Original: Japanese
Provisional translation

Analytical Method for Quinclorac (Agricultural Products)

1. Analytes

Quinclorac

Methyl 3, 7-dichloro-8-quinolinecarboxylate (hereafter referred to as metabolite C)

2. Applicable food

Grains, seeds, fruits, and other herbs

3. Instrument

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

4. Reagents

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

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

Reference standard of metabolite C: Contains not less than 98% of metabolite C.

5. Procedure

1) Extraction

i) Grains and seeds

Add 20 mL of water to 10.0 g of the sample and let stand for 30 min. Add 100 mL of 1 vol% hydrochloric acid-acetone solution to the sample, homogenize, and filter with suction. Add 50 mL of 1 vol% hydrochloric acid-acetone solution to the residue on the filter paper, homogenize, and filter as described above. Combine the resulting filtrates and add 1 vol% hydrochloric acid-acetone solution to make exactly 200 mL. Take exactly a 2 mL aliquot of the solution and add 20 mL of 0.1 vol% hydrochloric acid.

ii) Fruits and vegetables

Add 100 mL of 1 vol% hydrochloric acid-acetone solution to 20.0 g of sample, homogenize, and filter with suction. Add 50 mL of 1 vol% hydrochloric acid-acetone solution to the residue on the filter paper, homogenize, and filter as described above. Combine the resulting filtrates, and add 1 vol% hydrochloric acid-acetone solution to make exactly 200 mL. Take exactly a 1 mL aliquot of the solution, and add 20 mL of 0.1 vol% hydrochloric acid.

2) Clean-up

Inject 5 mL each of methanol and 0.1 vol% hydrochloric acid into an octadecylsilanized silica gel cartridge (1,000 mg) sequentially and discard each effluent. After transferring the solution obtained in 1) to the cartridge, add a 10 mL mixture of 0.1 vol% hydrochloric acid and methanol (4:1, v/v), and discard the effluent. Then, add a 10 mL mixture of 0.1 vol% hydrochloric acid and

methanol (1:4, v/v), collect the eluate, add a mixture of 0.1 vol% hydrochloric acid and methanol (1:4, v/v) to make exactly 10 mL, and use this solution as the test solution.

6. Calibration curve

Prepare stock standard solutions of the reference standard of quinclorac and metabolite C. Mix each stock standard solution appropriately, dilute with a mixture of 0.1 vol% hydrochloric acid and methanol (1:4, v/v), and prepare standard solutions of several concentrations. Inject each standard solution into LC-MS/MS respectively, and make calibration curves by peak-height or peak-area method. When the test solution is prepared following the above procedure, the concentration of quinclorac and metabolite C in the test solution corresponding to 0.01 mg/kg (The concentration of metabolite C is calculated as quinclorac.) in the sample results in 0.0001 mg/L (The concentration of metabolite C is calculated as quinclorac.) for each analyte.

7. Quantification

Inject the test solution into LC-MS/MS, calculate the concentration of quinclorac and metabolite C from the calibration curve made in 6., and calculate the concentration of quinclorac including metabolite C using the following equation.

Concentration of quinclorac (including metabolite C) (ppm) = A + B × 0.9452

A: Concentration of quinclorac (ppm)

B: Concentration of metabolite C (ppm)

8. Confirmation

Confirm using LC-MS/MS.

9. Measurement conditions

(Example)

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

Column temperature: 40°C

Mobile phase: Initially, acetonitrile and 0.1 vol% formic acid (2:3, v/v) for 6 min, followed by a linear gradient to (4:1, v/v) in 4 min, and hold for 5 min.

Ionization mode: ESI (+)

Major monitoring ion (*m/z*)

Quinclorac: Precursor ion 242, product ions 224, 161

Metabolite C: Precursor ion 256, product ions 196, 161

Injection volume: 10 μL

Expected retention time

Quinclorac: 5 min

Metabolite C: 11 min

10. Limit of quantification

0.01 mg/kg for each analyte (The concentration of metabolite C is calculated as quinclorac.)

11. Explanatory note

1) Outline of analytical method

The method consists of extraction of quinclorac and metabolite C from the sample with acetone under acidic condition using hydrochloric acid, clean-up with an octadecylsilanized silica gel cartridge, and quantification and confirmation using LC-MS/MS. In the method, quinclorac and metabolite C are quantified respectively. To calculate the concentration of quinclorac including metabolite C, the concentration of metabolite C is converted to the concentration of quinclorac by multiplying by a conversion factor, and the sum of the concentrations of quinclorac and metabolite C is regarded as the analytical result of quinclorac.

2) Notes

i) When the analytical methods for quinclorac and metabolite C using LC-MS/MS were developed, the following monitoring ions were used:

Quinclorac:

for quantitative ions *(m/z*): precursor ion 242, product ion 161

for qualitative ions (*m/z*): precursor ion 242, product ion 224

Metabolite C:

for quantitative ions *(m/z*): precursor ion 256, product ion 161

for qualitative ions (*m/z*): precursor ion 256, product ion 196

ii) Food items used to develop the analytical method: brown rice, blueberry, basil and rapeseed

12. References

None

13. Type

C