_T - A_B$, $\Delta A_S = A_S - A_B$. The blank and standard curve only need to be measured 1-2 times.

III. Calculation

1. Standard curve.

The concentration of standard solution as x-axis, ΔA_S as y-axis, obtain the equation y=kx+b. Take ΔA_T to the equation to acquire x value.

2. Calculate

1) Liquid sample

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

2) Sample weight

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

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] Zheng Q, Liu H, Zhang H, Han Y, Yuan J, Wang T, Gao Y, Li Z. Ameliorating Mitochondrial Dysfunction of Neurons by Biomimetic Targeting Nanoparticles Mediated Mitochondrial Biogenesis to Boost the Therapy of Parkinson's Disease. Adv Sci (Weinh). 2023 Aug;10(22): e2300758. doi: 10.1002/advs.202300758. Epub 2023 May 18. PMID: 37202595; PMCID: PMC10401119.

[2] Meng Y, Cui Y, Peng F, Guo L, Cui R, Xu N, Huang H, Han M, Fan Y, Zhang M, Sun Y, Wang L, Yang Z, Liu M, Chen W, Ni K, Wang D, Zhao L, Lu X, Chen X, Wang J, Wang S, Ye W. GhCYS2 governs the tolerance against cadmium stress by regulating cell viability and photosynthesis in cotton. Ecotoxicol Environ Saf. 2023 Sep 15; 263:115386. doi: 10.1016/j.ecoenv.2023.115386. Epub 2023 Aug 18. PMID: 37598545.

[3] Yang Q, Luo M, Zhou Q, Zhao Y, Chen J, Ji S. Insights into the loss of glucoraphanin in post-harvested broccoli--Possible involvement of the declined supply capacity of sulfur donor. Plant Sci. 2023 Mar; 328:111580. doi: 10.1016/j.plantsci.2022.111580. Epub 2022 Dec 30. PMID: 36587585.

[4] Zheng L, Zhou P, Pan Y, Li B, Shen R, Lan P. Proteomic profile of the germinating seeds reveals enhanced seedling growth in Arabidopsis rpp1a mutant. Plant Mol Biol. 2023 Oct;113(1-3):105-120. doi: 10.1007/s11103-023-01378-w. Epub 2023 Oct 7. PMID: 37804450.

[5] Zhang J, Zhang L, Zhou Y, Li K, Dai X, Bian L. The fluorescence regulation of a tri-functional oligonucleotide probe HEX-OND in detecting Pb (II), cysteine, and K(I) based on two G-quadruplex forms. Anal Bioanal Chem. 2023 Jun;415(14):2763-2774. doi: 10.1007/s00216-023-04681-z. Epub 2023 Apr 27. PMID: 37103561.

Related Products:

BC1550/BC1555	Glutamic-pyruvic Transaminase (GPT) Activity Assay Kit
BC1560/BC1565	Glutamic-oxalacetic Transaminase (GOT) Activity Assay Kit
BC0290/BC0295	Proline (Pro) Content Assay Kit
BC1570/BC1575	Amino Acid (AA) Content Assay Kit

Technical Specification:

Detection Limit: 0.0053 μmol/mL

Linear range: 0.03125-3 μmol/mL