Plant Total Phenol (TP)Assay Kit

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

Operation Equipment: Spectrophotometer

Cat No: BC1340

Size:50T/24S

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 60 mL×1 (Required but not provided)	RT
Reagent I	Solution 20 mL×1	2-8℃
Reagent II	Solution 25 mL×1	2-8℃
Standard	Powder×1	2-8℃

Solution preparation:

1. **Extract solution:** 60% alcohol (V: V), self-provided reagent, store at room temperature.
2. **Standard:** 5 mg of gallic acid. Before use, add 1 mL of distilled water, heat it at 50℃ and dissolve it to prepare 5mg/mL standard solution. It could be stored at 2-8℃ for two weeks.

Description:

Plant phenols have the function of scavenging free radicals, anti-oxidation and anti-aging. It is widely used in cosmetics, food, medicine and other fields because of its high nutritional value and health care function.

In alkaline conditions, phenolic substance reduce tungsten-molybdic acid to form blue compounds which has an absorption peak at 760 nm. The total phenol content of the sample is obtained by measuring the absorbance at 760nm.

Technical Index:

Minimum detection limit: 0.0012 mg/mL

linear range: 0.0024-0.156 mg/mL

Required but not provided:

Spectrophotometer, 1 mL glass cuvette, balance, oven, crusher/mortar, 30-50 sieve, ultrasonic cleaner, 60% alcohol, centrifuge, distilled water.

Procedure:

I. Total phenol extraction:

Dry the sample to constant weight, smash. After screening with the 30-50 mesh sieve, add 2.5 mL of

Extract solution to 0.1 g of tissue and extract by ultrasonic cleaner (power 300W, 60℃ for 30 min). Centrifuge at 12000 rpm for 10 min at 25℃.

II. Procedure:

1. Preheat spectrophotometer for 30 min, adjust wavelength to 760 nm, set zero with distilled water.

2. Dilution of standard solution: dilute the standard with distilled water to 0.15625, 0.078125, 0.039, 0.02, 0.01, 0.005, 0.0025mg/mL standard solution.

3. Standard solution dilution 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	5	125	875	0.625
2	0.625	250	750	0.15625
3	0.15625	200	200	0.078125
4	0.078125	200	200	0.039
5	0.039	200	200	0.02
6	0.02	200	200	0.01
7	0.01	200	200	0.005
8	0.005	200	200	0.0025

Note: Each standard tube in the following experiment requires 50 μL of standard solution (be careful not to directly test the absorbance of the standard solution in this step).

4. Operation table.

Reagent name (μL)	Control tube (A_C)	Test tube (A_T)	Standard tube (A_S)	Blank tube (A_B)
Sample	50	50	-	-
Standard	-	-	50	
Distilled water	-	-	-	50
Reagent I	-	250	250	250
Mix thoroughly，incubate at room temperature for 2 min.				
Reagent II	250	250	250	250
Distilled water	700	450	450	450
Mix thoroughly, incubate at room temperature for 10 min. Detect the absorbance of 760 nm in 1mL glass cuvette, and record it as A_C, A_T, A_S and 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.

1. Standard curve drawn.

Taking the concentration of each standard solution as the x-axis and its corresponding ΔA_S as

the y-

axis, draw a standard curve to get the standard equation y = kx + b, and bring ΔA_T into the equation to get x (mg/mL).

2. Calculation of plant total phenol

a. Sample weight

Total phenol (mg/g, weight) = $x \times V_E \div W = 2.5x \div W$

b. Protein concentration

Total phenol (mg/mg prot) = $x \times V_E \div (Cpr \times V_E) = x \div Cpr$

V_E: Extract solution volume；2.5 mL；

W: Sample weight, g；

Cpr: Protein concentration，mg/mL.

Note:

1. If the measured absorbance value exceeds the absorbance value in the linear range, you can increase the sample volume or dilute the sample before performing the measurement.
2. Reagent I have a certain irritation to the skin, please take precautions during operation.

Examples:

1. Add 0.1g treated yellow flower to 2.5mL extract solution, after treating sample follow the determination procedure to operate, and calculate: $\Delta A = A_T - A_B = 0.679 - 0.000 = 0.679$, standard curve: y=5.5245x+0.0102, calculate x=0.1211, according with mass of sample to calculate: Total phenol (mg/g weight) $= 2.5x \div W = 2.5 \times 0.1211 \div 0.1 = 3.0275$ mg/g weight.

Recent Product citations：

[1] Wang Y, Zhang Y, Yuan Y, Zhao Y, Nie J, Nan T, Huang L, Yang J. Nutrient content prediction and geographical origin identification of red raspberry fruits by combining hyperspectral imaging with chemometrics. Front Nutr. 2022 Oct 17;9:980095. doi: 10.3389/fnut.2022.980095. PMID: 36386936; PMCID: PMC9642070.

[2] Zhang C, Xin Y, Wang Z, Qin L, Cao L, Li H, Ma X, Yin J, Zhao Z, Liu P, Tang J, Dong C. Melatonin-induced myeloblastosis viral oncogene homologs alleviate fresh-cut lotus root browning during storage by attenuating flavonoid biosynthesis and reactive oxygen species. J Sci Food Agric. 2023 Aug 30;103(11):5452-5461. doi: 10.1002/jsfa.12619. Epub 2023 May 1. PMID: 37046375.

[3] Wang C, Chen Y, Chen S, Min Y, Tang Y, Ma X, Li H, Li J, Liu Z. Spraying chitosan on cassava roots reduces postharvest deterioration by promoting wound healing and inducing disease resistance. Carbohydr Polym. 2023 Oct 15;318:121133. doi: 10.1016/j.carbpol.2023.121133. Epub 2023 Jun 19. PMID: 37479443.

[4] Liang Z, Luo Z, Li W, Yang M, Wang L, Lin X, Li L. Elevated CO2 Enhanced the Antioxidant Activity and Downregulated Cell Wall Metabolism of Wolfberry *(Lycium barbarum L.*).

Antioxidants

(Basel). 2021 Dec 22;11(1):16. doi: 10.3390/antiox11010016. PMID: 35052519; PMCID: PMC8773196.

[5] Ling S, Qiu H, Xu J, Gu Y, Yu J, Wang W, Liu J, Zeng X. Volatile Dimethyl Disulfide from Guava Plants Regulate Developmental Performance of Asian Citrus Psyllid through Activation of Defense Responses in Neighboring Orange Plants. Int J Mol Sci. 2022 Sep 7;23(18):10271. doi: 10.3390/ijms231810271. PMID: 36142192; PMCID: PMC9499464.

Reference:

[1] Maryam Akhbari, Sepideh Hamedi, Zahra-sadat Aghamiri. Optimization of total phenol and anthocyanin extraction from the peels of eggplant (Solanum melongena L.) and biological activity of the extracts [J]. Journal of Food Measurement and Characterization, 2019, 13(4): 29-37.

Related Products:

BC1300/BC1305	Ceruloplasmin (CP) Assay Kit
BC1310/BC1315	Total antioxidant capacity (T-AOC) Assay Kit
BC1370/BC1375	Total Sulfhydryl Assay Kit

Tel: 86-010-50973105 https://www.solarbio.net E-mail: info@solarbio.com