Original: Japanese

Provisional Translation

Analytical Method for Flucetosulfuron (Agricultural Products)

1. Analyte

Flucetosulfuron (sum of isomers)

2. Instrument

Liquid chromatograph-mass spectrometer (LC-MS)

3. Reagents

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

Reference standard of flucetosulfuron: Contains not less than 98% of flucetosulfuron.

4. Procedure

1) Extraction

For fruits and vegetables, weigh 20.0 g of sample. For grains, legumes, nuts and seeds, weigh 10.0 g of sample, for tea leaves, weigh 5.00 g of sample, add 20 mL of water and let stand for 30 minutes.

Add 100 mL of acetonitrile/water (4:1, v/v), homogenize, and filter with suction. Add 50 mL of acetonitrile/water (4:1, v/v) to the residue on the filter paper, homogenize, and filter with suction. Combine the resulting filtrates, and add acetonitrile/water (4:1, v/v) to make exactly 200 mL. Take a 1 mL (2 mL for grains, legumes, nuts and seeds, 4 mL for tea leaves) aliquot of the extract, and add 9 mL (8 mL for grains, legumes, nuts and seeds, 6 mL for tea leaves) of acetonitrile/ water (4:1, v/v).

2) Clean-up

i) Graphitized carbon black column chromatography

Add 10 mL of acetonitrile/water (4:1, v/v) to a graphitized carbon black cartridge (500 mg), and discard the effluent. Transfer the extract obtained in 1) to the cartridge, elute with 50 mL of acetonitrile/water (4:1, v/v), collect the total eluate, concentrate to about 8 mL at below 40°C and remove acetonitrile. Add water to make about 10 mL solution, and add 0.1 mL of acetic acid.

ii) Octadecylsilanized silica gel column chromatography

Add 5 mL each of 1% acetic acid-acetonitrile and 1% acetic acid to an octadecylsilanized silica gel cartridge (1,000 mg) sequentially, and discard the effluents. Transfer the solution obtained in i) to the cartridge, add 10 mL of 1% acetic acid/1% acetic acid-acetonitrile solution (7:3, v/v), and discard the effluent. Elute with 10 mL of 1% acetic acid/1% acetic acid-acetonitrile (1:1, v/v), concentrate the eluate at below 40°C and remove the solvent. Dissolve the residue in 10 mL of acetonitrile/ water (4:1, v/v).

iii) Trimethylaminopropylsilanized silica gel column chromatography

Add 10 mL of acetonitrile/water (4:1, v/v) to a trimethylaminopropylsilanized silica gel

cartridge (500 mg), and discard the effluent. Transfer the solution obtained in ii) to the cartridge, elute with 10 mL of acetonitrile/water (4:1, v/v), collect the total eluate, concentrate at below 40°C and remove the solvent. Dissolve the residue in acetonitrile/water (2:3, v/v) to make exactly 2 mL, and use this solution as the test solution.

5. Calibration curve

Prepare 0.0005–0.01 mg/L flucetosulfuron standard solutions (acetonitrile/water (2:3, v/v)). Inject 5 µL of each standard solution to LC-MS, and make a calibration curve by peak-height or peak-area method.

6. Quantification

Inject 5 µL of the test solution to LC-MS and calculate the concentration of flucetosulfuron from the calibration curve made in **5**.

7. Confirmation

Confirm using LC-MS.

8. Measurement conditions

Example

Column: Octadecylsilanized silica gel, 2.0 mm in inside diameter, 150 mm in length and 5 µm in particle diameter

Column temperature: 40°C

Mobile phase: 0.1 vol% acetic acid/0.1 vol% acetic acid-acetonitrile solution (3:2, v/v).

Ionization mode: ESI (+)

Major monitoring ions (*m/z*): 510, 488

Expected retention time: 12 min

9. Limit of quantification

0.01 mg/kg

10. Explanatory note

1) Outline of analytical method

The method consists of extraction of flucetosulfuron from sample with acetonitrile/water (4:1, v/v), clean-up with a graphitized carbon black cartridge, an octadecylsilanized silica gel cartridge, and a trimethylaminopropylsilanized silica gel cartridge, quantification and confirmation using LC-MS.

2) Notes

i) The reference standard of flucetosulfuron is a mixture of *erythro* isomer and *threo* isomer (about 9:1). Although flucetosulfuron is detected as one peak under the measurement conditions described above, two isomers may be separately detected as two peaks depending on the measurement conditions. In such a case, make a calibration curve and quantify using the sum of heights or areas of both peaks.

ii) Flucetosulfuron was not eluted sufficiently from a graphitized carbon black/aminopropyl－silanized silica gel layered cartridge, and “Multi-residue Method I for Agricultural Chemicals by LC-MS (Agricultural Products)” was not applicable to flucetosulfuron.

iii) “Multi-residue Method II for Agricultural Chemicals by LC-MS (Agricultural Products)” is also applicable to flucetosulfuron in rice.

iv) When the analytical method for flucetosulfuron using LC-MS was developed, the following monitoring ions were used:

for quantification (*m/z*): 488

for confirmation (*m/z*): 510

11. References

None

12. Type

C