

# Plant Proanthocyanidins Assay Kit

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

**Operation Equipment:** Spectrophotometer/microplate Reader

**Catalog Number:** BC1355

**Size:**100T/48S

## Components:

Extract solution: Liquid 80 mL×1 bottle, storage at 4°C ;

Reagent I: Liquid 8 mL×1 bottle, storage at 4°C;

Reagent II: powder×1 bottle, storage at 4°C and protected from light, dissolve with 8 mL of extract solution before use;

Working solution: According to the usage, mix Reagent I and II as the ratio of 1:1 before use.

Standard: powder×1 bottle, 10 mg of Proanthocyanidins;

## Product Description:

Oligomeric proanthocyanidins (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.

## Reagents and Equipment Required but Not Provided:

Spectrophotometer/microplate reader, micro glass cuvette/96 well flat-bottom plate, the room temperature centrifuge, balance, distilled water.

## Procedure:

### I. Sample preparation:

1. Dry the sample to constant weight, crush and filtrate with 40 mesh sifter, add 1 mL of extract solution to 0.1 g of sample, ultrasonic (power 300W, work time 5s, interval 8s) 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/microplate reader for 30 min, adjust the wavelength to 500 nm, set the counter to zero with distilled water.

2. Dilute Proanthocyanidins standard with extract solution to 10 mg/mL for use. Dilute with extract solution to final concentrate 5, 2.5, 1.25, 0.625, 0.3125, 0.156, 0.078, 0.039, 0.02, 0.01 mg/mL.

3. Add the following reagents:

	Blank tube A2	Standard tube A1	Test tube A3	Control tube A4
Sample (μL)			40	40
Standard (mg/mL)		40		
Working solution (μL)	160	160	160	
H <sub>2</sub> O (μL)	40			160

Mix thoroughly, 30°C water bath for 30 min, take 200  $\mu$ L to micro cuvette/96 well flat-bottom plate, detect absorbance at 500 nm,  $\Delta A(\text{Standard}) = \Delta A(S) = A1 - A2$ ,  $\Delta A(\text{Test}) = \Delta A(T) = A3 - A4$ . Blank tube just test once or twice.

### III. Calculation:

1. Make standard curve:

Standard solution concentrate as the abscissa,  $\Delta A(S)$  as ordinate, establish the standard curve, get formula  $y = kx + b$ .

2. Calculation of OPC

The determination of  $\Delta A$  is introduced into the equation and  $x(\text{mg/mL})$  is obtained.

A. Sample weight:

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

B. Sample Protein concentration:

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

$C_{pr}$ : Protein concentration, mg/mL;

W: Sample weight. g;

$V_e$ : Extraction volume, 1 mL;

#### Note:

1. Use Reagent II as soon as possible, storage at 4°C no more than one month;
2. Add sample or dilute sample if the change of absorbance out range of 0.006-1.2, dilute times in the calculation formula also need to change.

#### Recent Product citations:

[1] Li Y, Cui W, Qi X, et al. MicroRNA858 negatively regulates anthocyanin biosynthesis by repressing AaMYBC1 expression in kiwifruit (*Actinidia arguta*)[J]. Plant Science, 2020: 110476.

#### Related products:

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