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Determination of organic acids in plant - Liquid chromatography-tandem mass spectrometry

植物中有机酸的测定 液相色谱-质谱/质谱法

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1 Scope

This document specifies the method for the determination of organic acids in plants, by liquid chromatography-tandem/mass spectrometry.

This document applies to the determination of fumaric acid, trans-aconitic acid, adipic acid, gallic acid, vanillic acid, chlorogenic acid, caffeic acid, syringic acid, p-coumaric acid, ferulic acid, p-hydroxybenzoic acid, salicylic acid, 2,5-dihydroxybenzoic acid, 3,4-dihydroxybenzoic acid, in citrus, apple, strawberry, cucumber, tomato, honeysuckle, codonopsis, hawthorn, medlar.

2 Normative references

The contents of the following documents constitute the indispensable clauses of this document through normative references in the text. Among them, for dated reference documents, only the version corresponding to that date is applicable to this document; for undated reference documents, the latest version (including all amendments) is applicable to this document.

GB/T 6682 Water for analytical laboratory use - Specification and test methods

3 Terms and definitions

The following terms and definitions apply to this document.

3.1

Organic acids

An acidic organic compound in plant materials.

4 Principle

After the specimen is heated and refluxed by 50% methanol solution, it is separated by reversed-phase chromatographic column, determined and

5.9 Internal standard solution: Accurately pipette 1.0 mL of the internal standard stock solution, into a 100 mL volumetric flask. Use 50% methanol solution to dilute it, to make the volume to the mark. Mix well, to prepare an internal standard solution, which has a concentration of 1.0 mg/L. Prepare it before use.

5.10 Mixed standard working solution: Accurately pipette an appropriate amount of mixed standard stock solution, into a 10 mL volumetric flask. Then accurately add 2.5 mL of internal standard solution respectively. Use 50% methanol solution to dilute it, to prepare the series of mixed standard working solutions, which have a concentration of 0.10 mg/L, 0.25 mg/L, 0.50 mg/L, 0.75 mg/L, 1.0 mg/L, 1.25 mg/L. Prepare it before use.

5.11 0.22 μm hydrophilic nylon microporous filter membrane.

6 Equipment

6.1 Liquid chromatography-tandem/mass spectrometer: Equipped with electrospray ion source (ESI).

6.2 Electronic balance: Sensitivity is 0.0001 g and 0.01 mg.

6.3 High-speed centrifuge: The speed is not less than 10000 r/min.

6.4 High-speed homogenizer.

6.5 High-speed crusher.

6.6 Condensation reflux device.

7 Analytical procedures

7.1 Sample preparation

Take about 1 kg of representative dried samples of honeysuckle, codonopsis, hawthorn, wolfberry, etc. Use a crusher to crush it. Mix well. Divide the prepared specimen equally into two sets. Contain them in clean vessels. Seal and mark it. Store it at 0 °C ~ 4 °C. Make determination within one week.

Take about 1 kg of representative fresh samples of citrus, apples, strawberries, cucumbers, tomatoes, etc. Use a homogenizer to homogenize them. Divide the prepared specimen equally into two sets. Contain them in clean vessels. Seal and mark it. Prepare it before use.

7.2 Processing of specimen

Weigh about 1.5 g of dry sample or 10 g of fresh sample (accurate to 0.001 g),

Perform two parallel determinations, on the same specimen, according to the provisions of 7.2 ~ 7.4.

8 Processing of test data

The calculation of the content of the analyte, in the sample, is as shown in formula (1):

$$X = \frac{\rho \times V}{m} \qquad \cdots\cdots(1)$$

Where:

X - The content of the tested component in the specimen, in milligrams per kilogram (mg/kg);

ρ - The mass concentration of the tested component solution, which is obtained from the standard working curve of internal standard, in micrograms per milliliter (μg/mL);

V - The total volume of the sample solution, in milliliter (mL);

m - The mass of the specimen, in grams (g).

The calculation result is expressed as the arithmetic mean of two independent determination results, which are obtained under repeatability conditions. The result retains to two decimal places.

9 Precision

The absolute difference between two independent determination results, which are obtained under repeatability conditions, shall not exceed 10% of the arithmetic mean.

10 Others

Refer to Table A.1 in Appendix A for the limits of quantification of 14 organic acids, which are determined by this method.

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