Tannin content Assay Kit

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

Operation Equipment: Spectrophotometer/microplate reader

Catalog Number: BC1395

Size:100T/96S

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	Storage
Extract Solution	Solution 75 mL×2	2-8℃
Reagent I	Powder×1	RT
Standard	Powder×1	2-8℃

Solution preparation:

Standard : 10 mg tannic acid. Add 1.175 mL of Extract solution to dissolve it into 5000 nmol/mL standard solution before use. It could be stored at 2-8℃ for two weeks.

Product Description

Tannins are also called plant polyphenols. It is a kind of polyphenol compound widely existing in plants. Tannins can be used as potential biomarkers. The ability to bind to proteins is also known as convergence or astringency. Its convergence is the basis of many physiological activities, such as hemostasis, anti-tumor, anti-aging and other physiological activities. It is also one of the factors that affect the taste of the product.

According to the spectral characteristics, tannins have strong UV absorption at 275 nm. Activated carbon can adsorb tannin specifically. The tannin content can be detected by this property.

Reagents and Equipment Required but Not Provided.

Ultraviolet spectrophotometer/microplate reader, micro quartz cuvette/96-well UV plate, centrifuge, water bath, adjustable pipette, 30-50 mesh screen, distilled water.

Procedure

I. Sample processing :

Dry the sample to constant weight and crush it. Over 30-50 mesh screen. Add 1 mL of Extract solution to 0.05 g sample weight. Sealing film to prevent liquid splashing. Extraction in 70℃ water bath for 30 min. Continuous oscillation. Centrifuge at 12000 rpm for 10 min at 25℃. Take the supernatant, and use the Extract solution to volume the supernatant to 1 mL for test.

II. Determination Procedure

1. Preheat the spectrophotometer/microplate reader for more than 30 minutes, adjust the wavelength to 275

nm and set spectrophotometer counter to zero with **extract solution**.

2. Standard working solution: dilute the 5000 nmol/mL standard solution to 25、12.5、6.25、3.125、1.5625、0.78125nmol/mL standard solution with extract solution.

3. The dilution of standard solution can refer to the following table:

Number	Pre dilution concentration (mg/mL)	Standard liquid volume (μL)	Volume of standard dilution solution (μL)	Diluted concentration (mg/mL)
1	5000	100	900	500
2	500	100	1900	25
3	25	1000	1000	12.5
4	12.5	1000	1000	6.25
5	6.25	1000	1000	3.125
6	3.125	1000	1000	1.5625
7	1.5625	1000	1000	0.78125

4. Sampling table :

Reagent	Test tube (A_T)	Control tube (A_C)	Standard tube (A_S)	Blank tube (A_B)
Reagent I (mg)	about 5-7 mg	-	-	about 5-7 mg
Extract solution (mL)	-	-	-	0.5
Standard (mL)	-	-	0.5	-
Sample (mL)	0.5	0.5	-	-
Mix thoroughly. Shock 5 min. Centrifuge at 13000 g for 20 min. Take 200 μL of supernatant to determine the absorbance at 275 nm. Record as A_T, A_C, A_S, A_B, and calculate $\Delta A_S = A_S - A_B$, $\Delta A_T = A_T - A_C$. Each test tube should be provided with one contrast tube. Standard curve and blank tube only need to be measured once or twice.				

III. Calculation of Tannin content:

IV. Standard curve

Taking the concentration of each standard solution as the y-axis and its corresponding ΔA_S as the x-axis, draw a standard curve to get the standard equation y = kx + b, and bring ΔA_T into the equation to get y (nmol/mL).

1. Calculate

1) Calculate by protein concentration

Tannin content (nmol/mg prot)=$y \times V_E \div (V_E \times Cpr) = y \div Cpr$

2) Calculate by sample weight

Tannin content (nmol/g fresh weight)=$y \times V_E \div W = y \div W$

V_E: Extract solution volume, 2 mL;

W: Sample weight, g.

Cpr: Sample protein concentration, mg/mL; (The protein concentration needs to be re extracted by

PBS and then determined.)

Note:

1. If the absorbance value exceeds the linear range, the sample size can be increased or diluted before proceeding with the measurement. Pay attention to synchronously modifying the calculation formula.
2. Extraction contains ingredients that denature proteins, and protein content needs additional measurement if tannin content would be calculated by protein concentration.

Related Products:

BC1300/BC1305	Ceruloplasmin (CP) Assay Kit
BC1310/BC1315	Total antioxidant capacity (T-AOC) Assay Kit
BC1360/BC1365	Uric acid (UA) Assay Kit