

Plant Proanthocyanidins Assay Kit

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

Operation Equipment: Spectrophotometer

Catalog Number: BC1350

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 80 mL×1	2-8°C
Reagent I	Solution 25 mL×1	2-8°C
Reagent II	Powder×1	2-8°C
Standard	Powder×1	2-8°C

Solution preparation:

Reagent II: Dissolve with 25 mL of extract solution before use, the configured reagent II can be stored at 2-8°C for 1 month.

Standard: 10 mg of Proanthocyanidins. Add 1mL of extraction solution before use, fully dissolve to obtain 10 mg/mL standard solution, and store at 2-8°C for two weeks;

Working Solution: Mix Reagent I and II in a 1:1 ratio according to the dosage before use, and prepare them as needed. Mix as much as you need.

Product Description:

Oligomeric proantho cyanidins (OPC) is a polyphenol compound of a flavanol monomer and polymer, which exists widely in various organs of plants. It has strong oxidation resistance and the ability of scavenging free radical. It used widely in pharmaceutical, food, cosmetics, health care products and so on.

Under acidic conditions, resorcinol and pyrogallol in A ring of plant OPC can react with vanillin to form colored compound, which can be detected by colorimetric assay at 500 nm and calculate the content of OPC.

Technical Index:

Minimum detection limit: 0.0622 mg/mL

linear range: 0.078-5 mg/mL

Reagents and Equipment Required but Not Provided:

Spectrophotometer, 1 mL glass cuvette, balance, centrifuge, crusher, ultrasonic cleaner, distilled water.

Procedure:

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I. Sample Preparation:

Dry the sample to constant weight, crush and filtrate with 30-50 mesh sifter, add 1 mL of extract solution to 0.1 g of sample, ultrasonic (power 300W) for 30 min, centrifuge at 12000 rpm and 25°C for 10 min. Add extract solution to supernatant, make final volume to 1 mL for test.

II. Determination Procedure

1. Preheat spectrophotometer for 30 min, adjust the wavelength to 500 nm, set the counter to zero with distilled water.
2. Preparation of standard solution: Dilute 10mg/mL standard solution with **Extract solution** to 4, 3, 2.5, 1.25, 0.625, 0.3125, 0.15625 mg/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	10	200	300	4
2	10	150	350	3
3	10	125	375	2.5
4	2.5	500	500	1.25
5	1.25	500	500	0.625
6	0.625	500	500	0.3125
7	0.3125	500	500	0.15625

Note: Each standard tube in the following experiment requires 200 μL of standard solution (be careful not to directly test the absorbance of the standard solution in this step).

4. Operation table

Reagent Name	Control Tube (A _C)	Test Tube (A _T)	Standard Tube (A _S)	Blank Tube (A _B)
Sample	200	200	-	-
Standard	-	-	200	-
Working Solution	-	800	800	800
H ₂ O	800	-	-	200

Mix thoroughly, 30°C water bath for 30 min, take 1 mL to 1 mL glass cuvette, detect absorbance at 500 nm, $\Delta A(\text{Standard}) = \Delta A(S) = A_S - A_B$, $\Delta A(\text{Test}) = \Delta A(T) = A_T - A_C$. The standard curve and blank tube only need to be measured 1-2 times.

III. Calculation:

1. Make standard curve:

According to concentration of standard solution and absorbance to create the standard curve, take standard solution as X-axis, $\Delta A(T)$ as Y-axis. Take $\Delta A(S)$ into the equation to obtain x (mg/mL).

2. Calculation of OPC

The determination of ΔA is introduced into the equation and x(mg/mL) is obtained.

A. Calculated based on sample weight:

$$\text{OPC (mg/g weight)} = x \times V_e \div W = x \div W$$

B. Calculated based on sample protein concentration:

$$\text{OPC (mg/mg prot)} = x \times V_e \div (C_{pr} \times V_e) = x \div C_{pr}$$

Cpr: Protein concentration, mg/mL;

W: Sample weight, g;

Ve: Extraction volume, 1 mL;

Note:

If the measured absorbance value exceeds the linear range, the sample size can be increased or the sample can be diluted before measurement, and attention should be paid to synchronously modifying the calculation formula.

Recent Product Citations:

[1] Hong Y, Lv Y, Zhang J, Ahmad N, Li X, Yao N, Liu X, Li H. The safflower MBW complex regulates HYSA accumulation through degradation by the E3 ligase CtBB1. *J Integr Plant Biol.* 2023 May;65(5):1277-1296. doi: 10.1111/jipb.13444. Epub 2023 Mar 27. PMID: 36598461.

[2] Sun H, Cui H, Zhang J, Kang J, Wang Z, Li M, Yi F, Yang Q, Long R. Gibberellins Inhibit Flavonoid Biosynthesis and Promote Nitrogen Metabolism in *Medicago truncatula*. *Int J Mol Sci.* 2021 Aug 27;22(17):9291. doi: 10.3390/ijms22179291. PMID: 34502200; PMCID: PMC8431309.

[3] Jiang G, Wang S, Xie J, Tan P, Han L. Discontinuous low temperature stress and plant growth regulators during the germination period promote roots growth in alfalfa (*Medicago sativa* L.). *Plant Physiol Biochem.* 2023 Apr;197:107624. doi: 10.1016/j.plaphy.2023.03.001. Epub 2023 Mar 16. PMID: 36948023.

Reference:

[1] Wang AG, Luo GH, Quantitative relation between the reaction of Hydroxylamine and Superoxide Anion Radicals in plants. *Plant Physiology Communication.* 1990(6): 55-57.

Related Products:

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