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### **Determination of Luteolin purity**

A.1 Methods The samples are extracted by ultrasonic wave and detected by high performance liquid chromatography.

A.2 Test instruments and appliances

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A.2.1 Analytical balance, accuracy is 1/100,000th

A.2.2 Ultrasonic Cleaning Instrument: 250W, 20KHz

A.2.3 High performance liquid chromatograph

A.3 Reagents and solutions

A.3.1 Acetonitrile, analytical pure

A.3.2 Water, secondary distilled water

A.3.3 Luteolin control comparison product.

A.3.4 Preparation of mobile phase: mixed with acetonitrile-water (30:70) ratio, obtained by filtration with microporous membrane.

A.3.5 Detector and detection wavelength: UV spectrophotometer, detection wavelength 360nm.

A.4 Methods of operation

A.4.1 Preparation of reference solution: Luteolin comparison product solution (accurate to 0.01mg) was accurately weighed and added with methanol to prepare A solution containing 70 $\mu$ g per 1mL as comparison product solution.

A.4.2 Preparation of test solution: Take A sample of luteolin (about 10mg), weigh it accurately, dissolve it with ultrasonic methanol, and use it as the test solution.

A.4.3 Determination methods

Precisely absorb 10 $\mu$ l of comparison product solution and test solution, inject into liquid chromatograph, and got

#### A.5 Calculation of results

The purity of luteolin was calculated according to Equation (B.1) :

$$\text{Luteolin (\%)} = \frac{S_1 \times C \times A}{S_0 \times (M - M \times B)} \times 100\% \dots \dots \dots \text{(B. 1)}$$

in:

S1-- Peak area value of test product solution;

S0-- Peak area value of comparison product solution;

C-- Concentration of comparison product solution (mg/ mL);

A-- Comparison product purity (%);

B-- Moisture purityof the sample (%);

M-- Concentration of test product solution (mg/ mL).