

Determination of 21 Pesticide Residues in *Cornus officinalis* Sieb. et Zucc. by UPLC-MS/MS

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Abstract

Purpose: A method based on dispersive solid-phase extraction (dSPE) combined with ultra-high performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) was applied for the simultaneous determination of 21 pesticide residues in *Cornus officinalis* Sieb. et Zucc., a food-medicinal homologous substance. Additionally, commercial samples were tested to evaluate their quality. **Methods:** Samples were extracted with acetonitrile-water (10: 5, v/v). The extract was purified by dSPE using 200 mg anhydrous magnesium sulfate + 50 mg primary secondary amine (PSA) + 50 mg C₁₈ as matrix dispersants. Gradient elution was performed using a mobile phase consisting of 0.01% formic acid, 2 mmol/L ammonium formate aqueous solution and 0.01% formic acid in methanol. UPLC-MS/MS with electrospray ionization (ESI) in positive ion mode and multiple reaction monitoring (MRM) was employed for detection. Matrix-matched standard curves were prepared, and quantification was carried out by the external standard method. **Results:** All 21 target pesticides showed good linearity in the concentration range of 2 ng/mL-200 ng/mL ($r \geq 0.994$). The limits of quantification (LOQ) for all pesticides were less than 7.6 $\mu\text{g}/\text{kg}$. At three spiked levels (5 $\mu\text{g}/\text{kg}$, 20 $\mu\text{g}/\text{kg}$, and 50 $\mu\text{g}/\text{kg}$), the recoveries of the 21 pesticides ranged from 72.24% to 115.0%, with relative standard deviations (RSDs) of 0.40% to 9.7%. **Conclusions:** The developed method meets the requirements for pesticide residue levels of insecticides, making it suitable for rapid confirmation and quantitative analysis of 21 pesticide residues in *Cornus officinalis* Sieb. et Zucc.



1 Introduction

Cornus officinalis Sieb. et Zucc. (abbreviated as *Cornus officinalis*) is a dried and mature fruit of the *Cornus officinalis* plant in the Cornaceae family. With the effects of nourishing the liver and kidneys, reducing astringency, and solidifying dehydration, it is applied for dizziness, tinnitus, waist and knee pain, impotence and spermatorrhea, enuresis and frequent urination, metrorrhagia and leukorrhea, excessive sweating, and internal heat to quench thirst [1]. Modern pharmacological research showed that the active substances in *Cornus officinalis* have a variety of pharmacological effects such as anti-tumor, hypoglycemic, antiarrhythmic, anti-inflammatory, antioxidant, bone metabolism regulation, etc. [2-8], which are used to prevent and treat diabetes [9], Alzheimer's syndrome [10,11], etc. As a substance used for both medicinal and food purposes, *Cornus officinalis* has a wide range of applications in health products and food, such as *Cornus officinalis* decoction pieces, *Cornus officinalis* wine, *Cornus officinalis* beverages [12], etc. With the vigorous development of the big health industry, the demand for *Cornus officinalis* in the market has increased dramatically. During the planting and growth process, *Cornus officinalis* is prone to pests and diseases. Therefore, the application of insecticides, fungicides, etc. is necessary for prevention and control during its growth, harvesting, and storage. This poses a safety hazard of pesticide pollution to the raw materials of *Cornus officinalis* and affects industrial development, necessitating effective monitoring methods for pesticide residues such as *Cornus officinalis*.

At present, there are numerous reports on the detection of insecticide and fungicide residues in medicinal plants using ultra-high performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) [13-16], but research has mainly focused on roots, stems, and flowers, with less

attention paid to fruits. The medicinal parts of *Cornus officinalis* are fruits, which are different from other medicinal parts due to their abundant interfering substances such as pigments and carbohydrates; research on pesticide residues in *Cornus officinalis* is relatively limited. Current methods primarily use gas chromatography (GC) and gas chromatography-mass spectrometry (GC-MS), while ultra-performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) has not yet been employed. This could serve as a novel approach for studying pesticide residues in *Cornus officinalis*. Thus, there are higher demands on detection techniques that may provide reference value for similar research in the later stage. Based on the current situation of pests, diseases and pesticide usage during the cultivation process of *Cornus officinalis* [17-19], and as per the current effective pesticide residue detection standards, this study aimed to optimize the extraction and purification conditions of the target substances, and establish an UPLC-MS/MS analysis method suitable for the pesticide residues (including 21 common pesticides) in the dried fruit flesh of *Cornus officinalis*, in order to ensure the safety of *Cornus officinalis* products.

2 Materials and methods

2.1 Instruments

AB SCIEX AB-5500 QTRAP UPLC-MS/MS equipped with an ESI electrospray ion source (USA); high-speed refrigerated centrifuge (Thermo Fisher, USA); Milli-Q Gradient ultrapure water system (Millipore, France); 0.22 µm organic filter membrane (Anpel, Shanghai); Shimadzu LC-30AD UPLC system (Japan); XPE-205 electronic balance (Mettler, Switzerland).

2.2 Reagents

Acephate (30560-19-1, BePure-20003-10 mg); Aldicarb (116-06-3, BePure-20179YM); Aldicarb sulfone (1646-88-4, BePure-20183); Aldicarb

sulfoxide (1646-87-3, BePure-20185-10 mg); Bifenthrin (BePure-20631, 82657-04-3); Carbofuran (1563-66-2, DRE-GS09010311AL); 3-Hydroxycarbofuran (16655-82-6, BePure-20957); Chlorpyrifos (2921-88-2, BePure-21307); Fenpyroximate (111812-58-9, BePure-22848); Imidacloprid (138261-41-3, BePure-23491); Isocarbophos (24353-61-5, BePure-23601-10 mg); Methomyl (16752-77-5, DRE-L15030000CY); Omethoate (1113-02-6, ZW-HPC-673399-5 mL); Phorate (298-02-2, DRE-GA09010337ME); Phorate sulfone (2588-04-7, BePure-24929); Phorate sulfoxide (2588-03-6, BePure-24931-10 mg); Prochloraz (67747-09-5, BePure-25165); Prochloraz-Desimidazole (139520-94-8, BePure-25299); Prochloraz-Desimidazole-formylamino (139542-32-8, BePure-25301); Pyridaben (96489-71-3, BePure-25335); Pyraclostrobin (1000 µg/mL, Bepure); acetonitrile and methanol (HPLC grade, Merck, Germany); PSA (40 µm, Agilent, USA); C₁₈ (40 µm, Phenomenex); Milli-Q ultrapure water as experimental water; other reagents were of analytical grade. Four batches of *Cornus officinalis* (20220514, 20220728, 20221108, 20230524; origin: Zhejiang) used in the experiment were provided by the Medicinal Materials and Ginseng Branch of Huadong Pharmaceutical Co., Ltd.

2.3 Pretreatment methods of samples

2 g *Cornus officinalis* samples were placed in a 50 mL plastic centrifuge tube, added with 5 mL of water and allowed to stand for 30 minutes. Then, 10 mL of acetonitrile and 1 ceramicbeads were added for 20 min vortex. 7.5 g of powder containing anhydrous magnesium sulfate, anhydrous sodium acetate and sodium chloride (4: 1: 1) were shaken vigorously with samples for 2 min, and centrifuged at 5000 r/min for 5 min.

2 mL of the supernatant after centrifugation was transferred to a plastic centrifuge tube containing purification materials (200 mg anhydrous magnesium

sulfate, 50 mg PSA and 50 mg C₁₈). After vortex for 1 minute and centrifugation at 5000 r/min for 5 min, the supernatant was filtered through a 0.22 µm microporous filter membrane, and the filtrate was stored for later use.

2.4 UPLC-MS/MS

Chromatographic conditions: AC Quity UPLC BEH C18 chromatographic column (2.1 mm × 100 mm); Mobile phase A: 0.01% formic acid-2 mmol/L ammonium formate aqueous solution, Mobile phase B: 0.01% formic acid methanol solution; Flow rate: 0.3 mL/min; Gradient elution program: 0.0-1.5 min, 3%-18% B; 1.5-2.5 min, 18%-50% B; 2.5-23 min, 50%-95% B; 23-25 min, 95% B; 25-26 min, 95%-3% B; 26-30 min, 3% B; Column temperature: 35 °C; Injection volume: 2 µL.

MS conditions: Electrospray ionization source mode (ESI); Scanning mode: positive ion, multiple reaction monitoring (MRM) mode; Nebulizer gas pressure: 1.72×10^5 Pa, Auxiliary gas pressure: 1.72×10^5 Pa, Curtain gas pressure: 2.04×10^5 Pa; Ion source temperature: 500 °C; Electrospray voltage: 5500 V. The remaining MS parameters were listed in the [Table 1](#) below.

3 Results

3.1 Selection of MS conditions

21 pesticide single-standard solutions (0.1 mg/L) were introduced into the ion source individually through direct injection, and then the parent ion was fully scanned in positive ion mode or negative ion mode, respectively. The quasi-molecular ion peaks of each target compound were obtained accordingly. Then, using the quasi-molecular ion peak of each target compound as the parent ion, secondary MS scanning was performed to obtain fragment ion information. Subsequently, the relevant parameters of the secondary MS of each target compound, such as collision energy, cone voltage, etc, were optimized.

Finally, MS parameters such as flow rate, ion source temperature, and desolvation gas temperature were optimized under MRM mode. Table 1 displayed the final optimized parameters. Under the above

chromatographic conditions, all 21 target objects exhibited good sensitivity and peak shape. The total ion chromatogram and the MRM plot were shown in Figure 1 and 2, respectively.

Table 1 Mass spectrometric parameters for 21 compounds.

| Compound | Mass-to-charge ratio (m/z) | | Declustering Potential/V | Collision Pressure/V |
|--------------------------------------|----------------------------|--------------|--------------------------|----------------------|
| | Parent ions | Daughter ion | | |
| Acephate | 184 | 143.0 * | 50 | 12 |
| | | 125 | | 25 |
| Aldicarb | 208.1 | 116.1 * | 20 | 11 |
| | | 89 | | 25 |
| Aldicarb sulfone | 210.1 | 118.0 * | 30 | 17 |
| | | 166.1 | | 16 |
| Aldicarb sulfoxide | 207.1 | 132.0 * | 55 | 9 |
| | | 89 | | 20 |
| Bifenthrin | 440.2 | 181.1 * | 40 | 22 |
| | | 166.1 | | 58 |
| Carbofuran | 222.1 | 165.1 * | 70 | 16 |
| | | 123 | | 29 |
| 3-Hydroxycarbofuran | 238.1 | 181.1 * | 70 | 16 |
| | | 163.1 | | 18 |
| Chlorpyrifos | 349.9 | 197.9 * | 75 | 28 |
| | | 97 | | 45 |
| Fenpyroximate | 422.2 | 366.1 * | 90 | 23 |
| | | 135 | | 43 |
| Iimidacloprid | 256.1 | 175.1 * | 45 | 27 |
| | | 209.1 | | 22 |
| Isocarbophos | 231 | 121.0 * | 100 | 26 |
| | | 109 | | 38 |
| Methomyl | 163.1 | 88.0 * | 38 | 12 |
| | | 106 | | 14 |
| Omethoate | 214 | 183.0 * | 60 | 16 |
| | | 109 | | 36 |
| Phorate | 261 | 75.0 * | 51 | 21 |
| | | 47 | | 53 |
| Phorate sulfone | 293 | 97.0 * | 65 | 50 |
| | | 115 | | 35 |
| Phorate sulfoxide | 277 | 199.0 * | 25 | 13 |
| | | 153 | | 19 |
| Prochloraz | 376.2 | 308.0* | 20 | 15 |
| | | 266 | | 22 |
| Prochloraz-desimidazole | 325 | 282.0 * | 90 | 21 |
| | | 284 | | 23 |
| Prochloraz-desimidazole-formyl amino | 535 | 308.0 * | 80 | 19 |
| | | 310 | | 20 |
| Pyridaben | 365.1 | 309.1 * | 77 | 17 |
| | | 147.1 | | 34 |
| Pyraclostrobin | 388.1 | 194.0* | 50 | 47 |
| | | 163.1 | | 32 |

Note: * represents Quantitative ion.

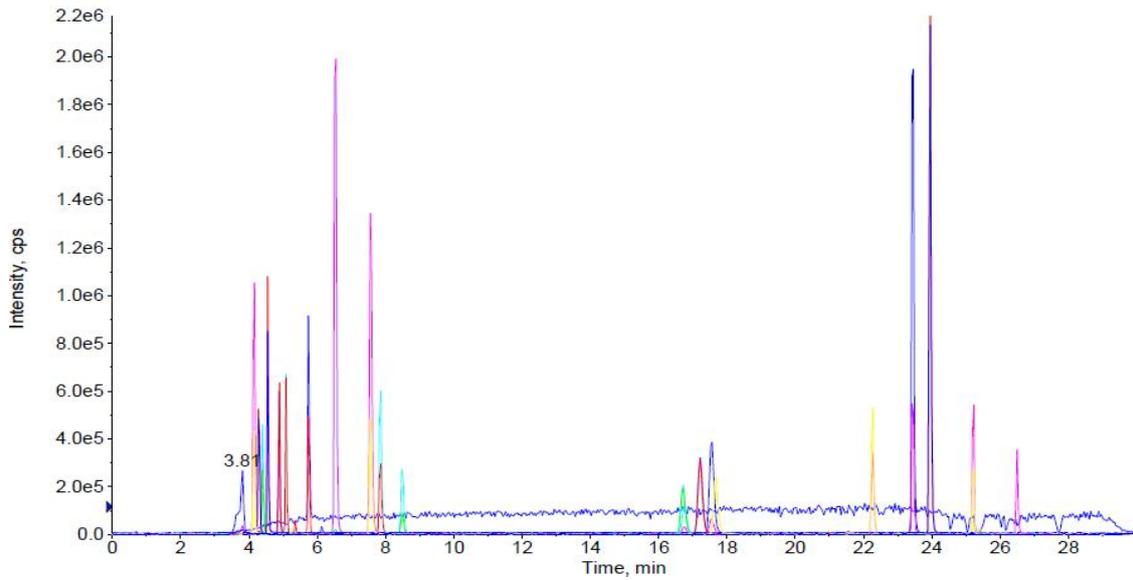


Figure 1 TIC plot of 21 pesticides in blank matrix.

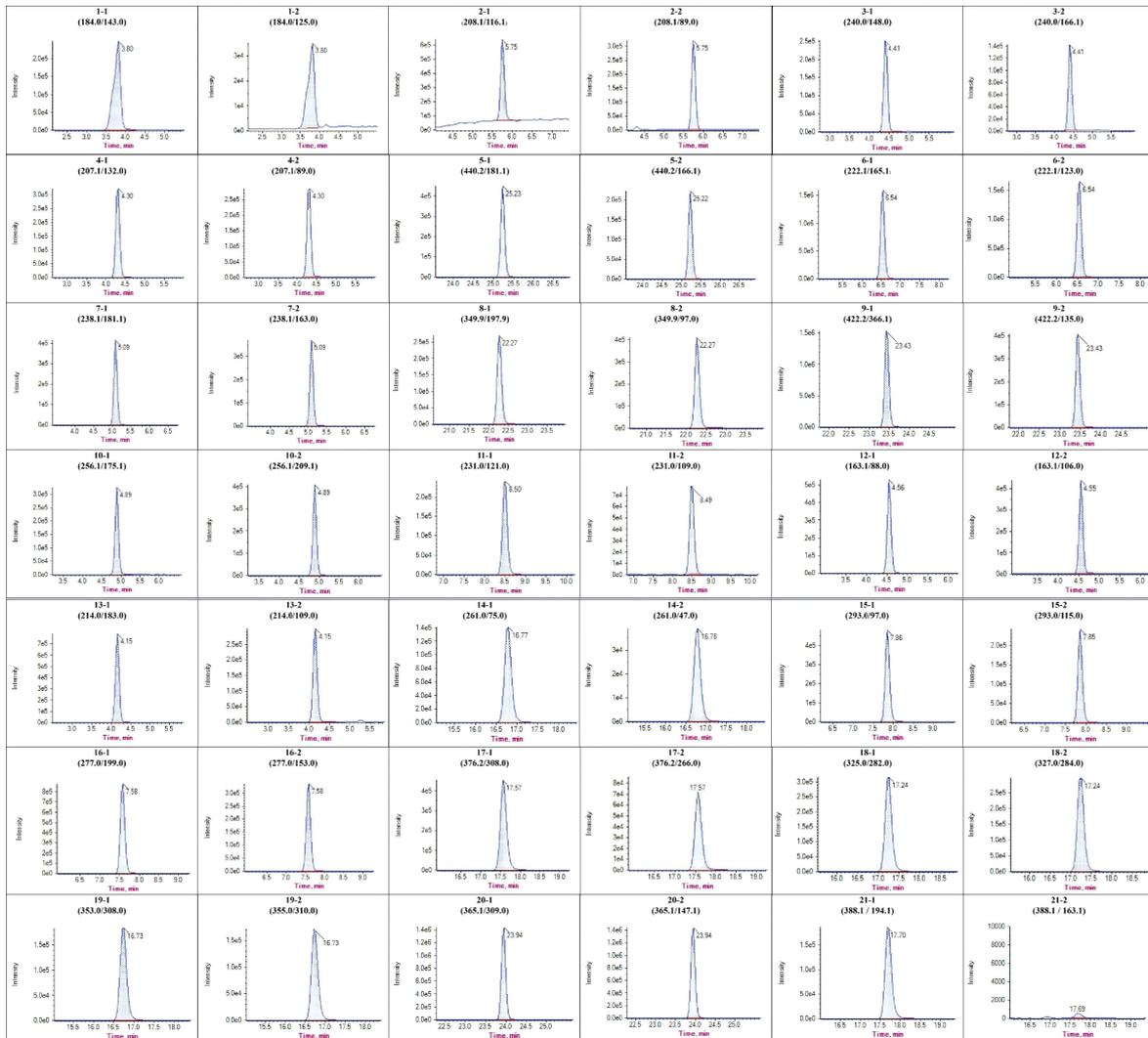


Figure 2 MRM chromatograms of 21 pesticides in blank matrix.

3.2 Optimization of chromatographic conditions

Two organic phases of methanol and acetonitrile were examined. The results showed that when methanol

was used as the mobile phase, the peak shapes of acephate and pyraclostrobin were significantly better than that of acetonitrile. Under the condition of 0.01% formic acid organic phase, the response of some compounds was higher. Under aqueous conditions containing ammonium formate at concentrations of 2 mmol/L, 5 mmol/L and 10 mmol/L, the peak shapes of compounds such as acephate, isocarbophos, prochloraz, and their metabolites were greatly improved. However, high concentrations of salt easily contaminated the MS. In the final experiment, 0.01% formic acid-2 mmol/L ammonium formate was selected as the aqueous mobile phase A, and 0.01% formic acid methanol was as the organic mobile phase B.

3.3 Optimization of sample pretreatment conditions

3.3.1 Selection of extraction solvent

The *Cornus officinalis* sample is a dehydrated and dried sample with relatively low moisture content. Adding a certain amount of water during the extraction process can soften and expand the sample, which contributes to enhancing the extraction efficiency of the target substance by the later extraction solvent. However, as the volume of water in the extraction solvent was increased, the amount of co-extracted interferences was also elevated, leading to a cloudy extraction solution. The commonly used extraction solutions in pesticide residue detection were acetonitrile, acetone, ethyl acetate, n-hexane, dichloromethane, etc. The *Cornus officinalis* sample was a dried fruit pulp sample, comprising abundant pigments, polysaccharides, phenolic acids, and other interfering impurities. Because acetonitrile extracts less sugars and pigments, with the strongest polarity, high extraction efficiency, and low volatility, this experiment selected acetonitrile as the extraction solvent. To investigate the effect of water infiltration volume on the recovery rate of samples, 2 g of *Cornus officinalis* samples were observed after addition of 0

mL, 2 mL, 3 mL, 5 mL, and 8 mL of water under the condition of 10 mL of acetonitrile as the extraction solvent. The results revealed that when the volume of water infiltration was greater than or equal to 5 mL, the sample was completely dispersed, but when the volume was 8 mL, the extraction solution became turbid and difficult to pass through the organic filter membrane. Taking into account factors such as sample dispersion, extraction solution, and filter membrane, a water infiltration volume of 5 mL was deemed more appropriate.

3.3.2 Selection of dispersive solid-phase extraction (dSPE) agent type

Due to the presence of interfering components such as polysaccharides and pigments in *Cornus officinalis*, which can affect pesticide residue detection, the purification effects of two purification methods, namely carbamate column and dSPE, were compared in the experiment. The experiment investigated the recovery rates of 21 target compounds at a concentration of 50 µg/kg in blank *Cornus officinalis* samples were examined. The results unveiled that the recovery rate of the target substance purified by the carbamate column ranged from 26.4% to 81.6%, and there were 7 compounds with a recovery rate of less than 60%. The purification process was time-consuming and inefficient. This experiment chose dSPE as the final purification method.

The commonly used solid-phase extraction materials in pesticide residue analysis include anhydrous magnesium sulfate, PSA, GCB, C₁₈, silica, etc. Based on the characteristics of *Cornus officinalis* samples, combined with the pretreatment methods in current effective national standards and existing literature research results, the experiment used the recovery rates of blank *Cornus officinalis* samples spiked at 50 µg/kg as the evaluation indicator, and examined four different dispersant combinations: (1) 100 mg anhydrous magnesium sulfate + 25 mg PSA + 50 mg

C₁₈, (2) 200 mg anhydrous magnesium sulfate + 50 mg PSA + 50 mg C₁₈, (3) 200 mg anhydrous magnesium sulfate + 100 mg PSA + 100 mg C₁₈, (4) 200 mg anhydrous magnesium sulfate + 50 mg PSA+50 mg C₁₈ + 5 mg GCB. The statistical recovery rate results were shown in Figure 3.

According to the results, in group containing GCB, the recovery rates of bifenthrin, pyraclostrobin, and 3-Hydroxycarbofuran were significantly affected, with recovery rates all below 60%. There was no significant difference in the results between Group 2 and Group 3,

but excessive PSA still adsorbed polar compounds, leading to lower recovery rates for some target compounds. The recovery rates of most compounds in Group 1 were lower than those in Group 2, which was attributed to incomplete removal of water from the extraction solution, resulting in changes in solution polarity and affecting the purification outcome of the target substance. Group 2 (200 mg anhydrous magnesium sulfate + 50 mg PSA + 50 mg C₁₈) was determined as the purification condition for the experiment.

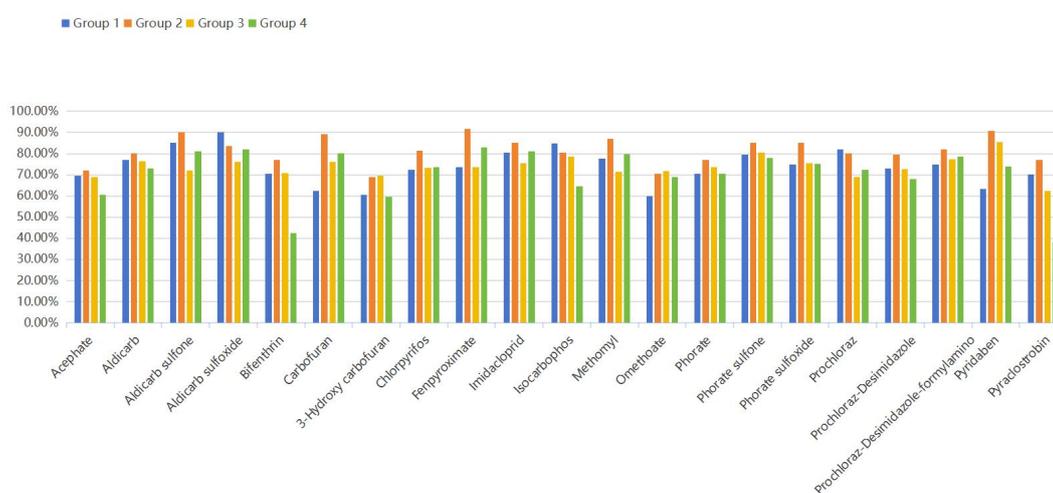


Figure 3 Effects of different dSPE material combination on the spike-and-recovery rates of 21 pesticide.

3.4 Methodological Investigation

3.4.1 Matrix effects (ME)

Using blank sample solution of *Cornus officinalis* and initial proportion mobile phase as solvents, 21 mixed reference stock solutions with concentrations of 50 μg/L were prepared. Under optimized experimental conditions, the evaluation value of ME was calculated by multiplying the peak area ratio of the corresponding target substances by 100%. An absolute value of ME between 80% and 120%, less than 80% and greater than 120% indicated an acceptable ME, a matrix suppression effect, and a matrix enhancement effect.

The experimental results showed that the ME values of 21 target substances ranged from 42.7% to 123.1%, among which imidacloprid and pyraclostrobin had

significant matrix inhibitory effects, while acephate, fenpyroximate, phorate, phorate sulfone, and phorate sulfide had notable matrix enhancing effects. Therefore, to alleviate the impact of ME on the detection of target compounds, the matrix matching standard curve was used in the experiment.

3.4.2 Investigation of linear relation method and limits of quantification (LOQ)

0.10 mL of the 21 reference standard stock solutions mentioned above were accurately aspirated and transferred into a 10.0 mL volumetric flask. The solutions were diluted to the mark with methanol as the solvent. After thorough mixing, the mixed reference solution I (10 μg/mL) was obtained.

Subsequently, 1.0 mL of mixed reference solution I

was precisely pipetted into another 10.0 mL volumetric flask, diluted to the mark with methanol, and thoroughly mixed to obtain mixed reference solution II (1000 ng/mL).

The blank *Cornus officinalis* samples were selected and the blank *Cornus officinalis* solution was prepared according to item "2.3". An appropriate amount of mixed reference solution II was precisely pipetted into a 5 mL volumetric flask, diluted serially with blank *Cornus officinalis* solution to the mark, and mixed thoroughly to obtain matrix standard curve solutions with concentrations of 2 ng/mL, 5 ng/mL, 10 ng/mL, 20 ng/mL, 50 ng/mL, 100 ng/mL, and 200 ng/mL. Analyses were performed in accordance with the experimental conditions under item 2.4. The corresponding peak area data were recorded, with mass concentration (x) as the horizontal axis in the coordinate system, and the recorded peak area (y) as the vertical axis. Linear regression was conducted using data analysis methods. The results proved that 21 pesticides had good linear relationships within the range of 2 ng/mL to 200 ng/mL (all $r > 0.994$, [Table 2](#)).

The LOQ was calculated based on a signal-to-noise ratio (S/N) of 10 using samples with a recovery rate spiked at 5 µg/kg. The results showed that the LOQ for all 21 pesticides were below 7.6 µg/kg ([Table 2](#)).

3.4.3 Precision and stability tests

50 ng/mL level point under the mixed reference curve in item "3.4.1" was measured continuously 6 times according to the experimental conditions in item "2.4". The peak area of the target substance was recorded. The results showed that the RSD range of peak areas for 21 pesticides was 0.168%-0.803% ($n = 6$), indicating good instrument precision.

50 ng/mL level point under the mixed reference curve

in item "3.4.1" was injected at 0, 1, 3, 5, 8, 15, 20, and 24 hours according to the experimental conditions in item "2.4", and the peak area of the target substance was recorded. The results demonstrated that the RSD of peak areas for 21 pesticides was less than 5.8% ($n = 8$), proving that the 21 pesticides were stable within 24 hours.

3.4.4 Spike-and-recovery test and repeatability investigation

In the recovery rate experiment, blank *Cornus officinalis* samples were spiked with reference standard solutions at three levels, 5 µg/kg, 20 µg/kg, and 50 µg/kg. 6 parallel operations were conducted for each concentration level and the RSD values were calculated. The results showed that the recovery rate was between 62.29% and 112.9%, and the RSD was within the range of 0.51% to 8.73%. Six of the aforementioned 50 µg/kg level spiked samples were analyzed under the experimental conditions in item "2.4", and the spiked recovery rates were calculated. The results indicated that the RSD of the spiked recovery rates for the 21 pesticides ranged from 1.1% to 7.6% ($n = 6$), implying the good repeatability of the experimental method ([Table 2](#)).

3.4.5 Sample determination

4 batches of *Cornus officinalis* samples were selected as test objects. The test sample solution was prepared according to the method described in item "2.3", and analyzed under the experimental conditions in item "2.4", to calculate the residual amounts of 21 pesticides in the samples. The results revealed that among the 4 batches of *Cornus officinalis* samples, 1 batch was detected to contain bifenthrin, but the content was below the LOQ. However, no residues of the 21 target pesticides were detected in the remaining three batches of samples.

Table 2 Linear relationships, LOQ, and recovery rates with relative standard deviations at three spiking levels in blank *Cornus officinalis* samples (n = 6).

| Compound name | Linear equation | Correlation coefficient (r) | LOQ µg/kg | 5 µg/kg | | 20 µg/kg | | 50 µg/kg | |
|-------------------------------------|--------------------------------|-----------------------------|-----------|-----------------|------|-----------------|------|-----------------|------|
| | | | | Recovery rate % | RSD% | Recovery rate % | RSD% | Recovery rate % | RSD% |
| Acephate | y = 5.92824e4 x + 3.06087e4 | 0.9999 | 7.6 | 89.2 | 2.0 | 72.3 | 1.1 | 72.3 | 3.5 |
| Aldicarb | y = 7.72403e4 x + 1.13500e5 | 0.9998 | 1.5 | 76.3 | 1.6 | 80.4 | 7.6 | 80.4 | 2.4 |
| Aldicarb Sulfone | y = 3.69524e4 x + -10925.39253 | 0.9998 | 1.5 | 85.3 | 5.7 | 90.2 | 4.5 | 90.0 | 5.4 |
| Aldicarb sulfoxide | y = 4.71851e4 x + 18906.93612 | 0.9999 | 1.5 | 104.6 | 1.7 | 83.2 | 4.7 | 83.2 | 1.8 |
| Bifenthrin | y = 6.17849e4 x + 4.34478e4 | 1.0000 | 7.6 | 81.4 | 2.7 | 77.3 | 1.4 | 77.3 | 2.9 |
| Carbofuran | y = 2.22737e5 x + 3.92725e5 | 0.9998 | 7.6 | 62.0 | 2.8 | 89.8 | 5.9 | 89.8 | 1.8 |
| 3-Hydroxycarbofuran | y = 5.29180e4 x + 26469.71025 | 0.9999 | 7.6 | 69.9 | 7.5 | 69.2 | 2.5 | 69.1 | 2.7 |
| Chlorpyrifos | y = 4.47214e4 x + 7663.58484 | 0.9999 | 7.6 | 82.6 | 0.5 | 81.1 | 4.8 | 81.1 | 2.0 |
| Fenpyroximate | y = 2.21133e5 x + 2.96317e5 | 0.9998 | 7.7 | 64.0 | 2.3 | 91.8 | 2.7 | 91.8 | 3.5 |
| Imidacloprid | y = 4.19566e4 x + 7.34665e4 | 0.9995 | 7.6 | 84.6 | 9.6 | 84.9 | 1.9 | 75.5 | 2.8 |
| Isocarbophos | y = 4.01376e4 x + 4.02697e4 | 0.9999 | 7.6 | 80.4 | 2.5 | 80.4 | 2.1 | 76.4 | 1.9 |
| Methomyl | y = 6.91591e4 x + 23502.03623 | 1.0000 | 7.6 | 77.8 | 2.2 | 86.2 | 1.6 | 86.2 | 1.5 |
| Omethoate | y = 1.08228e5 x + 1.39928e5 | 0.9996 | 7.6 | 60.2 | 1.4 | 84.2 | 1.7 | 70.4 | 1.6 |
| Phorate | y = 3.49628e4 x + -5986.58373 | 0.9996 | 3.8 | 112.0 | 1.4 | 77.8 | 6.3 | 70.2 | 1.7 |
| Phorate sulfone | y = 7.20593e4 x + 9.08992e4 | 0.9997 | 3.8 | 101.2 | 1.4 | 85.5 | 5.8 | 77.6 | 2.3 |
| Phorate sulfoxide | y = 1.47533e5 x + -4.40181e4 | 0.9993 | 3.8 | 75.2 | 2.5 | 84.4 | 2.7 | 84.4 | 1.6 |
| Prochloraz | y = 9.98866e4 x + 6.69619e4 | 1.0000 | 7.6 | 81.9 | 3.0 | 80.2 | 5.0 | 68.7 | 4.5 |
| Prochloraz-desimidazole | y = 7.32777e4 x + 3.95722e4 | 0.9999 | 7.6 | 102.3 | 0.4 | 79.7 | 4.5 | 79.5 | 4.5 |
| Prochloraz-desimidazole-formylamino | y = 4.33871e4 x + 19048.08966 | 0.9999 | 7.6 | 102.6 | 0.5 | 82.1 | 4.0 | 80.3 | 7.1 |
| Pyridaben | y = 2.07024e5 x + 3.38547e5 | 1.0000 | 7.6 | 63.6 | 1.0 | 90.5 | 2.2 | 73.7 | 1.9 |
| Pyraclostrobin | y = 4.14925e4 x + 1.08853e5 | 0.9999 | 7.6 | 81.4 | 9.4 | 77.1 | 2.2 | 77.1 | 2.3 |

4 Discussion

In this experiment, a UPLC-MS/MS method suitable for detecting 21 pesticide residues in *Cornus officinalis* was established by optimizing multiple factors, including chromatographic and mass spectrometric conditions, dispersive solid-phase extraction (d-SPE) purification conditions, and extraction solvents. As *Cornus officinalis* is a flesh fruit sample, its matrix characteristics differ from those of other plant parts commonly tested for pesticide residues. The use of existing methods may lead to issues such as insufficient sensitivity and significant interference, often resulting in low recovery rates for these 21 pesticides. In contrast, the method developed in this study improves and refines current national standards and previously reported analytical approaches. Through systematic optimization of various conditions, this method better meets the detection requirements for flesh fruit samples, achieving recovery rates above 60% for all 21 pesticides, thereby effectively enhancing detection accuracy.

Our method adopts acetonitrile combined with a buffer salt system comprising anhydrous magnesium sulfate, anhydrous sodium acetate, and sodium chloride for extraction. This extraction system destroys the cell structure of *Cornus officinalis* samples, allowing pesticide residues to be dissolved and extracted by acetonitrile. At the same time, the addition of buffer salts regulates the acidity and alkalinity of the extraction system, reducing the degradation of pesticides during extraction.

After purification with 200 mg anhydrous magnesium sulfate, 50 mg PSA and 50 mg C₁₈ purification materials, anhydrous magnesium sulfate can adsorb water in the extraction solution to prevent interference with subsequent detection. PSA specifically adsorbs impurities such as organic acids and sugars in the sample, while C₁₈ can adsorb impurities such as lipids and pigments. Through synergistic effects, the

interference of the sample matrix on the detection results can be reduced.

HPLC-MS/MS was used for detection, together with the electrospray positive ion mode and MRM mode. The electrospray positive ion mode can effectively ionize pesticide molecules in the ion source to form positively charged ions that are easily captured and detected by the instrument. The MRM mode can precisely target pesticide residues, elevate the specificity and sensitivity of detection, and ensure the identification of target pesticide residues at trace levels by monitoring specific parent ions and daughter ions.

Quantification is achieved using a matrix-matched standard curve with the external standard method. Due to the complex composition of the pulp matrix and the presence of matrix effects, direct quantification with a solvent-based standard curve can lead to biased results due to the influence of ME on detection signals. The matrix-matched standard curve is prepared by adding a standard solution to a pre-treated blank *Cornus officinalis* sample, effectively eliminating the influence of ME and ensuring the accuracy of quantitative results.

Our developed method is easy to operate, highly sensitive, efficient, fast, and environmentally friendly. It provides technical reference for the determination of pesticide residues in *Cornus officinalis* and technical support for the safety supervision of medicinal and edible substances such as *Cornus officinalis* that are increasingly in demand. Due to limitations in sample collection and other factors, this study only tested four batches of samples, which presents certain constraints. To obtain more robust and representative experimental data, it would test additional batches in future experiments.

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Conflicts of Interest

The author of this article, Zhaohuan Lou, is a member of the editorial office of this journal. All procedures during the editorial review process were conducted strictly in accordance with the journal's policies, and the author was not involved in handling any part of the process. .

Author Contributions

Substantial contributions to conception and design: L.W. Data acquisition, data analysis and interpretation: Y.Z. and K.L. Drafting the article or critically revising it for important intellectual content: M.C and Z.L. Final approval of the version to be published: All authors. Agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of the work are appropriately investigated and resolved: All authors.

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Availability of Data and Materials

The data presented in this study are available on request from the corresponding author.

Supplementary

Not applicable.

References

- [1] Chinese Pharmacopoeia Commission. Pharmacopoeia of the People's Republic of China, 1st ed; China Medical Science Press: Beijing, China, 2020; p29-30.
- [2] Li YL, Ma DL, Zhang L, et al. Pharmacological effects and mechanism of active components of *Cornus officinalis* on rapidly aging mice in the elderly. *Chinese Journal of*

Pharmacology and Toxicology 2021; 35(9): 652.

[3] Zhang GK, Liu X, Yang C, et al. Screening of anti-allergic active fractions from *Cornus officinalis* and their effects on RBL-2H3 cell degranulation. *Natural Product Research and Development* 2022; 34(7): 1196-1203.

[4] Zhang YH, Zhou XQ, Wu DH, et al. Study on Mechanism of Liuwei Dihuang Pills in Treating Alzheimer's Disease Based on Network Pharmacology. *Chinese Journal of Modern Applied Pharmacy* 2022; 39(2): 154-160.

[5] Nie ZY, Li SM, Wang WS, et al. Mechanism of *Cornus officinalis* polysaccharide on proliferation and apoptosis of gastric cancer cells. *Chinese Pharmacological Bulletin* 2022; 38(2): 222-227.

[6] Ma DL, Luo Y, Li L, et al. Effects and mechanism of iridoid glycosides from *Cornus officinalis* on psychiatric symptoms in tau protein transgenic mice. *Chinese Journal of Pharmacology and Toxicology* 2021; 35(9): 687-690.

[7] Zhang YY, Cui Y, Wang JM, et al. Study on the antidepressant effect of *Cornus officinalis* Sieb. et Zucc. *Journal of Li-shizhen Traditional Chinese Medicine* 2021; 32(7): 1574-1576.

[8] Zhang Y, Yang J, Gao JP, et al. Effect of *Cornus* iridoid glycosides on mouse vascular endothelial cells. *Journal of Biology* 2022; 39(1): 75-79.

[9] Han LZ, Hu KX, Ju HY, et al. The mechanism of action of *Eucommia ulmoides* and *Cornus officinalis* in the treatment of diabetes mellitus based on network pharmacology. *Natural Product Research and Development* 2019; 31(7): 1130-1137+1182.

[10] Wang JJ, Wang ZP, Wang XL, et al. Research progress on chemical composition and pharmacology of *Cornus officinalis*. *Chinese Traditional and Herbal Drugs* 2025; 56(0): 1088-1103.

[11] Peng ZC, He J, Pan XG, et al. Secoiridoid dimers and their biogenetic precursors from the fruits of *Cornus officinalis* with potential therapeutic effects on type 2 diabetes. *Bioorganic & Medicinal Chemistry Letters* 2021; 117: 105399.

[12] Li XJ, Guo J, Liu QY, et al. Research progress and development strategy of *Cornus officinalis* food. *Journal of Shaanxi University of Technology (Natural Science Edition)* 2024; 40(3): 77-83.

[13] Li C, Xiong Y, Gu LH, et al. Analysis and determination of 71 forbidden and restricted pesticide residues in *Citri reticulatae* 'Chachi'. *Chinese Journal of Pharmaceutical*

J. Exp. Clin. Appl. Chin. Med. 2026, 7(1), 15-26

Analysis 2020; 40(5): 843-853.

[14] Zheng JF, Dong YJ, Yu J, et al. Rapid determination of 112 pesticide residues in honey by dispersive solid-phase extraction and gas chromatography-mass spectrometry. *Journal of Zhejiang Agricultural Sciences* 2022; 63(4): 787-791.

[15] Liu LY, Guan X, Wang ZQ, et al. Determination of bromopropylate, spirotetramat and its metabolites in citrus by HPLC-MS/MS. *Agrochemicals* 2022; 61(6): 434-437.

[16] Zhang Y, Wei J, Long JH, Gao D, et al. QuEChERS-UHPLC-MS/MS method for simultaneous determination of 6 pesticide residues in pepper.

Agrochemicals 2022; 61(3): 198-198.

[17] Zhang H, Li T. Common pests of *Cornus officinalis* and their prophylaxis and treatment techniques. *Shaanxi Journal of Agricultural Sciences* 2018; 64(11): 101-104.

[18] Gao X, Xiong KP. Research and development of standardized cultivation techniques for *Cornus officinalis*. *Research and Information on Traditional Chinese Medicine* 2022; 4(1): 25-26.

[19] Ye XF, Fan XZ, Ying ZQ, et al. Resource distribution, as well as development and application of *Cornus officinalis*. *Agricultural Technology Service* 2021; 38(3): 84-87.