RSC Advances

PAPER

View Article Online
View Journal | View Issue

Cite this: *RSC Adv.*, 2020, **10**, 36906

Screening of multiclass pesticide residues in maca and *Moringa oleifera* by a modified QuEChERS sample preparation procedure and UPLC-ESI-MS/MS analysis

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In the present study, a modified QuEChERS (quick, easy, cheap, effective, rugged, and safe) method was proposed for the simultaneous analysis of 75 pesticides in maca and *Moringa oleifera* with ultra-performance liquid chromatography-electrospray ionization-tandem mass spectrometry (UPLC-ESI-MS/MS). The developed method was validated in accordance with linearity, linear range, limit of detection, limit of quantification, accuracy, precision, and matrix effect. Each analyte had good linearity (R^2 > 0.99) in the corresponding concentration range. The method LOD and LOQ values of all the analytes ranged from 0.01 μg kg^{-1} to 303.35 μg kg^{-1} and 0.03 μg kg^{-1} to 1011.15 μg kg^{-1}, respectively. The recoveries (n = 6) of the analyzed pesticides were in the range of 75.92–113.43%. The RSDs of precision were between 0.60% and 7.36%. All matrix effect values ranged from 81.79% to 118.71% and 80.36% to 119.64% in maca and *Moringa oleifera*, respectively. The analysis of 103 samples showed the presence of isofenphos-methyl in some of them. The method had a good application prospect and could be used as a general approach for the quantitative determination of pesticide residues in food.

Received 22nd July 2020
Accepted 17th September 2020

DOI: 10.1039/d0ra06375d

rsc.li/rsc-advances

1. Introduction

Maca can enhance physical strength, improve fertility, adjust endocrine, enhance immunity, and so on.[1–4] In 2011, China approved maca as a new resource food. At present, China has become the largest maca producing country in the world, with maca being widely cultivated in Lijiang and Diqing of Yunnan province.[5] *Moringa oleifera* can enhance immunity, promote digestion, and improve human intestinal health with rich nutrients and medicinal ingredients, so it is the umbrella of human health. *Moringa oleifera*, the Chinese *Ganoderma lucidum* and American ginseng were known as the "world's three treasures", and *Moringa oleifera* was even named as the "miracle tree" by scientists.[6,7] In November 2012, China's ministry of health approved *Moringa oleifera* leaves as a new resource food. It is widely cultivated in the Yunnan province of China.[8,9] Maca and *Moringa oleifera* can be used as both food and medicine with rich nutrients and secondary metabolites. With the growth of the socio-economic level, people's health consciousness constantly improve, get more attention to food safety, residents also have higher requirement for the quality of food.

Pesticide residues in food are one of the main factors that endanger food safety and human health.[10–12] In order to ensure food safety, many countries and regions in the world have established strict limits for pesticide residues in food, and a lot of researches have been conducted on the detection technology and methods. In recent years, techniques such as gas chromatography (GC), high-performance liquid chromatography (HPLC), GC-tandem mass spectrometry (GC-MS/MS), and LC-tandem mass spectrometry (LC-MS/MS) have been widely used in the detection of pesticide residues and the risk assessment in food.[13–18] However, the application of these technologies needs to be based on reliable sample pretreatment methods. In view of this, Anastassiades *et al.*[19] of the United States agricultural department in 2003 researched a new sample preparation method, namely the QuEChERS (quick, easy, cheap, effective, rugged, and safe) method. This method was mainly applied to the rapid extraction and purification of pesticide residues in food.[20–23] This technique can solve the problems of long pretreatment time, large amount of toxic solvent, qualitative and quantitative interference of coexisting substances in traditional analytical methods. However, because the physical and chemical properties of different analytes differ greatly, and different substrates have different effects on the same analyte, it is difficult to achieve the optimal recovery of all the analytes at the same time. In addition, since the main advantage of the pre-

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treatment method of QuEChERS is simple and fast, the matrix cannot be completely purified, and hence it is easy to produce a matrix effect affecting the accuracy and reliability of the test results when it is combined with various sophisticated high-end detection instruments for trace or ultra-trace analysis.[24,25] No current method can probe all pesticides from all matrices using a single approach. The multiclass and multi-residue analysis of pesticides is a constantly evolving process. The emergence of new pesticides, instruments and techniques has brought both opportunities and challenges.[26] QuEChERS is not just a method or a pair of official methods (AOAC2007.01 and EN 15662), but it is also a flexible approach for chemical sample preparation that is being used in multiple applications.[27] In order to improve the recovery rate and detection efficiency as much as possible, researchers have made a variety of improvements to the QuEChERS method.[28–30] Therefore, the QuEChERS method is optimized according to the differences in the properties of different samples and components to be tested, so as to improve the recovery rate, reduce the interference of impurities, and increase the types of components to be extracted at the same time.

A general method for a large-scale multi-residue pesticide analysis should be established. The pesticide residue analysis is an important quality assessment that solves consumer concerns about food safety and accords with national and international food safety regulations. With the establishment of the food safety standards agency, the scope of pesticide residue monitoring has been expanded dramatically over the past decade. As a result, a multi-residue analysis method should contain as many chemicals as possible and be applicable to multiple substrates.

There are four types of QuEChERS, namely the original QuEChERS method,[19,31] AOAC QuEChERS method,[32,33] European (EN) official QuEChERS method,[34] and modifications of the standard QuEChERS technique.[29,30] Food matrices in pesticide analysis using QuEChERS are as follows: fruits and vegetables,[35] cereals,[36] and animal products.[37] This method has the advantages of dynamic, simple, rapid, fewer analysis steps, and fewer errors. In addition, it is cheap and environmentally friendly due to the use of small amounts of organic solvents. However, few studies have combined the QuEChERS technique with other pre-concentration methods regarding maca and *Moringa oleifera*. In order to improve the extraction efficiency and selectivity, the coupling of QuEChERS is the focus of future research.

In this study, a modified QuEChERS-UPLC-ESI-MS/MS method was developed for multiclass pesticide analysis in maca and *Moringa oleifera*. In the proposed method, the N-EVAP-based extraction method has been added with an optimized concentrated volume that can provide a high enrichment factor for the determination of trace amounts of pesticides in the samples. Furthermore, after using the QuEChERS technology, a certain proportion of matrix interference components is still present in the samples, and these interfering components may get coextracted and adversely affect the chromatographic analysis. Therefore, a pretreatment C18 column was added to the UPLC-MS/MS column to make the sample clean enough and greatly improve the cleanliness of the samples. The increase in the concentration of the measured components in the clean samples can reduce the matrix effect and improve the sensitivity, accuracy, and selectivity of the target compounds.

2. Materials and methods

2.1 Chemicals and reagents

The certified reference standards of all the test compounds were of >99% purity and purchased from A ChemTek, Inc. (Worcester, MA, USA). Deionized water was obtained from a Milli-Q filtration system (Millipore Filter Cor., Bedford, MA, USA). LC/MS grade acetonitrile and formic acid were obtained from Fisher (Thermo Fisher Scientific Inc., Waltham, MA, USA). Other chemicals and solvents were of analytical grade and purchased from Zhiyuan Chemical Factory (Tianjin, China). A dispersive solid-phase extraction purification tube (anhydrous magnesium sulfate ($MgSO_4$): 1500 mg, N-primary secondary amine (PSA): 500 mg, C18: 500 mg, silicone: 500 mg, graphitized carbon black (GCB): 150 mg) was purchased from Agela Technologies (Bona Agela Technology Co. Ltd, Tianjin, China).

2.2 Apparatus

A grinding machine (200 g capacity, model DFT-200, Wenling Linda Machinery Co. Ltd, Zhejiang, China), a vortex mixer (XW-90, Instrument Factory of Shanghai Medical University, Shanghai, China), an oscillator (SHA-C, Jintan Kexi Instrument Co. Ltd, Jiangsu, China), a centrifuge (TGL-16C, Shanghai Precision Instrument Co. Ltd, Shanghai, China), and N-EVAP(ND200-1, Hangzhou Ruicheng Instrument Co. Ltd, Zhejiang, China) were used. The residue analysis was performed using an LC-MS/MS [Acquity I-class UPLC connected to ESI TQD (Waters Corp., Milford, MA, USA) mass spectrometer].

2.3 Selection of pesticides

The paper studied 75 multiclass pesticides with different biological activities such as fungicides, acaricides, insecticides, herbicides, and plant growth regulators. The analytes were selected considering the MRL database of the National Health Commission in china.

2.4 Preparation of standard solutions

The stock solutions of certified reference standards of about 100 $\mu g\ mL^{-1}$ were prepared in acetonitrile. The intermediate standard mixtures of 1 $\mu g\ mL^{-1}$ and working standard solutions of 0.1 $\mu g\ mL^{-1}$ were prepared by diluting the standard stock solutions with acetonitrile. The stock, intermediate, and working standard solutions were stored at $-20\ ^{\circ}C$ before use. All the solutions were filtered through a 0.22 μm membrane filter before injection.

2.5 Liquid chromatographic conditions

In this study, the chromatographic analysis was performed on an Acquity I-class UPLC system (Waters Corp., Milford, MA,

Table 1 The MS/MS parameters for the determination of pesticide residues in the MRM ESI^+ and ESI^- modes

Pesticide	Type of pesticide	Retention time (min)	Precursor (m/z)	Product (m/z)	Cone (V)	Collision energy (eV)	Remark
Chlorfluazuron	Insect growth regulator	9.12	539.8	158.0	42	20	Quantifier
				382.9	42	20	Qualifier
Methamidophos	Insecticide	1.44	142.0	93.9	28	13	Quantifier
				124.9	28	13	Qualifier
Carbaryl	Insecticide	5.39	202.0	117.0	28	28	Quantifier
				145.0	28	22	Qualifier
Dichlorvos	Insecticide, acaricide	5.08	221.0	79.0	34	34	Quantifier
				109.0	34	22	Qualifier
Parathion-methyl	Insecticide	6.41	263.9	79.0	38	36	Quantifier
				109.0	38	22	Qualifier
Phorate sulfoxide	Insecticide	5.35	277.0	96.9	24	32	Quantifier
				143.0	24	20	Qualifier
Pendimethalin	Herbicide	9.01	282.2	194.1	21	17	Quantifier
				212.2	21	10	Qualifier
Parathion	Insecticide	7.35	291.9	110.0	36	33	Quantifier
				236.0	36	14	Qualifier
Phorate sulfone	Insecticide	5.96	293.0	96.9	24	30	Quantifier
				115.0	24	24	Qualifier
Diflubenzuron	Insecticide	6.93	311.1	227.0	32	8	Quantifier
				269.0	32	8	Qualifier
Chlorpyrifos	Insecticide, acaricide	8.99	349.9	97.0	36	32	Quantifier
				198.0	36	20	Qualifier
Fenpropathrin	Insecticide, acaricide	8.99	350.1	97.0	24	34	Quantifier
				125.0	24	14	Qualifier
Chlorantraniliprole	Insecticide	5.79	484.0	286.0	18	12	Quantifier
				453.0	18	17	Qualifier
Tau-fluvalinate	Insecticide, acaricide	10.20	503.0	181.1	24	30	Quantifier
				208.1	24	12	Qualifier
Abamectin (B1a)	Insecticide, acaricide, bactericide	9.69	890.6	305.2	24	25	Quantifier
				567.4	24	11	Qualifier
Methomyl	Insecticide	3.78	163.0	88.0	26	10	Quantifier
				106.0	26	10	Qualifier
Carbendazim	Bactericide	3.72	192.0	105.0	24	41	Quantifier
				160.0	24	16	Qualifier
Pyrimethanil	Bactericide	6.10	200.0	82.0	51	24	Quantifier
				107.0	51	24	Qualifier
Carbofuran	Insecticide	5.28	222.1	123.0	34	16	Quantifier
				165.1	34	16	Qualifier
Acetamiprid	Insecticide	4.42	223.0	56.1	34	15	Quantifier
				126.0	34	20	Qualifier
Imidacloprid	Insecticide	4.31	256.1	175.1	34	20	Quantifier
				209.1	34	15	Qualifier
Chlorobenzuron	Insecticide	6.94	310.1	43.1	42	24	Quantifier
				70.2	42	18	Qualifier
Iprodione	Bactericide	6.76	330.0	245.0	35	15	Quantifier
				288.1	35	15	Qualifier
Isofenphos-methyl	Insecticide	11.18	332.4	58.1	44	30	Quantifier
				91.0	44	28	Qualifier
Tebufenozide	Insecticide	7.03	353.1	133.0	19	20	Quantifier
				297.1	19	8	Qualifier
Pyridaben	Acaricide	9.76	365.1	147.1	28	24	Quantifier
				309.1	28	12	Qualifier
Prochloraz	Bactericide	6.94	376.1	266.0	27	17	Quantifier
				308.0	27	12	Qualifier
Dimethomorph	Bactericide	5.88	388.1	165.0	41	30	Quantifier
				300.9	41	20	Qualifier
Emamectin benzoate	Insecticide, acaricide	7.49	886.6	126.0	45	38	Quantifier
				158.0	45	37	Qualifier
Propamocarb	Bactericide	3.26	189.1	102.0	31	17	Quantifier
				144.0	31	12	Qualifier
Aldicarb sulfoxide	Insecticide	3.19	207.0	89.0	22	14	Quantifier
				132.0	22	10	Qualifier

Table 1 (*Contd.*)

Pesticide	Type of pesticide	Retention time (min)	Precursor (*m*/*z*)	Product (*m*/*z*)	Cone (V)	Collision energy (eV)	Remark
Aldicarb	Insecticide	4.85	213.1	89.1	30	16	Quantifier
				116.1	30	11	Qualifier
Omethoate	Insecticide	3.13	214.1	125.1	26	22	Quantifier
				183.1	26	11	Qualifier
Aldicarb sulfone	Insecticide	3.63	223.0	86.0	31	14	Quantifier
				148.0	31	10	Qualifier
Carbofuran-3-hydroxy	Insecticide	4.23	238.0	163.0	34	16	Quantifier
				181.0	34	10	Qualifier
Forchlorfenuron	Plant growth regulator	5.33	248.1	93.0	36	35	Quantifier
				129.0	36	15	Qualifier
Fenitrothion	Insecticide	6.77	278.0	79.1	38	34	Quantifier
				109.1	38	20	Qualifier
Thiamethoxam	Insecticide	3.96	292.0	132.0	28	22	Quantifier
				211.2	28	12	Qualifier
Triadimefon	Bactericide	6.39	294.1	69.3	31	20	Quantifier
				197.2	31	15	Qualifier
Phoxim	Insecticide	7.80	299.0	129.0	22	13	Quantifier
				153.0	22	7	Qualifier
Phosmet	Insecticide	6.19	318.0	77.0	28	46	Quantifier
				160.0	28	22	Qualifier
Azoxystrobin	Bactericide	6.22	404.0	329.0	28	30	Quantifier
				372.0	28	15	Qualifier
Fipronil sulphone	Insecticide	5.77	453.0	112.0	60	54	Quantifier
				194.0	60	32	Qualifier
Mevinphos	Insecticide	4.52	225.1	127.1	24	15	Quantifier
				193.1	24	8	Qualifier
Phorate	Insecticide, acaricide	7.94	261.0	75.0	17	12	Quantifier
				97.0	17	32	Qualifier
Fenthion	Insecticide	7.43	279.1	169.1	36	16	Quantifier
				247.1	36	13	Qualifier
Phosphamidon	Insecticide	4.76	300.1	127.1	28	25	Quantifier
				174.1	28	14	Qualifier
Tolclofos-methyl	Bactericide	4.74	302.1	127.5	43	20	Quantifier
				176.5	43	13	Qualifier
Fenamiphos	Insecticide	6.26	304.1	202.1	36	36	Quantifier
				217.1	36	24	Qualifier
Triazophos	Insecticide	6.75	314.1	118.9	31	35	Quantifier
				161.9	31	18	Qualifier
Isazofos	Insecticide	6.75	314.1	97.0	34	30	Quantifier
				162.2	34	20	Qualifier
Dichlofenthion	Insecticide	7.74	316.2	46.2	20	12	Quantifier
				74.2	20	30	Qualifier
Phenthoate	Insecticide	7.48	321.0	135.0	18	20	Quantifier
				163.0	18	12	Qualifier
Malathion	Insecticide	6.73	331.0	99.0	20	24	Quantifier
				127.0	20	12	Qualifier
Phosalone	Insecticide, acaricide	7.83	367.9	110.9	22	42	Quantifier
				181.9	22	14	Qualifier
Profenofos	Insecticide	8.23	372.9	127.9	36	40	Quantifier
				302.6	36	20	Qualifier
Difenoconazole	Bactericide	7.28	406.0	111.1	46	60	Quantifier
				251.1	46	25	Qualifier
Dimethoate	Insecticide, acaricide	4.41	230.1	125.0	24	20	Quantifier
				199.0	24	10	Qualifier
Methacrifos	Insecticide, acaricide	6.16	241.1	125.0	20	20	Quantifier
				209.1	20	8	Qualifier
Ethoprophos	Insecticide	6.58	243.2	97.0	32	31	Quantifier
				131.0	32	20	Qualifier
Fonofos	Insecticide	7.81	247.1	109.0	24	20	Quantifier
				137.0	24	10	Qualifier
Cadusafos	Insecticide	7.72	271.1	131.0	28	22	Quantifier
				159.0	28	16	Qualifier

Table 1 (*Contd.*)

Pesticide	Type of pesticide	Retention time (min)	Precursor (*m*/*z*)	Product (*m*/*z*)	Cone (V)	Collision energy (eV)	Remark
Fosthiazate	Insecticide	5.43	284.0	104.0	28	22	Quantifier
				228.0	28	10	Qualifier
Etrimfos	Insecticide	7.61	293.1	125.0	38	26	Quantifier
				265.1	38	16	Qualifier
Quinalphos	Insecticide	7.28	299.0	96.9	24	30	Quantifier
				162.9	24	24	Qualifier
Methidathion	Insecticide	6.14	303.0	85.1	18	20	Quantifier
				145.0	18	10	Qualifier
Diazinon	Insecticide	7.75	305.1	96.9	31	35	Quantifier
				169.0	31	22	Qualifier
Pirimiphos-methyl	Insecticide, acaricide	8.14	306.1	108.1	36	32	Quantifier
				164.1	36	22	Qualifier
Fensulfothion	Insecticide	5.54	309.0	157.1	36	25	Quantifier
				173.1	36	22	Qualifier
Azinphos-methyl	Insecticide, acaricide	6.18	318.0	160.0	20	8	Quantifier
				261.0	20	8	Qualifier
EPN	Insecticide	8.03	324.0	157.0	31	25	Quantifier
				296.0	31	14	Qualifier
Isofenphos	Insecticide	8.14	346.1	217.0	16	22	Quantifier
				245.1	16	12	Qualifier
Ethion	Insecticide, acaricide	9.00	385.0	142.9	30	25	Quantifier
				199.0	30	10	Qualifier
Fipronil desulfinyl	Insecticide	7.33	386.9	282.0	35	30	Quantifier
				351.0	35	15	Qualifier
Fipronil sulphide	Insecticide	7.64	418.9	262.0	35	25	Quantifier
				383.0	35	10	Qualifier

USA) coupled with an autosampler, two pumps, a controller, and a degasser. The separation of pesticides was carried out using an ACQUITY UPLC BEH C18 column (100 mm × 2.1 mm, 1.7 μm) kept at 40 °C, and the autosampler was maintained at 10 °C. The mobile phase consisted of acetonitrile (A) and 0.1% formic acid aqueous solution (B) and a chromatographic gradient program of 5% (A) for 0.0–1.0 min, 5–60% (A) for 1.0–4.0 min, 60–100% (A) for 4.0–10.0 min, 100% (A) for 10.0–12.0 min, 100–5% (A) for 12.0–12.2 min, and 5% (A) for 12.2–16.0 min was used. The flow rate of the mobile phase was set at 0.2 mL min^{-1}, and the injection volume was 1 μL.

2.6 MS/MS conditions

A Xevo tandem quadruple detector (TQD) mass spectrometer was operated using an electrospray ionization (ESI) source (Waters Corp., Milford, MA, USA) with positive and negative modes (ESI^+ and ESI^-). After the direct infusion of reference standards, the multiple reaction monitoring (MRM) conditions were optimized for each pesticide. Other MS conditions were as follows: capillary voltage, 3 kV; cone voltage, 32 V; desolvating temperature, 350 °C; source temperature, 130 °C; source desolvating gas flow, 1000 L h^{-1}; and cone gas flow, 50 L h^{-1}. Each pesticide had two different products. The MS/MS parameters (the type of pesticide, retention time, precursor/product, cone voltage, and collision energy) for the determination of pesticide residues in the MRM ESI^+ and ESI^- modes are presented in Table 1.

2.7 Sample preparation

One hundred and three samples were collected from February to March 2018 in Yunnan province for the quantification of multiclass pesticide residues. Each sample was smashed and passed through a 100 mesh sieve. Five grams of each sample powder was accurately weighed in 100 mL polystyrene centrifuge tubes, followed by the addition of 25 mL of 1% glacial acetic acid solution, which was then dispersed by vortexing and placed for 30 min. Subsequently, 25 mL of acetonitrile was added, and the mixture was agitated vigorously for 1 min and 5 min using a vortex mixer and an oscillator, respectively. Then, 12.5 g of the salt mixture of anhydrous magnesium sulphate ($MgSO_4$) and anhydrous sodium acetate (NaAc) (4 : 1 w/w) was added to the tube and oscillated forcibly for 3 min, followed by placing the tube in cold water for 10 min to cool the sample. The sample was then centrifuged for 5 min at 4000 rpm, and 15 mL of the supernatant was added into a dispersive solid-phase extraction purification tube (anhydrous magnesium sulfate ($MgSO_4$): 1500 mg, N-primary secondary amine (PSA): 500 mg, C18: 500 mg, silicone: 500 mg, graphitized carbon black (GCB): 150 mg). The tube was vortexed fully and oscillated tempestuously for 5 min, and then centrifuged for 5 min at 4000 rpm. Subsequently, 8 mL of the supernatant was dried to approximately 0.6 mL under nitrogen gas at 40 °C. Then, the concentrate was diluted to 1 mL with acetonitrile and vortexed adequately. Lastly, the solution was filtered through a 0.22 μm membrane filter before the UPLC-ESI-MS/MS (LC-MS/MS using

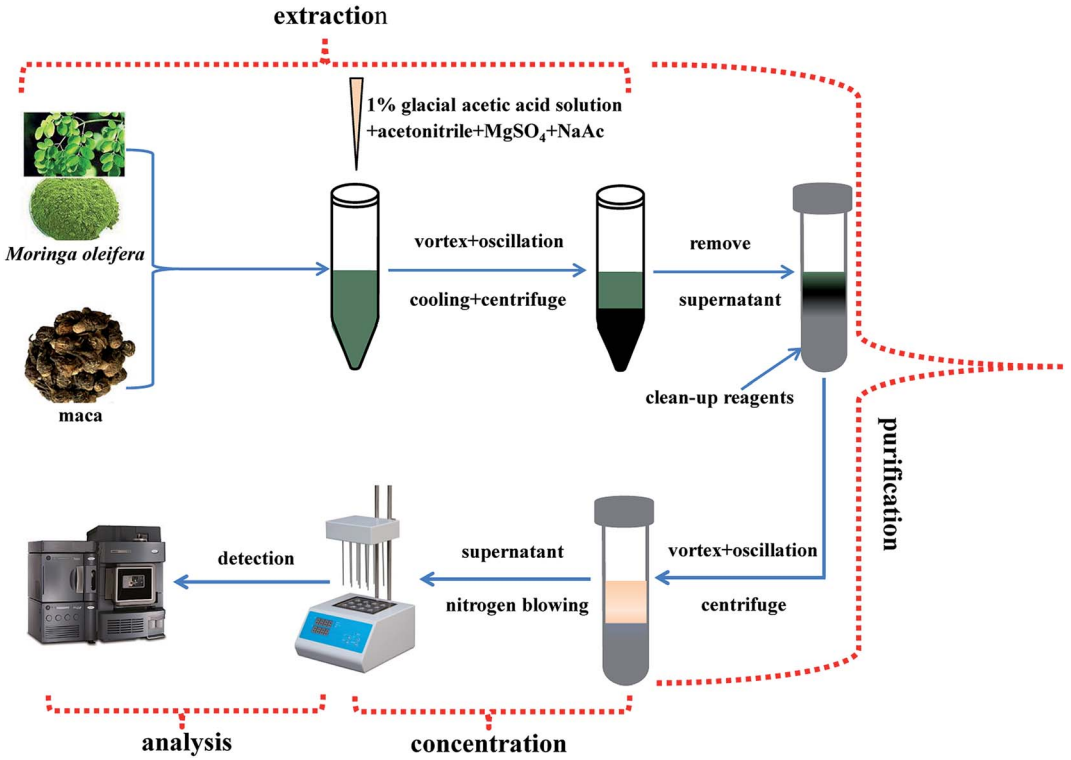

Fig. 1 The flowchart of the sample preparation.

a pretreatment C18 column) analysis. The flowchart of the sample preparation is shown in Fig. 1.

2.8 Method validation

The validation of the method was performed in accordance with the following parameters: linearity, linear range, limit of detection, limit of quantification, accuracy, precision, and matrix effect. Linearity was established on the basis of the quantitative ion peak area (*Y*-axis) *versus* the corresponding concentration (*X*-axis, μg mL^{-1}) for the injection of standard solutions of appropriate concentrations in the linear range 1–500 ng mL^{-1}. The limits of detection (LOD) and quantification (LOQ) were defined as the lowest concentration with signal-to-noise ratios (S/N) of 3 and 10, respectively. To identify a compound, the qualifier ion must have an S/N ratio higher than 3, and the S/N quantifier was many times higher than the qualifier. Indeed, the quantifier yielded an S/N ratio higher than 10, thereby permitting the quantification of the residue. Accuracy was expressed in terms of recovery, and the spiking experiments were carried out in duplicate to estimate the recovery of the target analytes. Precision was usually evaluated in terms of the relative standard deviation (RSD). The matrix effect might affect the exact content of the target compound in a complex sample matrix. In the experiment, sample 1 was a sample spiked to a concentration of 0.1 μg mL^{-1} with a negative sample, and sample 2 was a standard solution of 0.1 μg mL^{-1}. The matrix effect was calculated using the equation given below, where "*A*" represents the area of the corresponding sample. Commonly, the matrix effect < 0.8 indicates significant matrix suppression, while the matrix effect > 1.2 represents the matrix enhancement.[38,39]

$$\text{Matrix effect} = \frac{A_{\text{spiked sample (1)}}}{A_{\text{standard solution (2)}}} \times 100\%$$

3. Results and discussion

3.1 Optimization of UPLC and MS conditions

The mobile phase, flow rate, and chromatographic column for UPLC were chosen to sufficiently acquire the optimal signal response of each target analyte. Different proportions of the mobile phase composition (methanol/water or acetonitrile/water) could not obtain better improvement in the peak shape. However, the mobile phase consisting of acetonitrile and 0.1% formic acid aqueous solution with gradient elution could get good symmetric peaks and preferable analyte ionization, with the elution condition being optimized, as described in the section "*Liquid chromatographic conditions*". Different flow rates (0.1 mL min^{-1}, 0.2 mL min^{-1}, and 0.3 mL min^{-1}) were performed, and the best separation was achieved when the flow rate was 0.2 mL min^{-1}. Agilent Eclipse Plus (50 × 2.1 mm, 1.8 μm), ACQUITY UPLC BEH C18 (50 × 2.1 mm, 1.7 μm), and ACQUITY UPLC BEH C18 (100 × 2.1 mm, 1.7 μm) were used for the separation, and it was observed that better chromatographic peaks were obtained by using the ACQUITY UPLC BEH C18 (100 × 2.1 mm, 1.7 μm) column. In a representative sample, the total ion chromatogram (TIC) of target analytes is shown in Fig. 2.

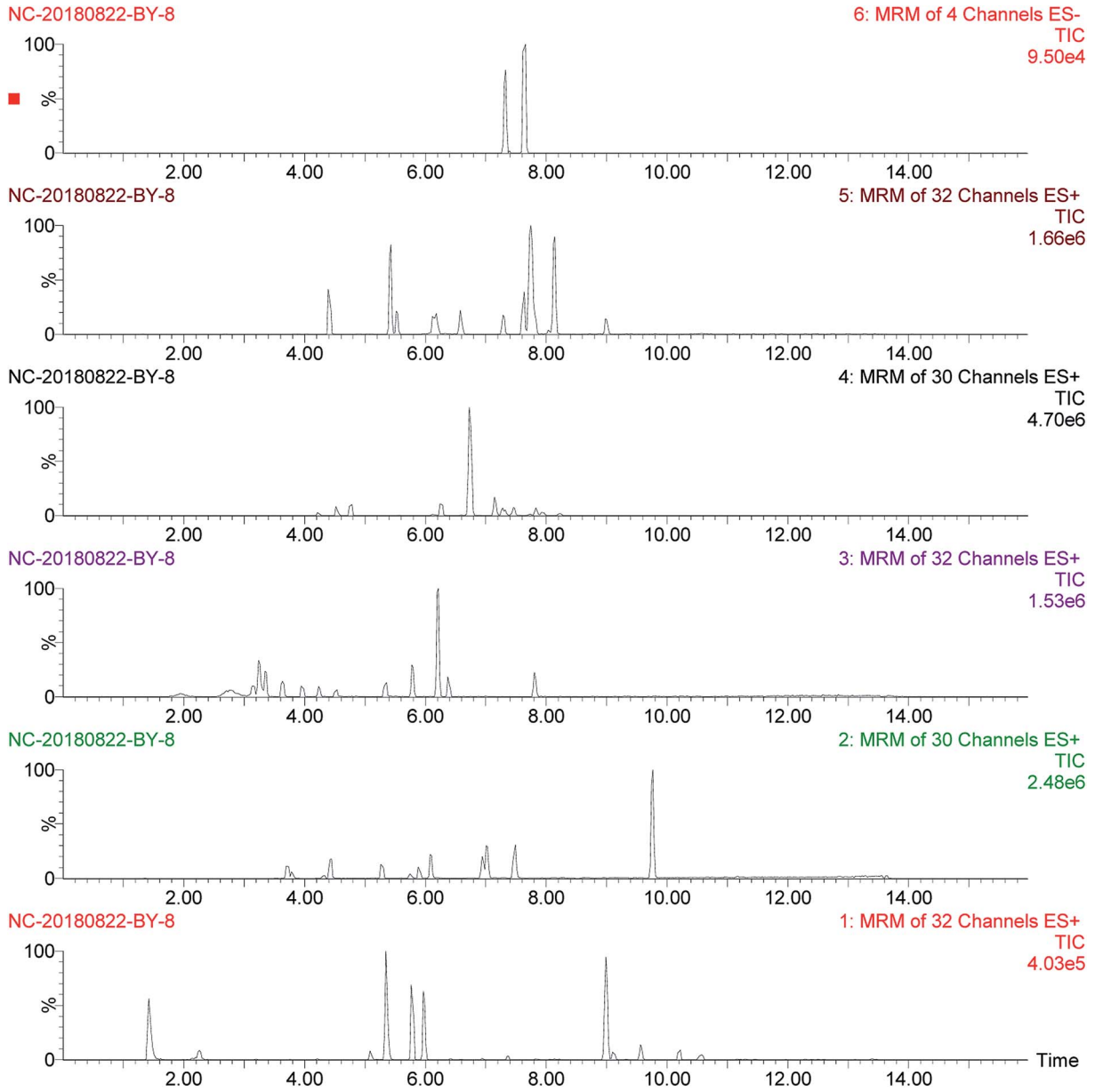

Fig. 2 The total ion chromatogram (TIC) of target analytes in a representative sample.

Each standard solution at a concentration of 50 ng mL^{-1} was injected directly into the mass spectrometer. In order to acquire maximum signal strength for the quantifier and qualifier of each analyte, an electrospray ionization source with positive and negative modes (ESI^+ and ESI^-) was selected. After fragmentation by collision gas, two characteristic peaks of fragment ions with strong responses were found. Therefore, the largest response product was regarded as the quantifier, and the other one was viewed as the qualifier. The multiple reaction monitoring (MRM) conditions of pesticide residues *via* tandem quadruple detector-mass spectrometry (TQD-MS) are summarized in Table 1.

3.2 Optimization of sample pretreatment

Selection of extraction solvent. Due to the large structural difference and wide polarity range of the analyzed target, ways to extract the traces of the target pesticide from the sample was the primary problem to be solved in the pesticide residue analysis. During the pretreatment of the pesticide residue samples, the commonly used extraction agents were acetonitrile, methanol, ethyl acetate, and acetone. The subsequent steps of salting out and water removal could not be carried out with methanol extraction. The content of the pigment in the acetone extract was relatively large, but as acetone and water are mutually soluble, it was difficult to achieve the complete separation of the organic solvent and water. By using ethyl acetate, it was easy to extract non-polar interferences such as wax and fat, but the transfer of most polar pesticides to the organic phase was difficult. Acetonitrile had strong versatility and penetration, good solubility, and it could be separated from water by salting out, thereby providing the best

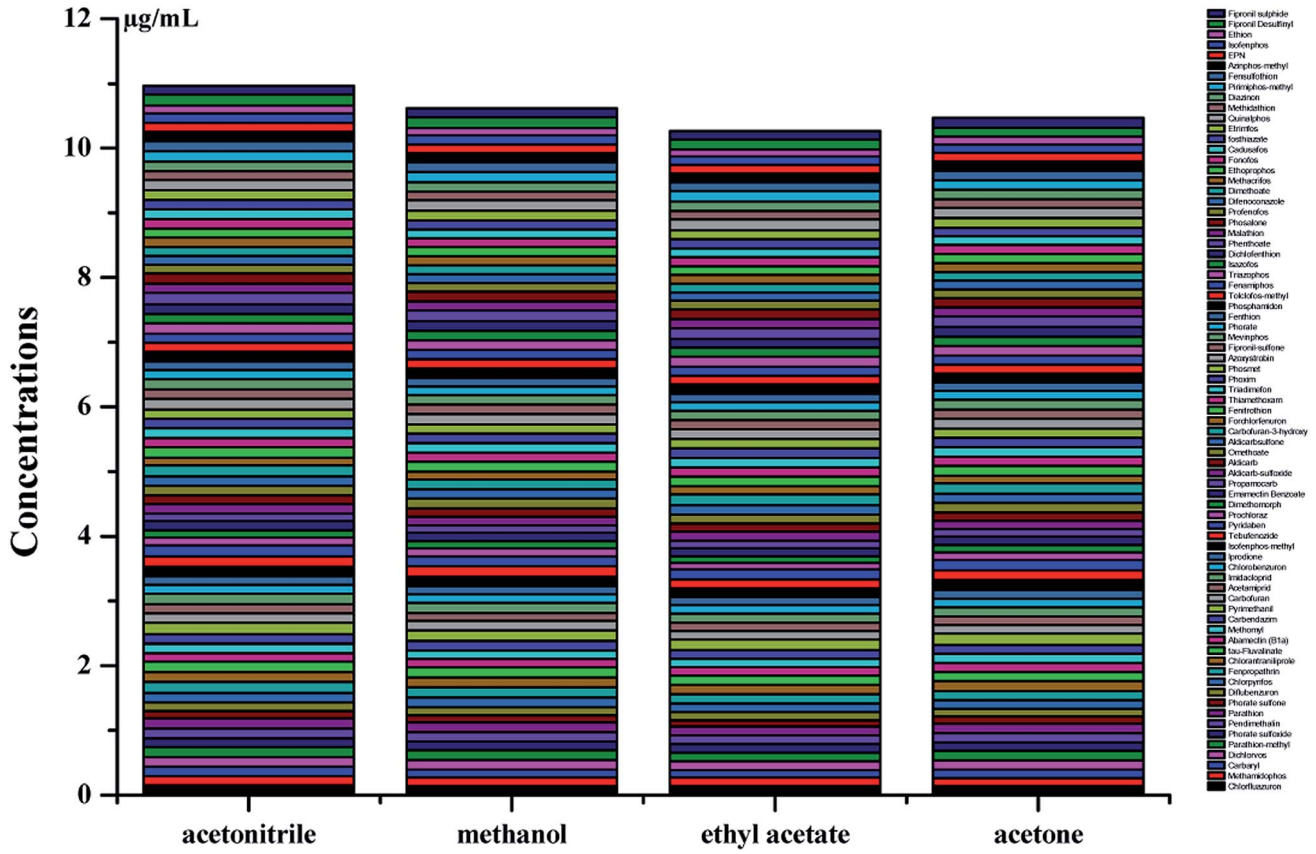

Fig. 3 The effect of extraction solvents in the extraction procedure.

characteristics for extracting the broadest range of pesticides with the least number of co-extractables.[40] However, due to the pH difference in different substrates, some pesticides sensitive to the alkaline environment were easily degraded in the extraction process. Few common pesticides in multi-residue monitoring applications degrade in acidic conditions. Since the pesticides selected were mostly acidic, 1% acetic acid (acetic acid/water, 1/99) was added to the extraction solution to improve the extraction efficiency of the alkaline sensitive pesticides and ensure the stability of the determination results.[41–43] Consequently, the optimal extraction solvent selected was acetonitrile (Fig. 3).

The effect of dehydrating agent. Anhydrous magnesium sulfate ($MgSO_4$) with a strong dehydration effect could promote the distribution of solvents, which leads to the effective extraction of pesticides.[44] The addition of anhydrous sodium acetate (NaAc) and anhydrous magnesium sulfate ($MgSO_4$) could absorb the water in the sample and achieve salting out due to which the substance to be measured remains dissolved in the organic solvents, which was conducive to reducing the subsequent nitrogen blowing and the dewatering pressure. However, anhydrous magnesium sulfate ($MgSO_4$) would release a certain amount of heat in the process of water absorption, resulting in the transformation or decomposition of some unstable pesticides, so cooling in the ice bath could increase the stability of pesticides.[42] The recoveries of 11 typical pesticides with different proportions of dehydrating agents are shown in Fig. 4. Therefore, one can deduce that the optimal dehydrating agent was 12.5 g of the salt mixture of anhydrous magnesium sulphate ($MgSO_4$) and anhydrous sodium acetate (NaAc) (4 : 1 w/w).

Optimization of purification condition. The commonly used clean-up reagents in pesticide residue detection were anhydrous magnesium sulfate ($MgSO_4$), silicone, C18, PSA, and GCB. The sample matrix included pigment, sugar, and organic acid. Anhydrous magnesium sulfate ($MgSO_4$) could remove water from the organic phase;[32] silicone could remove moisture and pigment; C18 could remove long-chain fatty compounds, sterols, and other non-polar interferences;[45] PSA was used in the removal of sugars, fatty acids, organic acids, lipids, and some pigments, and when used in combination with C18, additional lipids and sterols can be removed;[41] GCB was a strong sorbent for removing pigments and polyphenols.[46] Therefore, various options for the clean-up of special commodity co-extractives were presented such as GCB for chlorophyll, ODS for lipids, and PSA for fermented products.[47] A pretreatment C18 column was coupled with LC-MS/MS, the automated pretreatment with the effective removal of proteins and other compounds in the sample matrix could improve the cleanliness of the injected solution once again. The recoveries of 6 typical pesticides with different purification conditions are shown in Fig. 5. Thus, the optimal

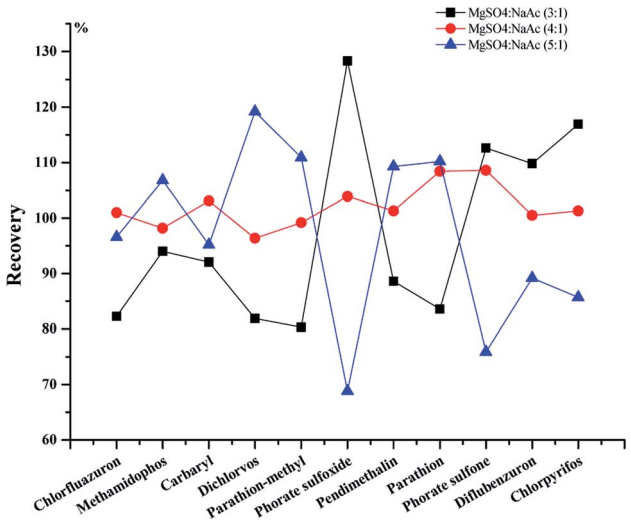

Fig. 4 The recoveries of 11 typical pesticides with different proportions of dehydrating agents.

purification conditions were as follows: anhydrous magnesium sulfate ($MgSO_4$) (1500 mg), N-primary secondary amine (PSA) (500 mg), C18 (500 mg), silicone (500 mg), and graphitized carbon black (GCB) (150 mg).

Optimization of concentration methods. The common methods for concentrating the sample extraction solution were rotary evaporation, freeze-drying, heat drying, and nitrogen blowing under water-bath. The recoveries of 6 typical pesticides with different concentration methods are shown in Fig. 6. The rotary evaporation could only handle a single large amount sample; freeze-drying was slow as it generally requires overnight treatment; heat drying was suitable for thermally stable substances; nitrogen blowing under water-bath was fit for the concentration of less amount of the sample with faster drying speed, thereby improving the sensitivity of the target analyte, so the optimal concentration method was nitrogen blowing under water-bath. The operation method was performed as described in the section "*Sample preparation*".

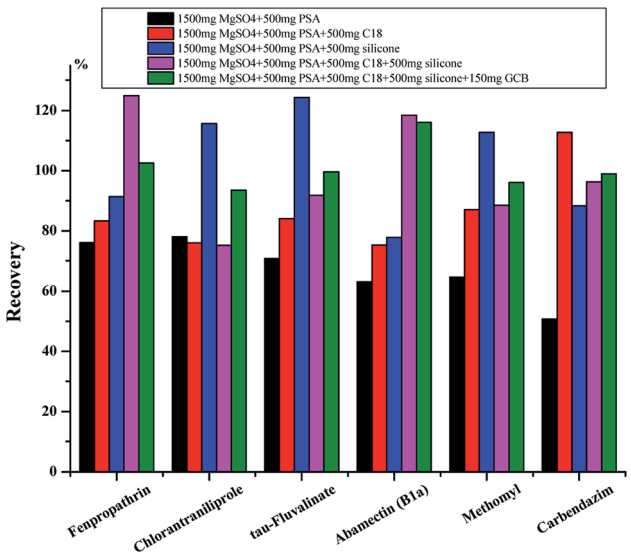

Fig. 5 The recoveries of 6 typical pesticides with different purification conditions.

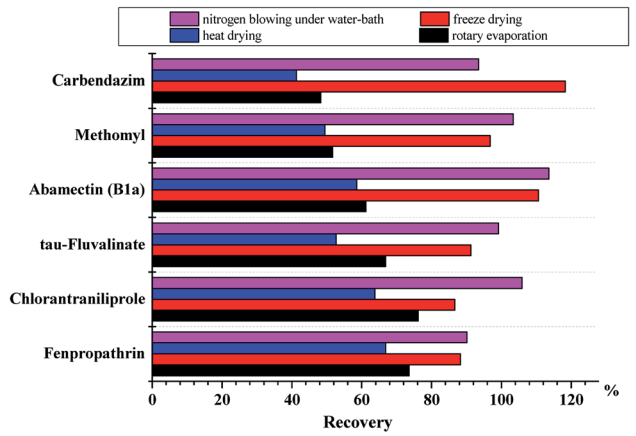

Fig. 6 The recoveries of 6 typical pesticides with different concentration methods.

3.3 Method validation

In this paper, the standard curves for each pesticide acquired good linearity in the range of 1–500 ng mL^{-1} with the correlation coefficient (R^2) > 0.990. The LOD and LOQ values of all the analytes ranged from 0.01 μg kg^{-1} to 303.35 μg kg^{-1} and 0.03 μg kg^{-1} to 1011.15 μg kg^{-1}, respectively, thereby manifesting the high sensitivity of the proposed method. Furthermore, the recoveries ($n = 6$) of the analyzed pesticides were in the range of 75.92–113.43%, which demonstrated that the developed method had excellent accuracy for the determination of the target analytes. The precision based on RSD from six replicate samples was evaluated, and the RSDs were between 0.60% and 7.36%, revealing the good repeatability of the developed UPLC-ESI-MS/MS method. Simultaneously, the matrix effect was considered to be one of the most important and common problems in the pesticide analysis, which had adverse effects on the quantitative analysis. The matrix effect was usually caused by the insufficient removal of fatty acids, phospholipids, pigments, and sugars from the extracting solution. All the matrix effect values ranged from 81.79% to 118.71% and 80.36% to 119.64% in maca and *Moringa oleifera*, respectively, which showed that the quantitative detection of the sample was not affected by the matrix. The regression equations, linear ranges, R^2, LODs, LOQs, recoveries, RSDs, and matrix effects of 75 compounds are listed in Table 2. Meanwhile, the proposed method was compared with the published literature about the determination of pesticides in floristics (Table 3).

Table 2 Linear equations, linear ranges, R^2, LODs, LOQs, recoveries, RSDs, and matrix effects of 75 pesticides in 103 samples

Compounds	Linear equation	Linear range (ng mL^{-1})	R^2	LOD (μg kg^{-1})	LOQ (μg kg^{-1})	Recovery (%)	RSD$_{precision}$ (n = 6) (%)	Matrix effect (%)	
								Maca	*Moringa oleifera*
Chlorfluazuron	$y = 4403.98x + 127.899$	1–100	0.9967	0.73	2.42	95.32	5.71	96.61	101.23
Methamidophos	$y = 60\,388.7x + 4201.16$	5–500	0.9982	4.42	14.72	94.11	3.52	99.22	106.81
Carbaryl	$y = 12\,001.3x - 248.311$	1–100	0.9948	0.52	1.71	103.10	4.83	95.19	92.10
Dichlorvos	$y = 16\,032.7x - 255.536$	10–500	0.9977	76.84	256.15	96.42	4.29	81.88	119.18
Parathion-methyl	$y = 1622.44x - 100.796$	1–200	0.9999	3.14	10.45	99.91	7.17	99.16	100.86
Phorate sulfoxide	$y = 143\,529x - 2705.6$	2–500	0.9946	13.61	45.36	103.89	2.92	81.79	99.12
Pendimethalin	$y = 71\,799.7x - 413.757$	1–100	0.9984	0.07	0.22	98.84	2.73	97.88	88.63
Parathion	$y = 4189.73x - 83.6592$	1–200	0.9955	1.14	3.81	97.86	4.77	110.16	83.56
Phorate sulfone	$y = 53\,992.1x + 195.135$	1–200	0.9905	0.08	0.27	75.92	2.57	91.08	80.36
Diflubenzuron	$y = 1981.39x - 105.615$	2–200	0.9982	3.65	12.18	89.22	5.60	100.47	109.79
Chlorpyrifos	$y = 21\,095x - 120.144$	1–100	0.9964	0.22	0.73	101.25	4.19	116.92	97.46
Fenpropathrin	$y = 33\,394.6x - 107.92$	1–100	0.9957	0.14	0.46	110.16	3.43	104.87	93.28
Chlorantraniliprole	$y = 76\,379.3x - 796.367$	1–100	0.9968	0.07	0.22	95.74	3.63	103.23	99.51
Tau-fluvalinate	$y = 7740.1x + 69.9103$	1–200	0.9963	0.44	1.47	94.14	4.27	99.59	104.28
Abamectin (B1a)	$y = 156.624x - 11.4867$	10–500	0.9976	35.84	119.48	107.78	7.34	99.14	93.06
Methomyl	$y = 22\,844.9x - 63.5845$	20–500	0.9937	303.35	1011.15	97.09	3.98	96.10	99.42
Carbendazim	$y = 134\,884x - 1543.79$	10–500	0.9986	26.52	88.42	88.33	2.50	98.36	90.75
Pyrimethanil	$y = 87\,330.1x + 63.1455$	1–100	0.9938	0.05	0.18	94.44	3.03	118.23	110.11
Carbofuran	$y = 102\,373x - 1772.69$	1–100	0.9975	0.05	0.17	93.26	3.05	98.02	105.37
Acetamiprid	$y = 185\,492x - 9477.32$	1–100	0.9963	0.04	0.13	94.02	1.22	107.48	105.89
Imidacloprid	$y = 14\,718.9x - 201.094$	2–200	0.9984	0.68	2.27	81.39	4.08	99.25	105.13
Chlorobenzuron	$y = 66\,129.6x + 437.562$	1–100	0.9973	0.05	0.18	87.67	2.62	93.20	119.08
Iprodione	$y = 3156.04x - 240.147$	2–200	0.9985	3.27	10.89	90.82	6.29	100.63	109.36
Isofenphos-methyl	$y = 6207.22x - 125.747$	2–200	0.9998	0.81	2.69	103.49	5.53	103.04	97.74
Tebufenozide	$y = 121\,816x + 468.153$	1–100	0.9918	0.04	0.13	96.56	4.34	108.78	107.32
Pyridaben	$y = 459\,203x + 216.086$	1–100	0.9965	0.01	0.03	104.22	1.73	96.16	105.41
Prochloraz	$y = 84\,479.1x - 922.982$	1–100	0.9943	0.07	0.22	77.14	3.20	115.02	101.26
Dimethomorph	$y = 86\,433.9x - 621.198$	1–100	0.9912	0.07	0.23	89.17	2.79	89.11	105.43
Emamectin benzoate	$y = 283\,956x - 1525.82$	1–100	0.9992	0.02	0.06	95.71	1.76	111.73	96.24
Propamocarb	$y = 82\,570.4x + 1634.29$	1–100	0.9908	0.02	0.07	93.14	1.67	97.71	109.32
Aldicarb-sulfoxide	$y = 17\,944.6x - 152.452$	1–200	0.9956	0.25	0.83	107.52	5.44	96.74	95.92
Aldicarb	$y = 1650.99x - 52.811$	2–500	0.9978	3.48	11.59	90.91	7.24	101.51	107.62
Omethoate	$y = 103\,447x - 1454.94$	1–100	0.9978	0.09	0.30	92.62	7.36	100.78	106.59
Aldicarbsulfone	$y = 53\,481.2x + 726.102$	1–100	0.9994	0.08	0.26	95.82	4.00	102.89	101.56
Carbofuran-3-hydroxy	$y = 31\,453.9x - 289.78$	1–200	0.9972	0.22	0.73	85.13	4.44	113.04	85.13
Forchlorfenuron	$y = 74\,444.9x - 1291.53$	1–200	0.9936	0.16	0.53	95.11	3.05	83.62	102.53
Fenitrothion	$y = 3535.14x - 39.6721$	5–500	0.9992	2.18	7.25	86.36	5.75	97.41	114.84
Thiamethoxam	$y = 52\,061.2x - 716.041$	1–100	0.9975	0.10	0.32	94.67	3.93	101.25	81.63
Triadimefon	$y = 99\,365.3x - 1039.06$	1–100	0.9950	0.05	0.18	86.68	3.63	96.62	108.32
Phoxim	$y = 52\,111.5x - 135.064$	1–100	0.9989	0.10	0.32	85.80	2.87	100.51	91.23
Phosmet	$y = 29\,610.6x - 549.165$	1–200	0.9982	0.18	0.59	95.16	4.64	102.83	107.92
Azoxystrobin	$y = 378\,351x - 2332.66$	1–100	0.9956	0.01	0.04	101.354	2.26	111.45	109.94
Fipronil-sulfone	$y = 60\,332.4x - 511.308$	10–500	0.9975	44.17	147.24	94.09	2.72	118.71	84.13
Mevinphos	$y = 67\,838.5x - 775.089$	1–200	0.9969	0.11	0.36	102.91	5.23	102.54	93.62
Phorate	$y = 70\,294.9x - 1152.52$	1–100	0.9976	0.07	0.23	92.89	2.09	95.22	115.23
Fenthion	$y = 19\,726.6x - 243.559$	1–200	0.9976	0.28	0.93	95.90	3.96	93.74	107.81
Phosphamidon	$y = 175\,158x - 2071.14$	1–100	0.9976	0.05	0.16	88.23	1.32	100.43	101.14
Tolclofos-methyl	$y = 4072.56x - 89.1861$	2–200	0.9942	1.23	4.11	90.40	4.19	112.03	93.52
Fenamiphos	$y = 179\,909x - 1479.86$	1–100	0.9987	0.03	0.09	96.68	2.17	101.78	107.28
Triazophos	$y = 379\,606x - 950.747$	1–100	0.9984	0.01	0.04	94.21	1.93	102.34	105.78
Isazofos	$y = 686\,684x - 3209.08$	1–200	0.9993	6.43	21.44	96.29	0.60	103.13	101.02
Dichlofenthion	$y = 12\,725.3x + 483.144$	1–500	0.9906	250.41	834.71	108.2	4.72	99.62	119.64
Phenthoate	$y = 47\,548x + 145.255$	1–100	0.9963	0.09	0.31	97.14	2.45	98.21	86.78
Malathion	$y = 367\,467x - 2199.49$	1–100	0.9992	0.01	0.05	92.18	2.28	103.62	110.23
Phosalone	$y = 103\,828x - 138.642$	1–100	0.9992	0.05	0.16	82.38	1.66	110.04	108.52
Profenofos	$y = 20\,307.6x - 42.0899$	1–200	0.9962	0.23	0.76	87.8	4.23	105.67	99.43
Difenoconazole	$y = 128\,220x - 635.362$	1–100	0.9990	0.05	0.15	85.39	1.78	101.31	104.04
Dimethoate	$y = 234\,934x - 2342.4$	1–100	0.9985	0.03	0.11	84.46	2.31	103.78	109.56
Methacrifos	$y = 21\,311.1x - 138.44$	1–200	0.9984	0.23	0.77	95.51	3.58	101.62	111.89
Ethoprophos	$y = 82\,643.6x - 947.642$	5–500	0.9991	34.26	114.19	96.47	2.43	101.85	97.64

Table 2 (*Contd.*)

Compounds	Linear equation	Linear range (ng mL^{-1})	R^2	LOD (μg kg^{-1})	LOQ (μg kg^{-1})	Recovery (%)	$RSD_{precision}$ ($n = 6$) (%)	Matrix effect (%)	
								Maca	*Moringa oleifera*
Fonofos	$y = 67\,849.2x - 686.454$	1–200	0.9987	0.43	100.40	100.70	3.70	100.42	105.51
Cadusafos	$y = 176\,382x - 868.372$	5–500	0.9934	9.82	32.73	97.42	1.60	102.62	87.05
Fosthiazate	$y = 286\,765x - 2455.26$	1–100	0.9953	0.05	0.17	83.91	2.39	99.60	96.02
Etrimfos	$y = 158\,015x - 1371.18$	1–100	0.9988	0.03	0.10	92.89	2.79	96.81	118.33
Quinalphos	$y = 70\,587.5x - 319.706$	2–200	0.9985	1.29	4.29	102.67	4.32	110.14	89.32
Methidathion	$y = 58\,479.6x - 309.929$	1–100	0.9973	0.09	0.31	98.42	1.81	97.21	88.89
Diazinon	$y = 296\,816x - 751.324$	1–100	0.9993	0.02	0.06	91.56	1.69	97.42	93.56
Pirimiphos-methyl	$y = 283\,855x - 85.2483$	1–100	0.9983	0.02	0.05	113.43	1.74	107.04	82.11
Fensulfothion	$y = 44\,725.6x - 360.82$	1–200	0.9984	0.15	0.52	80.40	4.52	98.23	117.31
Azinphos-methyl	$y = 107\,309x - 753.843$	1–100	0.9957	0.08	0.25	104.90	2.98	112.65	97.14
EPN	$y = 14\,800.7x - 55.1401$	1–200	0.9997	0.32	1.07	96.54	3.82	100.33	106.22
Isofenphos	$y = 18\,093.2x - 109.971$	1–200	0.9965	0.52	1.72	76.12	3.23	101.02	96.63
Ethion	$y = 70\,378.8x - 664.326$	1–100	0.9948	0.07	0.23	81.27	2.39	99.82	93.41
Fipronil desulfinyl	$y = 22\,500.5x - 79.4482$	1–200	0.9985	0.21	0.70	98.65	3.47	98.56	109.32
Fipronil sulphide	$y = 26\,024.9x - 25.3713$	1–200	0.9979	0.17	0.58	96.43	3.84	103.53	93.67

3.4 Analysis of real samples

The developed modified QuEChERS-UPLC-ESI-MS/MS method was used to analyze the contents of 75 pesticide residues in 103 samples (Table 4 and Fig. 7). Among 103 samples (63 dried fruits of maca and 40 *Moringa oleifera* leaves), isofenphos-methyl was detected in all maca samples with the concentrations in the range of 0.11–3.08 mg kg^{-1}. However, other pesticides were not detected. As a result, 40 *Moringa oleifera* leaves were found as negative samples. Isofenphos-methyl is a kind of soil insecticide, which has a strong contact action and causes gastric toxicity to pests. Meanwhile, it is a broad-spectrum insecticide with a long residual period, mainly used for preventing and treating grubs, mole crickets, wireworms, and other underground pests coming from wheat, peanuts, soybeans, corns, sweet potatoes, beets, apples, and other crops but is only used in seed mixing or soil treatment; however, it is prohibited from spraying on the fruit tree leaves. For maca, the edible part was mainly the root of maca that was used as the research object in this paper. In order to prevent and control pests, the planting farmers may add isofenphos-methyl to seed dressing or soil treatment, as a result, isofenphos-methyl remains in the root of maca. Regarding isofenphos-methyl, no MRLs had been reported in the dried maca and *Moringa oleifera* samples by the Codex Alimentarius.[59]

Table 3 The comparison of the proposed method with other methods used in the literature for the determination of pesticides in floristics

Floristics	Analytes	Chromatographic methods	Extraction technique	Analyte time	Recovery	LOQ	References
Tea	10 pesticides	HPLC-MS/MS	QuEChERS	15 min	72–116%	1.7–9.0 μg kg^{-1}	48
Pepper	3 pesticides	UPLC-MS/MS	QuEChERS	15 min	104.91%	2–10 μg kg^{-1}	49
Green tea	102 pesticides	HPLC-MS/MS	Modified QuEChERS	42 min	62–125%	0.1–50 μg kg^{-1}	50
Maca and *Moringa oleifera*	75 pesticides	QuEChERS-UPLC-ESI-MS/MS	Modified QuEChERS	16 min	75.92–113.43%	0.03–1011.15 μg kg^{-1}	This method
Tomato	3 pesticides	UPLC-MS/MS	QuEChERS	15 min	60–140%	—	51
Strawberry	16 pesticides	UPLC-MS/MS	Modified QuEChERS	3.5 min	81.8–117.2%	0.3–2.8 μg kg^{-1}	52
Cinnamon bark	60 pesticides	UPLC-MS/MS	d-SPE and QuEChERS	16 min	71–118%	0.5–50 μg kg^{-1}	53
Pecan nuts	47 pesticides	UPLC-MS/MS	Modified QuEChERS	30 min	70–120%	5–10 μg kg^{-1}	54
Oat	60 pesticides	UPLC-MS/MS	QuEChERS	25 min	70–120%	5–10 μg kg^{-1}	55
Strawberry	203 pesticides	UPLC-MS/MS	QuEChERS	27 min	70–120%	2–10 μg kg^{-1}	56
Cucumber and grapefruit	233 pesticides	HPLC-MS/MS	QuEChERS	—	77.87–104.15%	0.42–39.35 μg kg^{-1}	57
Zizania latifolia	25 pesticides	UPLC-MS/MS	QuEChERS	10 min	72–118%	0.5–3.3 μg kg^{-1}	58

Table 4 The pesticide content of the positive samples in 103 samples analyzed by the modified QuEChERS-UPLC-ESI-MS/MS method[a]

Sample name	No.	Sample source	Date of collection	Pesticides detected	Content (mg kg^{-1})	Codex Alimentarius MRLs (mg kg^{-1})
Maca	S1	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.58 ± 0.05	n.e.
	S2	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.24 ± 0.11	n.e.
	S3	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.17 ± 0.02	n.e.
	S4	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.40 ± 0.03	n.e.
	S5	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.83 ± 0.06	n.e.
	S6	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.51 ± 0.05	n.e.
	S7	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.88 ± 0.06	n.e.
	S8	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.61 ± 0.05	n.e.
	S9	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.49 ± 0.13	n.e.
	S10	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.43 ± 0.03	n.e.
	S11	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.53 ± 0.05	n.e.
	S12	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.21 ± 0.10	n.e.
	S13	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.59 ± 0.14	n.e.
	S14	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.71 ± 0.15	n.e.
	S15	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.35 ± 0.12	n.e.
	S16	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.56 ± 0.14	n.e.
	S17	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	2.29 ± 0.19	n.e.
	S18	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.73 ± 0.06	n.e.
	S19	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	2.77 ± 0.23	n.e.
	S20	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	3.06 ± 0.26	n.e.
	S21	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	3.08 ± 0.26	n.e.
	S22	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.00 ± 0.07	n.e.
	S23	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.10 ± 0.08	n.e.
	S24	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.48 ± 0.13	n.e.
	S25	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.21 ± 0.10	n.e.
	S26	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.11 ± 0.01	n.e.
	S27	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.14 ± 0.10	n.e.
	S28	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.38 ± 0.03	n.e.
	S29	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.11 ± 0.08	n.e.
	S30	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.29 ± 0.11	n.e.
	S31	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.68 ± 0.14	n.e.
	S32	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.66 ± 0.05	n.e.
	S33	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.72 ± 0.15	n.e.
	S34	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	2.20 ± 0.19	n.e.
	S35	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.68 ± 0.06	n.e.
	S36	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.92 ± 0.07	n.e.
	S37	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.60 ± 0.05	n.e.
	S38	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.34 ± 0.12	n.e.
	S39	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.91 ± 0.07	n.e.
	S40	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.06 ± 0.08	n.e.
	S41	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.95 ± 0.07	n.e.
	S42	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.09 ± 0.08	n.e.
	S43	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.35 ± 0.12	n.e.
	S44	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.42 ± 0.13	n.e.
	S45	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.43 ± 0.13	n.e.
	S46	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.09 ± 0.08	n.e.
	S47	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.12 ± 0.08	n.e.
	S48	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	2.91 ± 0.25	n.e.
	S49	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.80 ± 0.06	n.e.
	S50	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.58 ± 0.14	n.e.
	S51	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.99 ± 0.07	n.e.
	S52	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.54 ± 0.05	n.e.
	S53	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	3.02 ± 0.26	n.e.
	S54	Yongsheng (Lijiang)	Mar-18	Isofenphos-methyl	1.35 ± 0.12	n.e.
	S55	Yongsheng (Lijiang)	Mar-18	Isofenphos-methyl	1.22 ± 0.11	n.e.
	S56	Yongsheng (Lijiang)	Mar-18	Isofenphos-methyl	1.02 ± 0.09	n.e.
	S57	Litang (Ganzi)	Mar-18	Isofenphos-methyl	0.41 ± 0.03	n.e.
	S58	Litang (Ganzi)	Mar-18	Isofenphos-methyl	0.77 ± 0.06	n.e.
	S59	Xianggelila (Yunnan)	Mar-18	Isofenphos-methyl	1.62 ± 0.14	n.e.
	S60	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.56 ± 0.05	n.e.
	S61	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	1.42 ± 0.12	n.e.
	S62	Yulong (Lijiang)	Mar-18	Isofenphos-methyl	0.69 ± 0.05	n.e.
	S63	Luquan (Kunming)	Mar-18	Isofenphos-methyl	0.26 ± 0.02	n.e.

[a] n.e.: not established by the Codex Alimentarius.

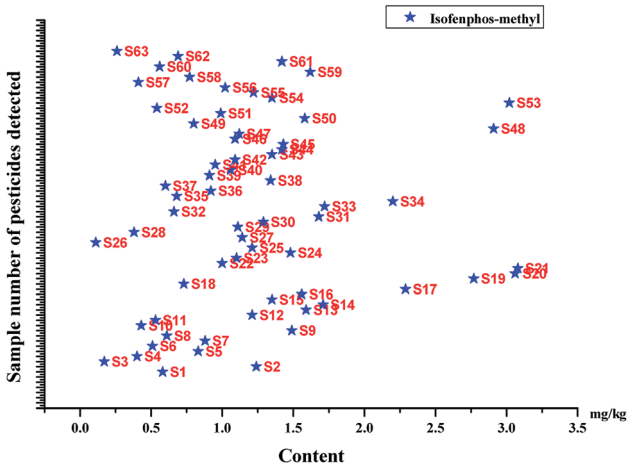

Fig. 7 Sample number of pesticides detected in 103 samples.

4. Conclusions

A sensitive and effective modified QuEChERS coupled with the UPLC-ESI-MS/MS method was successfully applied for the determination of 75 multiclass pesticides in maca and *Moringa oleifera* samples. The feasibility of the method was evaluated in the light of linearity, linear range, LOD, LOQ, accuracy, precision, and matrix effect. The validation studies manifested that the proposed method had good linearity, accuracy, and precision. The developed method was used to analyze the practical samples, and the analyses of 63 maca samples from Yunnan in China demonstrated the presence of isofenphos-methyl, but the MRL was not set by the Codex Alimentarius. Consequently, the risk and exposure levels of isofenphos-methyl in the dried fruit of maca should be further investigated and explored. Meanwhile, this method showed a good application prospect and could be used as a general method for the quantitative determination of pesticide residues in food.

Conflicts of interest

The authors have declared no conflict of interest.

Acknowledgements

The paper was supported by the Analysis Test Fund of Kunming University of Science and Technology (No. 2020T20070029) and Natural Science Fund of Yunnan Province (No. 2017ZF004).

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