ThP-171

Determination of 33 pesticide residues in Ginseng using gas chromatography-triple quadrupole mass spectrometry

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1. Overview

The 33 pesticide residues in traditional Chinese medicine specified in the fifth method of general rule 2341 of Chinese Pharmacopoeia (2020 Edition) were determined by Shimadzu gcms-tq8050 NX. The method has high sensitivity, good repeatability, and can fully meet the requirements of the 2020 Edition of Ch.P.

2. Introduction

In August 2019, the official website of the Chinese State Pharmacopoeia Commission published *the general rules of 0212* for the determination of Chinese herbal medicines and cut crude drugs of Chinese pharmacopoeia 2020 Edition. The most concerned content of *the general rule 0212* is the limit value of 33 banned pesticides in Chinese herbal medicines and cut crude drugs (plants). There are 53 residues of 33 banned pesticides in traditional Chinese medicine. The fifth method of general rule 2341 corresponding to general rule 0212 stipulates the GC-MS/MS and LC-MS/MS analysis methods of 33 banned pesticides. In this paper, referring to the fifth method of general rule 2341, a quantitative analysis method of 33 banned pesticide residues in ginseng was established by GC-MS/MS.

3. Methods

Weigh 3g of ginseng powder and put it into 50 mL centrifuge tube, add 15 mL 1% acetic acid aqueous solution, vortex to make the powder fully soaked and stayed at room temperature for 30min. Add 15mL acetonitrile to the centrifuge tube, vortex and shake for 5min, then add anhydrous magnesium sulfate and anhydrous sodium acetate mixed powder (4:1,M/M) and shake immediately to make it evenly dispersed. Place the centrifuge tube in an ice water bath to cool for 10min, then centrifuge at 4000 r/min for 5min.

Pipette 9 mL of the supernatant into a 15 mL solid phase dispersion tube, vortex and shake for 5min, and centrifuge at 4000 r/min for 5min. Pipette 5 mL of supernatant into a new 15 mL centrifuge tube, purge with nitrogen to about 0.4 mL, and use acetonitrile to bring the volume to 1 mL. The fixed volume liquid was filtered through a 0.22μm filter membrane, and then 0.3 mL of triphenyl phosphate acetonitrile solution was added, and vortexed to mix evenly for analysis.

High Speed Mass Spectrometer

Ultra Fast Scan Speed
- Max. 20000 amu/sec

Ultra Fast MRM
- Max. 888 transition /sec

Figure 1 GCMS-TQ8050 NX triple quadrupole mass spectrometer

4. Results

GC-MS/MS conditions

Instrument: GCMS-TQ8050 NX (Shimadzu Corporation, Japan)
Injection mode: Splitness mode
Column: SH-Rxi-17Sil MS, 30m×0.25mm×0.25μm
Column oven temp.: 60℃ (1 min)_10℃/min_160 ℃ _ 2℃/min_230 ℃ _15℃/min _300℃(6 min)
Carrier gas: Helium
CID gas: Argon
Ionization mode: EI
Detector voltage: Tuning result+0.7kV
Interface temp.: 250℃
Ion source temp.: 250℃

The n-alkanes (C16-C34) standard solution was determined by the analysis method of n-alkanes in the Smart Database, which was used to predict the retention time of the 33 banned pesticide residues and triphenyl phosphate (internal standard). The mass chromatogram of n-alkanes is shown in Figure 2.

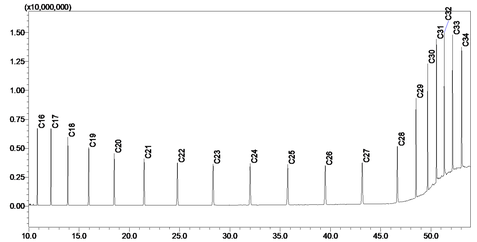

Figure 2 The mass chromatogram of n-alkanes

Table 1 MRM transition of the 33 banned pesticide residues

No.	Compound	Retetion time (min)	CAS .No.	MRM transition	CE (eV)
1	Demeton（O&S）	15.885（O） 20.512（S）	8065-48-3	88.0>60.0	4
				88.0>59.0	20
2	Ethoprophos	16.891	13194-48-4	157.8>96.7	20
				199.7>157.8	5
3	Chlordimeform free base	17.675	6164-98-3	152.0>117.0	15
				196.0>181.0	5
4	Sulfotep	18.377	3689-24-5	322.0>174.0	15
				322.0>294.0	10
5	Phorate	18.610	298-02-2	260.0>75.0	5
				230.8>175.0	10
6	alpha-BHC	19.298	319-84-6	181.0>145.0	15
				218.7>182.9	5
7	Terbufos	20.302	13071-79-9	230.8>129.0	25
				230.8>175.0	13
8	gamma-BHC	22.058	58-89-9	181.0>145.0	15
				218.7>182.9	5
9	Monocrotophos	22.585	6923-22-4	127.0>109.0	12
				127.0>95.0	16
10	Fipronil-desulfinyl	23.858	205650-65-3	388.0>333.0	20
				388.0>281.0	35
11	beta-BHC	24.124	319-85-7	181.0>145.0	15
				218.7>182.9	5
12	delta-BHC	26.240	319-86-8	181.0>145.0	15
				218.7>182.9	5
13	Aldrin	26.319	309-00-2	262.7>192.7	30
				255.0>220.0	20
14	Parathion-methyl	28.112	298-00-0	263.1>109.0	13
				263.1>136.0	5
15	Fipronil-sulfide	30.134	120067-83-6	420.0>351.0	12
				420.0>255.0	20
16	Fipronil	30.355	120068-37-3	351.0>255.0	20
				367.0>213.0	35
17	Parathion	30.586	56-38-2	291.0>109.0	25
				291.0>81.0	30
18	Dicofol deg.	31.257	0-00-0	250.0>139.0	15
				250.0>215.0	5
19	Isofenphos-methyl	32.189	99675-03-3	241.0>120.8	20
				241.0>199.0	5
20	Isocarbophos	33.476	24353-61-5	335.7>108.0	15
				120.7>65.0	20
21	alpha-Endosulfan	34.463	959-98-8	240.8>205.6	15
				240.8>170.0	25
22	Fipronil-sulfone	36.373	120068-36-2	383.0>255.0	20
				383.0>213.0	32
23	Dieldrin	37.233	60-57-1	263.0>193.0	35
				276.8>240.7	10
24	p,p'-DDE	37.243	72-55-9	246.0>176.0	30
				316.0>246.0	25
25	Fenamiphos	38.964	22224-92-6	303.1>122.0	20
				303.1>154.0	30
26	Phosfolan-methyl	39.899	5120-23-0	168.0>109.0	15
				227.0>92.0	10
27	o,p'-DDT	41.760	789-02-6	235.0>165.0	25
				235.0>199.0	15
28	Nitrofen	41.768	1836-75-5	201.8>138.7	28
				282.8>253.0	10
29	p,p'-DDD	42.767	72-54-8	235.0>165.0	25
				237.0>165.0	25
30	beta-Endosulfan	42.897	33213-65-9	206.8>171.8	15
				194.8>124.7	30
31	p,p'-DDT	45.148	50-29-3	235.0>165.0	25
				235.0>199.0	15
32	Endosulfan sulfate	46.991	1031-07-8	271.8>236.7	15
				271.8>141.0	40
33	Triphenyl phosphate（IS）	48.830	115-86-6	326.0>233.0	10
				326.0>215.0	25
34	Coumaphos	52.564	56-72-4	361.8>109.0	16
				361.8>81.0	32

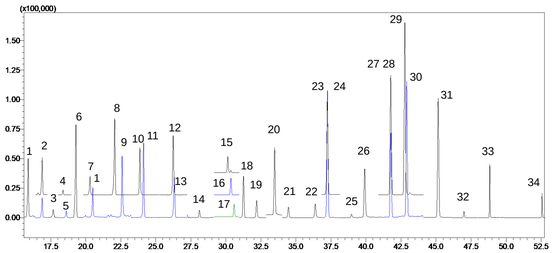

Figure 3 MRM chromatograms of the 33 banned pesticide residues and triphenyl phosphate (internal standard) mixed standard solution in ginseng matrix（concentration : 10ng/mL-25ng/mL）

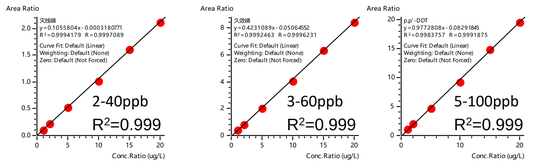

Figure 4 Representative calibration curve （Ethoprophos, Moncrotophos, p,p'-DDT）

33 mixed standard solutions of the banned pesticide residues (the content of the banned pesticide residues in ginseng is 0.002mg/kg~0.005mg/kg) with concentration of 2ng/mL~5ng/mL were prepared with ginseng blank matrix. The MRM chromatogram of some pesticide residues is shown in Figure 3.

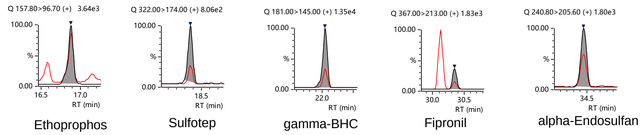

Figure 5 Representative MRM chromatograms of some banned pesticide residues （Ethoprophos, Sulfotep, gamma-BHC, Fipronil and alpha-Endosulfan)

As shown in Table 2, the recovery test were carried out for ginseng samples spiked with all 33 banned pesticide residues at levels of 0.01mg/kg~0.025mg/kg. As a result, the recoveries of all compounds were in the range of 60%~110%.

Table 2 Recovery results (n=3)

No.	Compound	加标浓度（mg/kg）	平均回收率(%)	No.	Compound	加标浓度(mg/kg)	平均回收率(%)
1	Demeton(O&S)	0.01	101.2	18	Dicofol deg.	0.025	81.89
2	Ethoprophos	0.01	91.80	19	Isofenphos-methyl	0.01	101.2
3	Chlordimeform free base	0.01	61.14	20	Isocarbophos	0.025	99.89
4	Sulfotep	0.01	108.6	21	alpha-Endosulfan	0.025	93.11
5	Phorate	0.01	97.91	22	Fipronil-sulfone	0.01	84.61
6	alpha-BHC	0.025	79.82	23	Dieldrin	0.025	76.99
7	Terbufos	0.005	103.0	24	p,p'-DDE	0.025	73.20
8	gamma-BHC	0.025	82.43	25	Fenamiphos	0.01	108.8
9	Monocrotophos	0.015	74.80	26	Phosfolan-methyl	0.015	82.41
10	Fipronil-desulfinyl	0.01	95.06	27	o,p'-DDT	0.025	87.98
11	beta-BHC	0.025	86.65	28	Nitrofen	0.025	82.11
12	delta-BHC	0.025	83.61	29	p,p'-DDD	0.025	92.20
13	Aldrin	0.025	76.41	30	beta-Endosulfan	0.025	85.60
14	Parathion-methyl	0.01	93.56	31	p,p'-DDT	0.025	97.31
15	Fipronil-sulfide	0.01	91.15	32	Endosulfan sulfate	0.025	79.90
16	Fipronil	0.01	104.3	33	Triphenyl phosphate(IS)	-	-
17	Parathion	0.01	96.83	34	Coumaphos	0.025	93.22

5. Conclusions

Using Shimadzu GCMS-TQ8050 NX Triple Quadrupole Gas Chromatography-Mass Spectrometer to determine the 33 banned pesticide residues in the fifth method of the 2020 version of the Chinese Pharmacopoeia 2341 "*Pesticide Residue Determination Method*". Ginseng samples were prepared by the QuEChERS method. In the concentration range of 2ng/mL~40ng/mL (taking parathion as an example), the linear correlation coefficient of the target was greater than 0.995; the mixed standard solution of pesticide residues in the ginseng matrix was prohibited (2ng/mL~5ng/mL) Repeat the injection 6 times, and the RSD of the peak area of each component is less than 5%. The method has good repeatability; For all of the 33 pesticides, acceptable recoveries in the range of 60-110% were achieved at spiking levels of 10μg/kg~25μg/kg. This method fully meets the requirements of the 2020 edition of the Chinese Pharmacopoeia for banned pesticide limit values.