Original: Japanese

Provisional Translation

Analytical Method for Fluopicolide (Agricultural Products)

1. Analyte

Fluopicolide

2. Instruments

Liquid chromatograph-mass spectrometer (LC-MS)

Liquid chromatograph-tandem mass spectrometer (LC-MS/MS)

3. Reagents

Use the reagents listed in Section 3 of the General Rules, except the following.

Reference standard of fluopicolide: Contains not less than 97% of fluopicolide. Melting point of the standard is 150–154°C.

4. Procedure

1) Extraction

For fruits and vegetables, weigh 20.0 g of sample. For grains, legumes, nuts and seeds, add 20 mL of water to 10.0 g of sample and let stand for 2 hours. For tea leaves, add 20 mL of water to 5.00 g of sample and let stand for 2 hours.

Add 100 mL of acetonitrile, homogenize, and filter with suction. Add 50 mL of acetonitrile to the residue on the filter paper, homogenize, and filter as described above. Combine the resulting filtrates and add acetonitrile to make exactly 200 mL.

2) Clean-up

i) Graphitized carbon black column chromatography

Add 5 mL of acetonitrile to a graphitized carbon black cartridge (500 mg) and discard the effluent. Transfer the extract obtained in 1) (5 mL for fruits and vegetables, 10 mL for grains, legumes, nuts and seeds, 20 mL for tea leaves) to the cartridge and elute with 10 mL of acetone. Collect the total eluate, add 10 mL of water, and concentrate to about 10 mL at below 40°C.

ii) Octadecylsilanized silica gel column chromatography

Add 5 mL each of acetonitrile and water to an octadecylsilanized silica gel cartridge (1,000 mg) sequentially, and discard the effluent. Transfer the solution obtained in i) to the cartridge. Wash the container with 10 mL of acetonitrile/water (3:7, v/v), add the washing to the cartridge, and discard the effluent. Elute with 10 mL of acetonitrile/water (3:2, v/v), add acetonitrile/water (3:2, v/v) to the eluate to make exactly 10 mL, and use this solution as the test solution.

5. Calibration curve

Prepare 0.0005–0.01 mg/L fluopicolide standard solutions (acetonitrile/water (3:2, v/v)) of several concentrations. Inject 10 µL of each standard solution to LC-MS, or 4 µL to LC-MS/MS and make a calibration curve by peak-height or peak-area method.

6. Quantification

Inject 10 µL of the test solution to LC-MS, or 4 µL to LC-MS/MS, and calculate the concentration of fluopicolide from the calibration curve made in **5**.

7. Confirmation

Confirm using LC-MS or LC-MS/MS.

8. Measurement conditions

Column: Octadecylsilanized silica gel, 2.1 mm in inside diameter, 150 mm in length and 5 µm in particle diameter

Column temperature: 40°C

Mobile phase: Acetonitrile/2 mmol/L ammonium acetate solution (1:1, v/v)

Ionization mode: ESI (+)

Major monitoring ions (*m/z*)

1) LC-MS: 383

2) LC-MS/MS: precursor ion 383, product ion 173, 109

Injection volume

1) LC-MS: 10 µL

2) LC-MS/MS: 4 µL

Expected retention time: 10 min

9. Limit of quantification

0.01 mg/kg

10. Explanatory note

1) Outline of analytical method

The method consists of extraction of fluopicolide from sample with acetonitrile, clean-up with a graphitized carbon black cartridge and an octadecylsilanized silica gel cartridge, and quantification and confirmation using LC-MS or LC-MS/MS.

2) Notes

i) If sample and solvent are not uniformly mixed during extraction, add 2 g of Celite to prepare a uniformly homogenized sample.

ii) It has been confirmed that the “Multi-residue Method I for Agricultural Chemicals by LC-MS (Agricultural Products)” was applicable to fluopicolide in potato and tea leaves but not applicable in orange. Therefore, the multi-residue method can be used under the condition that the performance is verified before use.

11. References

None

12. Type

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