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Method of Analysis for Curcumin

1. Method summary

After the sample was dissolved by ultrasound, it was measured by high performance liquid chromatography and quantified by external standard method.

2. Apparatus and Utensils

Analytical balance, (0.00001g).

Ultrasonic cleaner: 250W, 20kHz.

High performance liquid chromatograph. (equipped with UV detector)

3. Reagents and solutions

- ① Acetonitrile. Chromatographically pure;
- ② Methanol. Chromatographically pure;
- ③ Glacial acetic acid. Chromatographically pure;
- ④ HPLC grade water or MILIQ water;

4. Curcumin reference substance: HPLC \geq 98%.

5. Chromatographic conditions and system applicability

5.1 Chromatographic conditions

- a) Chromatographic column: octadecylsilane bonded silica gel column, .6mm \times 250mm.
- b) Mobile phase: Acetonitrile-4% glacial acetic acid solution (48:52), filtered through an organic 0.45 μ m membrane filtration and ultrasound are available for use.
- c) Flow rate: 1.00ml/min.
- d) Detection wavelength: 430nm.
- e) Column oven temperature: 30 $^{\circ}$ C

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5.2 System suitability

The number of theoretical plates should not be less than 4000 calculated based on the curcumin peak.

6. Operation method

6.1 Sampling

Refer to the method for sampling in Appendix IIA of the 2010 edition of the Pharmacopoeia of the People's Republic of China.

6.2 Preparation of reference solution

Accurately weigh about 10mg of curcumin reference substance, place it in a 100mL brown measuring flask, and add methanol dissolve by ultrasonic and dilute to the mark to obtain a 0.10mg/ml reference solution.

6.3 Preparation of test solution

Take an appropriate amount of turmeric extract powder (depending on the content), accurately weigh it, and place it in 100mL brown, added about 80 mL of methanol to the color measuring flask, take it out after ultrasonic extraction for 15 minutes, and place it at room temperature. Dilute with methanol to the mark and shake well. Filter with an organic 0.45um microporous membrane, set the filtrate in the brown sample bottle, the test solution is obtained.

6.4 Determination method

Precisely draw 5ul of the reference solution and the test solution respectively into the liquid chromatograph, press the content was determined by the external standard method.

Note: Curcumin is chemically unstable. The reference substance and test solution should be prepared in test within 2h.

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7 Result calculation

The content is calculated according to formula (C1):

$$\text{Curcumin content(\%)} = \frac{A_x * C * V}{A_s * m} * 100\% \dots\dots\dots (C1)$$

Where:

A_x —The total peak area of the three components of the test solution in the chromatogram;

A_s ——the peak area of curcumin in the chromatogram of the reference substance solution;

C —The concentration of curcumin in the reference solution, in milligrams per milliliter (Mg/mL);

V ——The volume of the test solution, in milliliters (mL);

m ——The weighing amount of the sample, the unit is milligram (mg)