



# Plant flavonoids detection kit

(Cat/No.:BC161 Size:50T/48S)

## 1. Metermination significance

Flavonoids are a class of polybenzene compounds, which belong to plant secondary metabolites, which have anti-inflammatory, antibacterial, lower blood lipid, remove hydroxyl free radicals in the body, cancer prevention and other effects

## 2. Determination principle (colorimetric method)

In the alkaline nitrite solution, flavonoids and aluminum ions form a red complex with a characteristic absorption peak at 502nm. The flavonoid content of the samples can be calculated by measuring the light absorption value of the sample extract at 502nm.

## 3. Instrument and Equipment (self-provided)

Oven, grinder, mortar, sieve, 60% ethanol, centrifuge, visible spectrophotometer, 60 °C shock incubator.

## 4. Reagent compositio

Extract: 60% ethanol, self-prepared.

**Reagent 1:**liquid 2 mL , 4 °C of storage.

**Reagent 2:**liquid 2 mL, 4 °C.

**Reagent 3:**liquid 25 mL , 4 °C.

**Reagent 4:**1mg ×1 standard powder, add 1mL 60% ethanol, shake at 60 °C for 10 minutes to dissolve it completely, mixed with 1mg / mL standard application solution.

## 5. operating steps

### 1) Flavonoid extraction:

Dry sample: dry the sample until constant weight, crush, screen after 40 mesh, weigh about 0.02g, add 2mL extract, 60 °C shock extract for 2 hours, 10000g, centrifuge at 10min at room temperature, remove the supernatant, to be measured.

Fresh sample: after washing the plant with normal saline, dry the surface moisture, cut, grind liquid

nitrogen into powder, weigh about 0.05g, add 2 mL, extract (low content can reduce the amount of extract), 60 °C shock extraction for 2 hours, 10000g, centrifuge at room temperature for 10min, take the supernatant, to be measured.

### 2) Operating table:

|  | Blank tube | Determi<br>ned<br>tube | standard<br>tube |
|--|------------|------------------------|------------------|
| distilled water (mL)                               | 0.50       |                        |                  |
| Sample solution to be tested (mL)                  |            | 0.50                   |                  |
| Different concentrations of standard solution (mL) |            |                        | 0.50             |



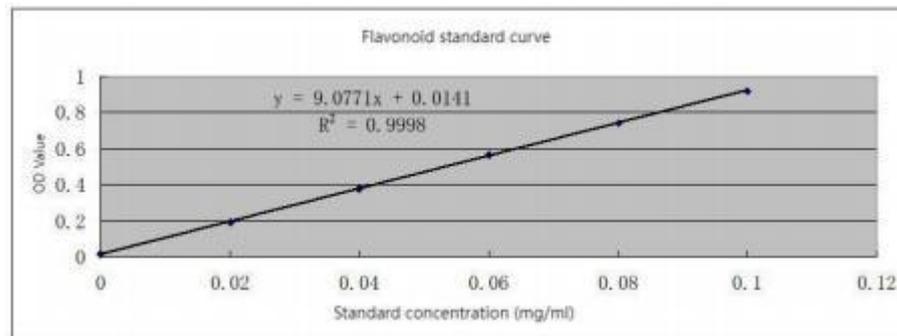
|  |      |      |      |
|--|------|------|------|
| Reagent 1 (mL)   | 0.03 | 0.03 | 0.03 |
| Mix well and stand at room temperature for 5min  |      |      |      |
| Reagent 2 (mL)   | 0.03 | 0.03 | 0.03 |
| Mix well and stand at room temperature for 5min  |      |      |      |
| Reagent 3 (mL)   | 0.40 | 0.40 | 0.40 |
| distilled water (mL)   | 0.04 | 0.04 | 0.04 |
| Mix well, stand at room temperature for 15 min, adjust the distilled water to zero, with the wavelength of 502nm, and determine the absorbance value of each tube ( $\Delta A_{502} = A_{\text{determine}} - A_{\text{blank space}}$ ) . |      |      |      |

**Note:** It is better to take 1-2 samples before the formal experiment,  $\Delta A_{\text{determine}}$  If greater than 1, dilute the supernatant of the sample before measuring.

## 6. Calculation formula

Making standard curve: 1 mg/mL standard solution was diluted with 60% ethanol into 0.1 mg/mL, 0.08 mg/mL, 0.06 mg/mL, 0.04 mg/mL, 0.04 mg/mL and 0.02mg / mL according to the standard tube and blank tube in the operation table, and the OD value was obtained and the standard curve was drawn as follows:

|  |       |       |       |       |       |       |
|--|-------|-------|-------|-------|-------|-------|
| Standard liquid concentration ( $\mu\text{g/mL}$ ) | 0     | 20    | 40    | 60    | 80    | 100   |
| OD price   | 0.015 | 0.192 | 0.375 | 0.566 | 0.743 | 0.917 |



Then the calculation formula is:

Flavonoid content (mg / g) =  $(\Delta A_{\text{determine}} - 0.0141) \times 9.0771 \times \text{Total volume of the extract (2 mL)} / \text{Sample mass (g)}$

## 7. Technical parameters

- 1, the detection limit of the kit is 1  $\mu\text{g/mL}$ , if  $\Delta A_{\text{determine}}$  the value is greater than 1, the sample supernatant shall be measured after appropriate dilution;
2. The recovery rate of this kit can reach 95% -100%, with good stability and high accuracy;
3. The low temperature of standard configuration will make it dissolve slowly. When  $60^\circ\text{C}$  oscillation solves, it should be sealed to prevent liquid overflow caused by loss;
4. If there is no spectrophotometer, it can also be measured by a microplate reader with the same wavelength (or selected within the fluctuation range of  $\pm 10\text{nm}$ ).