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Research Article

Determination of Polar Pesticides Based on a Modified QuPPe with Liquid Chromatography Coupled to Tandem Mass Spectrometry

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An analytical approach for determining polar pesticides using a Hypercarb column that is based on a modified quick polar pesticide (QuPPe) extraction process combined with liquid chromatography-tandem mass spectrometry (LC-MS/MS) was examined. Maleic hydrazide, glyphosate, glufosinate N-acetyl, glufosinate ammonium, fosetyl Al, ethephon, chlormequat chloride, aminomethyl phosphonic acid N-acetyl, aminomethyl phosphonic acid, cyanuric acid, ethylene thiourea, phosphonic acid, propylene thiourea, and acidified methanol solution were used to extract tomato, wheat, olive, sunflower, and herbal tea samples. The amount of solvent, extraction period, and mobile phases used in the experiment were all changed; the analysis included various stationary phases. The method was validated in five matrices spiked at 0.01 and 0.05 mg/kg in accordance with the EU guidance document SANTE/11312/2021 method performance criteria, using six replicates for each concentration for one individual. The limit of detection and limit of quantification (LOQ) values were determined and found to range from 1.82 to 2.44 and 6.07 to 8.13 mg/kg. For all spike levels studied, the approximate recoveries for the pesticides ranged from 85 to 118%, with RSD values of less than 20%. Plant-origin foods from diverse field experiments were effectively processed using the validated approach. This newly developed analytical process can meet the stringent requirements for plant-origin food analysis.

1. Introduction

Pesticide analysis in environmental and biological samples has received considerable attention in recent years, owing to the widespread use of pesticides in agricultural and home applications, as well as their environmental impact [1]. High-quality assessments and food safety standards, especially the control of chemical contaminants, are critical, with a focus on pesticides and crop protection chemicals, which are widely used in modern agriculture and can cause food and environmental problems [2]. Many of these crops are grown and stored using traditional agricultural methods, including the application of pesticides to control insects and other pests [3].

Pesticides released into the environment can be absorbed into various matrices, including water, soil, and crops, posing a significant threat to human health [4]. Pesticides can cause various health problems by accumulating in the human body. Consequently, in recent years, there has been a surge in scientific interest in the potential negative health effects of pesticide exposure in humans [5]. High polar herbicides (HPHs) are commonly used in modern agricultural practices for weed control as well as desiccation in cereal, corn, and rape crops or vegetable production. Nonselective, postemergence, highly polar herbicides have gained popularity in recent years owing to their low cost and broad spectrum [6].

Pesticides with very different polarities are currently available on the market. Gas chromatography (GC) has traditionally been used to determine nonpolar pesticides but is ineffective for highly polar pesticides. Although liquid chromatography (LC) can be used to identify polar

substances, it is not always the best method for this purpose [7]. Pesticides and their metabolites may be present in trace amounts, necessitating the use of sensitive analytical methods [8]. To date, many methods for determining polar pesticides have been developed, including gas chromatography-tandem mass spectrometry (GC-MS/MS), liquid chromatography-ultraviolet (LC-UV), liquid chromatography-fluorescence, and liquid chromatography-tandem mass spectrometry (LC-MS/MS) [9].

In many countries, fosetyl and its aluminum salt (fosetyl-Al) are systemic fungicides used to control oomycetes and ascomycete fungi, as well as some plant pathogenic bacteria, in fruit trees, vegetables, and ornamental plants [10]. Glyphosate (N-phosphonomethyl glycine) is a nonselective postemergence herbicide used to control a wide variety of grass and broad-leaf-weed species in agricultural and industrial settings [11]. It is a broad-spectrum herbicide commonly used in agricultural and forestry settings, as well as in nonagricultural settings such as water systems, parks, road verges, and gardens [12]. Aminomethyl phosphonic acid (AMPA) is a primary metabolite [13]. They are both polar and amphoteric chemicals that are highly soluble in water, which complicates analysis [14].

The general public may be exposed to ethylene bisdithiocarbamates (EBDCs) if they consume food products previously treated with the same. In and on crops treated with the EBDC group of fungicides, EBDC and ethylenethiourea (ETU) residues have been detected. The residual levels change throughout storage, processing, and cooking since the parent chemicals may be converted to ETU by various processes [15]. While the International Agency for Research on Cancer (IARC) categorized ETU as "not classifiable as to its carcinogenicity to people" (IARC 2001), the National Toxicology Program (NTP) of the United States classified the substance as "probably carcinogenic to humans" (NTP 2011). The commission has classified ETU as a category 3 carcinogen ("suspected carcinogens") [16].

Cyanuric acid is an oxytriazine analog of melamine that is produced as a byproduct of the melamine manufacturing process. It is frequently used for sanitizing chemicals and processing animal feed additives. The prevalence of cyanuric acid in food is linked to the use of dichloroisocyanurates as disinfectants for water and product contact items [17].

Maleic hydrazide (MH) is an herbicidal plant growth regulator that stops cell division [11]. It has been used to prevent potatoes, onions, and garlic from sprouting and inhibits tobacco sucker development and weed growth. Colorimetric, chromatographic, electrochemical, and other techniques for determining MH have been developed [18].

Ethephon is a plant growth regulator that boosts sugar content, improves fruit abscission for mechanical harvesting, and promotes or inhibits blooming [19]. Ethephon analysis is required for many foods, including table grapes, under the coordinated community control program specified in Commission Regulation (EC) No. 788/2012 [11]. Chlormequat (Cq) is a 2-chloro-N, N, N-trimethylethylammonium salt. Chlormequat chloride is frequently used as a plant-growth regulator and is widely used to prevent cereals, pears, and grapes from becoming too large. Several pesticides are being examined for modification according to World Health Organization safety guidelines, including Cq [20].

While the fundamental technique presented is acceptable for a wide range of chemicals owing to their unique physicochemical properties (usually high polarity), some compounds, known as single-residue-method compounds, require dedicated techniques because of their poor chromatographic behavior on standard reversed-phase HPLC columns and/or significant decomposition/losses during QuEChERS extraction. The quick polar pesticides method (QuPPe) [21], which uses different chromatographic separation techniques, including hydrophilic interaction chromatography (HILIC), to assess highly polar pesticides in plant-based foods, has been developed by the European Union Reference Laboratories [22].

The Hypercarb column requires a long equilibration of the column before stable retention times can be achieved [23]. Before first use, Hypercarb columns must be thoroughly primed to cover certain active sites on the surface. Priming may be performed by multiple injections of a QuPPe spinach extract or grape skin extract solution [21].

Representative matrices can be used to validate multi- and single-residue methods. At a minimum, one representative commodity from each commodity group must be validated, depending on the intended scope of the method [24]. Using liquid chromatography-tandem mass spectrometry (LCMS/MS) technology, a method for residue-level detection of fosetyl Al, glyphosate, glyphosate ammonium, ethephon, chlormequat, and chlormequat chloride in five matrices was confirmed to represent these groups. Pesticide residues were extracted using a modified QuPPe method that was developed to evaluate polar and non-QuEChERS-amenable pesticide residues. To achieve chromatographic separation, gradient elution was employed, and MS/MS in negative and positive polarity was performed using an electrospray ionization (ESI) probe in the multiple reaction monitoring (MRM) mode.

This study aimed to develop and evaluate a simple and effective simultaneous analytical method for routinely measuring highly polar herbicides and their major metabolites in various plant-based foods. Glyphosate, glufosinate N acetyl, glufosinate ammonium, fosetyl Al, ethephon, chlormequat chloride, aminomethyl phosphonic acid N-acetyl, aminomethyl phosphonic acid, ethylene thiourea, propylenethiourea, and phosphonic acid were used to (i) modify and optimize the QuPPe extraction process, (ii) optimize LC-MS/MS parameters, (iii) investigate possible matrix effects, and (iv) determine the validation approach.

2. Materials and Methods

2.1. Samples. For each sample, 1000 g of tomato, wheat, olive, sunflower, and herbal tea was randomly purchased from bazaar, supermarkets, and greengroceries. The samples were transported to the lab and kept at 4°C until analysis. All samples were homogenized in a blender (R23; Robot Coupe, Jackson, MI, USA). For the analysis, 100 g of the homogenate sample was used.

2.2. Chemicals, Reagents, and Standards. Analyte-gradepesticide standards of maleic hydrazide, glyphosate, glufosinate N-acetyl, glufosinate ammonium, fosetyl Al, ethephon, chlormequat chloride, aminomethyl phosphonic acid N-acetyl, aminomethyl phosphonic acid, cyanuric acid, ethylene thiourea, phosphonic acid, and propylenethiourea were purchased from Sigma-Aldrich (St. Louis, MO, USA). High-performance liquid chromatography (HPLC) grade methanol (MeOH), formic acid (HCOOH), and acetic acid (CH_3COOH) were purchased from Merck (Darmstadt, Germany). A Purelab Option *Q* ultrapure water system from Elga LabWater (Woodridge, IL, USA) was used throughout the study to obtain the HPLC water used during the analyses. Standard solutions of the target compounds were prepared by dissolving an accurately weighed portion of the pesticide (approximately 10 mg powder or liquid) in 10 mL of an appropriate solvent. Stock standard pesticide solutions were prepared at 1,000 mg/L in 10% acetonitrile in water (methanol, only for chlormequat chloride, and maleic hydrazide) and stored at −18°C. Stock solutions were regenerated annually.

2.3. Instruments and Apparatus. The pesticides were extracted using a high-speed blender (R23, Robot Coupe), Waring blender (8011S, Waring, Torrington, CT, USA), and Sartorius analytical balance (model ED323S-CW; Sartorius AG, Göttingen, Germany). For the pesticides under study, chromatographic analyses were performed using a 6470 Triple Qual LC/MS with a Hybercarb 100 × 2.1 mm column [25] (Agilent Technologies, Santa Clara, CA, USA). For gas flow in the LC-MS-MS, a nitrogen generator (Peak Scientific, Scotland, UK) was used at a flow rate of 9.0 L/min. Mobile phase A comprised water with 1% acetic acid and 5% methanol, and mobile phase B comprised acetic acid/methanol (1 : 99) (Table 1). The column temperature was set at 40°C, with a flow rate of 0.2 mL/min. Extracted sample volumes of 5 μL were injected, and from stock standard solutions, the matrix-matched calibration standard solutions (5–100 μm) were prepared in the blank matrix extracts. The electrospray ionization (ESI) interface was operated with positive polarity, and its parameters were as follows: sheath gas flow, 3.0 L/min; sheath gas temperature, 509°C; gas flow, 3.0 L/min; gas temperature, 300°C; and capillary current, 34 nA. The collision-induced dissociation gas was argon (Ar, 99.999%) at 230 kPa. All instrument parameters were controlled using MassHunter Workstation Software® (version B.08.00).

2.4. Extraction Procedure. The tomato, wheat, olive, sunflower, and herbal tea sample treatments were modified from QuPPe protocols [21]. Fresh fruit and vegetable samples were immediately mashed in a blender, and homogenization was performed on dry samples (less than 30% water content) by adding 85 mL of water to 50 g of the material.

Analysis was conducted on a 10 ± 0.1 g sample with high water content and a 5 ± 0.05 g sample with low water content. The amount to be extracted from the homogenized product and the amount of water to be added varied according to the product group. These amounts are listed in Table 2 [22]. Water was added to the sample in a 50 mL tube of the homogenized product and vortexed. Thereafter, methanol (10 mL) containing 1% formic acid was added and mixed for 15 min, after which the mixture was centrifuged at 4,000 rpm for 5 min, and the supernatant was filtered through a 0.45-micron PTFE filter. Finally, 5 μL of elute was subjected to LC-MS/MS.

Table 1: Optimal mobile phase conditions used for chromatographic separation.

Time (min)	%A[a]	%B[b]
0	100	0
10	70	30
11	70	30
18	70	30
19	10	90
22	10	90
22.1	100	0
30	100	0

[a]94% water, 5% methanol, 1% acetic acid. [b]99% methanol, 1% acetic acid.

Table 2: Water content of selected foods and water amount to be added to test portions prior to extraction depending on the analytical approach.

Matrix	Sample weight (g)	Typical natural water content (g/100 g)	Water to be added
Tomato	10	95	0.5
Wheat	5	<10	10
Olive	10	50	5
Sunflower	5	<10	10
Herbal tea	2	<10	10

3. Results and Discussion

3.1. Matrix Effects. It is well documented that the performance of the LC-MS interface is considerably influenced by the composition of the liquid entering the detector, i.e., the type and amount of organic phase modifiers and volatile buffers, as well as the type and amount of sample matrix components play an influential role [26].

In the present study, five different matrices were selected to evaluate matrix effects on tomato, wheat, olive, sunflower, and herbal tea. These fruits and vegetables are representative of the high water content, high oil content, very low and intermediate water content, high starch and/or protein content, low water and fat content, difficult or unique commodities, high acidity of most product types, and high sugar content of many fruits. The linearity of the system was evaluated using matrix-matched calibration and blank extracts spiked with different concentrations (5–100 ng/g). Figure 1 depicts the chromatogram of a tomato sample containing 13 polar pesticides at 10 and 50 ng/g. In the investigated matrices, the lowest spiking level was 0.01 ng/g stances, which corresponded to the limit of quantification (LOQ). The determined LOQ was less than or equal to the MRLs of the target chemicals in the tested matrices, as defined by the European Parliament in Regulation No. 396/2005. To assess repeatability at the 2 concentration levels, 10 duplicates were used (10 and 50 ng/g).

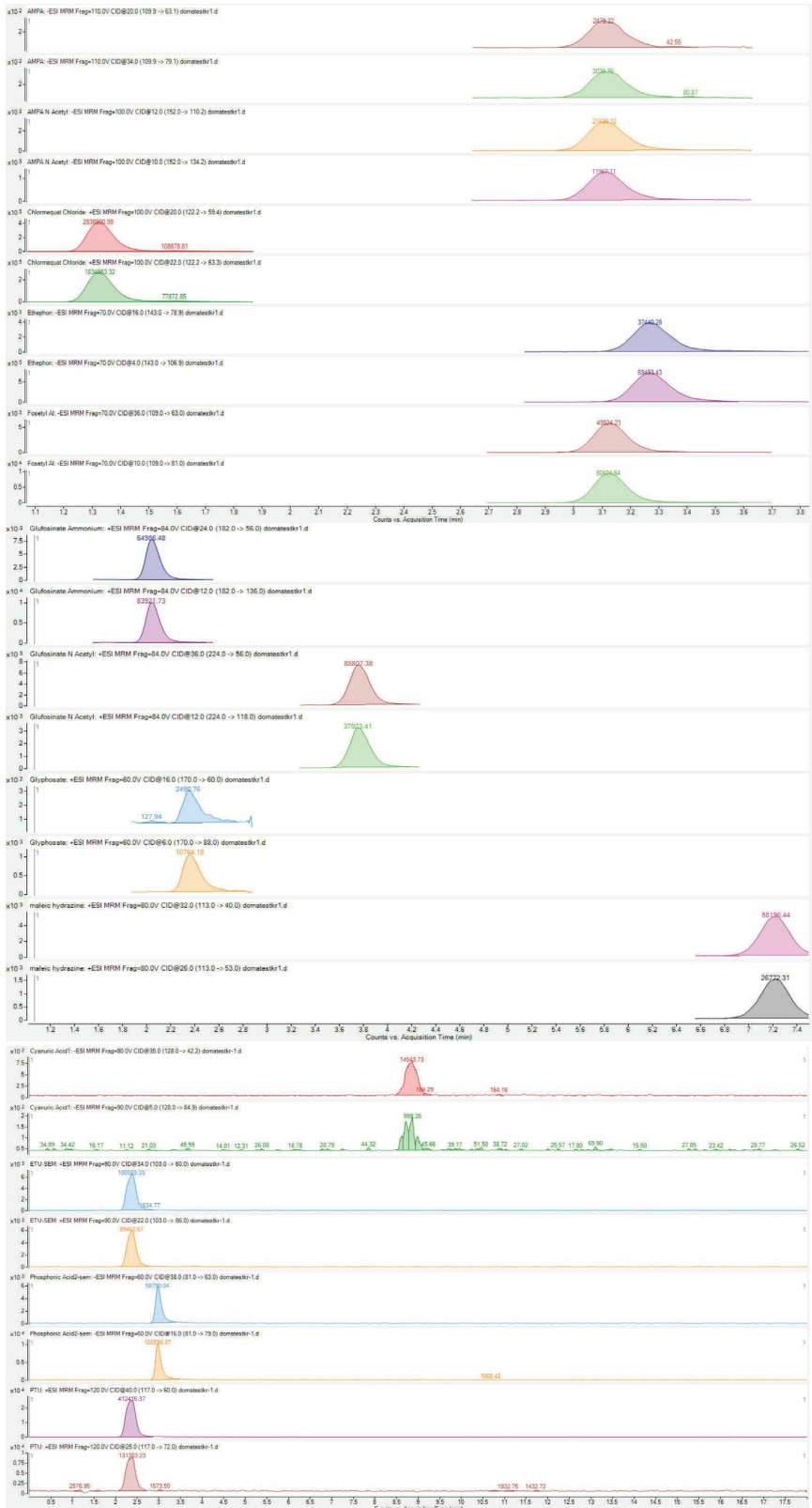

Figure 1: Chromatograms after additional injection of approximately 0.05 mg/kg QuPPe-extracts in tomato.

Calibration curves constructed using matrices with similar physical and chemical properties can be used for other matrix types. For example, because the calibration slopes constructed with olive and sunflower matrices are similar, it is thought that if they are used, they will provide accurate results for the samples. Incidentally, when the

Table 3: LC-MS/MS parameters for the analyzed plant origine.

Compound name	Precursor ion	Product ion	Dwell	Fragmentor	Collision energy	Cell accelerator voltage	Polarity
AMPA	109.9	79.1 63.1	10	110	34 20	3	Negative
AMPA N Acetyl	152.0	134.2 110.2	10	100	10 12	5	Negative
Chlormequat chloride	122.2	63.3 59.4	10	100	22 20	5	Positive
Cyanuric acid	128.0	84.9 42.2	10	90	5 35	5	Negative
Ethephon	143.0	106.9 78.9	10	70	4 16	3	Negative
ETU	103.0	86.0 60.0	10	90	22 34	5	Positive
Fosetyl Al	109.0	81.0 63.0	10	70	10 36	3	Negative
Glufosinate ammonium	182.0	136.0 56.0	10	84	12 24	3	Positive
Glufosinate N Acetyl	224.0	118.0 56.0	10	84	12 36	5	Positive
Glyphosate	170.0	88.0 60.0	10	60	6 16	3	Positive
Maleic hydrazine	113.0	53.0 40.0	10	80	26 32	4	Positive
Phosphonic acid	81.0	79.0 63.0	10	60	16 38	5	Negative
PTU	117.0	72.0 60.0	10	120	25 40	5	Positive

results were evaluated, it was determined that the matrix effect could be reduced by calibrations prepared with the matrix.

3.2. Selection of the LC-MS/MS Conditions. To validate this improved QuPPe approach, researchers have employed the European SANTE/11312/2021 Guidance Document [24]. Validation research examined linearity, the limit of detection (LOD), the LOQ, recovery, precision, and measurement uncertainty. For MS/MS detection, multiple reaction monitoring (MRM) parameters were thoroughly evaluated. Each polar pesticide was fine-tuned to obtain the highest possible sensitivity.

First, both positive and negative electrospray ionization modalities (ESI+ and ESI−) were investigated. Glyphosate, glufosinate N acetyl, glufosinate ammonium, chlormequat chloride, ethylene thiourea, and propylene thiourea are the finest for fosetyl Al, ethephon, aminomethyl phosphonic acid N-acetyl, aminomethyl phosphonic acid, cyanuric acid, phosphoric acid, and maleic hydrazide. Table 3 lists the MS-MS parameters of each analyte.

Precision between days was measured at 10 and 50 ng/g, and spiked samples were examined every day for 5 days. The LC-MS/MS measurement method was chosen according to European Union Regulation SANTE/11312/2021. The calibration curves were linearly fitted with $1/x$ weight and showed good linearity with coefficients of determination (R^2) greater than 0.995. The selectivity of the method was determined by analyzing the reagent blanks and blank samples spiked at the lowest fortification level. The approximate pesticide recoveries ranged from 85 to 118% across all spike levels studied, with RSD values less than 20%. In Alwis et al.'s [27] study, trueness was between 90 and 104%; the values obtained in both studies were in accordance with SANTE/11312/2021.

3.3. Optimization of the Extraction Procedure. The LC parameters (mobile phase combination, amount of solvent, and extraction duration) were adjusted to provide the best selectivity and sensitivity. Eluents containing 1% (v/v) acetic acid, as well as methanol, water, and a water-methanol mixture, were tested. The combination of mobile phase A and acid produced peaks with excellent shape. Two different extraction times, 5 and 10 min, were used for modification since extraction time is an important parameter that strongly influences the amount of analyte recovered. Recovery rates and relative standard deviations were calculated to determine the optimal extraction time. After 10 min, the homogeneous mixture was found to perform better. The amount of solvent was changed based on the initial procedure, and acetonitrile was used in subsequent trials. To reduce the problem of peak tailing, 1% formic acid in the volume ratio was added to methanol. Thirteen polar pesticides were evaluated.

Table 4: Average recoveries (R), repeatability and reproducibility (intraday and interday) of polar herbicides.

Pesticide	Intra-day				Inter-day				
	Recovery (%)	Repeatability (% RSD. $n = 10$)	Recovery (%)	Repeatability (% RSD. $n = 10$)	Recovery (%)	Reproducibility (% RSD. $n = 10$)	Recovery (%)	Reproducibility (% RSD. $n = 10$)	Measurement uncertainty (%)
	0.01 (mg/kg)		0.05 (mg/kg)		0.01 (mg/kg)		0.05 (mg/kg)		
Tomato									
AMPA	114	4.83	117	13.84	99	2.96	87	19.31	46
AMPA N acetyl	115	1.21	119	1.98	110	7.89	93	19.06	44
Chlormequat chloride	114	1.98	115	0.89	93	15.96	107	6.01	36
Cyanuric acid	93	5.73	101	3.31	108	7.61	103	4.36	20
Ethephon	115	2.78	116	2.45	102	13.29	88	18.00	45
ETU	89	11.19	110	3.42	96	15.36	106	9.27	36
Fosetyl Al	118	1.98	117	2.29	109	8.89	98	19.82	45
Glufosinate ammonium	116	2.17	116	1.54	106	9.26	92	19.77	44
Glufosinate N acetyl	97	4.11	111	2.66	97	11.82	92	18.15	39
Glyphosate	104	9.22	100	7.55	102	11.09	95	17.74	37
Maleic hydrazine	101	6.53	116	2.08	99	14.50	102	17.41	42
Phosphonic acid	89	7.99	101	3.52	100	15.16	105	6.93	32
PTU	102	12.53	114	2.60	109	9.04	110	7.58	30
Wheat									
AMPA	111	5.77	92	3.18	95	14.71	91	18.93	43
AMPA N acetyl	115	2.53	90	1.61	110	5.83	97	17.24	37
Chlormequat chloride	108	1.44	106	1.59	94	10.87	105	2.39	22
Cyanuric acid	94	8.50	99	3.52	10	9.44	105	4.51	22
Ethephon	116	2.51	103	1.50	104	12.16	92	19.61	43
ETU	92	7.89	113	3.52	96	14.77	105	7.78	34
Fosetyl Al	117	1.40	111	0.98	111	7.23	98	18.43	40
Glufosinate ammonium	116	2.21	108	1.08	106	9.32	95	17.50	38
Glufosinate N acetyl	114	4.06	95	1.33	101	12.83	90	17.79	41
Glyphosate	107	5.36	86	3.06	102	5.93	94	18.15	36
Maleic hydrazine	113	2.84	101	2.19	99	19.15	98	14.65	43
Phosphonic acid	91	8.29	97	3.40	100	13.23	101	9.69	30
PTU	99	10.12	115	2.91	93	9.53	107	7.34	29
Olive									
AMPA	111	5.50	96	3.60	97	14.65	88	15.10	39
AMPA N acetyl	114	3.08	92	2.41	108	5.95	94	19.45	39
Chlormequat chloride	102	2.20	106	0.63	91	9.04	103	2.44	19
Cyanuric acid	108	6.05	102	2.98	103	11.53	104	7.15	26
Ethephon	113	4.12	104	1.47	102	13.88	89	18.73	42
ETU	113	4.95	114	2.29	100	16.43	106	10.17	39
Fosetyl Al	114	3.80	110	0.70	110	8.66	94	17.63	39
Glufosinate ammonium	115	2.85	112	1.06	106	9.82	93	17.95	40
Glufosinate N acetyl	107	3.42	94	4.31	99	15.41	89	17.06	41
Glyphosate	107	6.10	93	2.91	100	6.19	93	16.95	33
Maleic hydrazine	111	8.03	101	1.69	98	16.66	97	13.86	39

Table 4: Continued.

Pesticide	Intra-day				Inter-day				
	Recovery (%)	Repeatability (% RSD. $n = 10$)	Recovery (%)	Repeatability (% RSD. $n = 10$)	Recovery (%)	Reproducibility (% RSD. $n = 10$)	Recovery (%)	Reproducibility (% RSD. $n = 10$)	Measurement uncertainty (%)
	0.01 (mg/kg)		0.05 (mg/kg)		0.01 (mg/kg)		0.05 (mg/kg)		
Phosphonic acid	99	8.64	101	4.43	105	14.29	101	6.28	26
PTU	94	9.38	115	2.12	98	11.02	106	6.94	30
Sunflower									
AMPA	109	8.86	93	3.76	99	13.77	88	8.59	43
AMPA N acetyl	96	4.60	86	1.13	102	10.16	95	17.92	38
Chlormequat chloride	85	0.96	112	0.85	85	3.33	105	4.51	26
Cyanuric acid	105	6.85	112	4.24	102	12.09	107	7.85	30
Ethephon	96	3.99	102	1.69	99	15.41	91	19.89	43
ETU	112	3.62	116	2.59	100	13.34	106	9.04	35
Fosetyl Al	109	0.55	115	0.95	110	7.74	97	19.81	41
Glufosinate ammonium	111	1.22	109	0.71	104	9.90	94	16.98	37
Glufosinate N acetyl	115	3.35	95	3.61	99	15.62	89	18.16	45
Glyphosate	103	7.12	87	3.55	99	6.94	92	14.53	32
Maleic hydrazine	106	2.51	86	5.54	100	16.35	94	17.71	44
Phosphonic acid	94	8.18	101	3.74	99	12.77	103	6.60	27
PTU	95	9.27	113	3.82	99	10.24	109	8.45	29
Herbal tea									
AMPA	103	8.01	91	3.73	96	12.33	88	19.22	41
AMPA N acetyl	88	2.64	88	2.05	105	11.97	91	18.68	42
Chlormequat chloride	86	1.16	109	0.54	89	2.33	105	3.40	21
Cyanuric acid	109	9.73	104	3.70	106	4.35	104	10.40	25
Ethephon	95	2.26	102	1.61	101	12.60	91	19.44	40
ETU	113	2.33	115	2.56	99	16.20	106	10.26	38
Fosetyl Al	115	1.15	113	0.95	111	7.73	99	19.78	42
Glufosinate ammonium	108	1.35	109	0.85	105	7.84	91	18.69	37
Glufosinate N acetyl	115	2.98	97	1.82	101	13.25	90	17.77	41
Glyphosate	95	3.96	87	3.11	99	8.66	93	17.83	37
Maleic hydrazine	109	5.02	98	3.31	101	14.61	96	14.56	36
Phosphonic acid	100	10.49	98	3.08	101	10.35	98	7.18	26
PTU	98	13.72	114	1.03	101	8.96	108	7.79	29

3.4. Method Validation. In the present study, the chromatographic and MS/MS settings were adjusted to achieve sufficient sensitivity, reliable target chemical detection, and short analysis time. No interference or less than 30% LOQ at the retention time of all target analytes in the blank matrix indicated that the method was selective. The LOD and LOQ were based on 10 replicates for each matrix and analyte and ranged from 1.82 to 2.44 and 6.07 to 8.13 mg/kg, respectively. Precision and reproducibility determinations (the precision of the method and the instrumental technique) were performed using five replicates at two different concentration levels for each matrix. Chromatograms of the field-treated samples are shown in Figure 1. The RSD values for 13 highly polar pesticides in terms of repeatability and reproducibility ranged from 0.54 to 13.84 and 2.33 to 19.89, respectively, at low and high fortification levels (Table 4). Golge [28] reported that repeatability at low and high concentration ranges was between 2.07 and 15.56% and between 1.57 and 6.78%, respectively.

The overall results from a study by Adams et al. [29] supported the application of LC-MS/MS as a multiresidue detection method, with successful validations for 12 analytes in the cereal matrix and 13 analytes in the grape matrix. In our study, the method was validated in five spiked matrices, and 13 pesticide residues were successfully validated.

The average recovery rates were 70–120% for all investigated analytes, as recommended by the SANTE guidelines [24]. For accuracy assessment, independent verification of method performance was evaluated using a proficiency test in soya beans (dried). In the present study, 2 of the 12 pesticides in the FAPAS sample were scanned using MRM functions. To evaluate pesticide residues, matrix-matched calibration curves were used, which were established using a blank FAPAS sample of soya beans. All the scanned pesticides within the analytical scope of the method were correctly identified with excellent quantitative results; respective z-score values of glyphosate and AMPA were found to be 0.8 and −0.9, rendering the obtained results more than satisfactory for the accuracy assessment ($|z| < 2$). The analysis of these samples demonstrated that this method is acceptable for extracting plant-origin food matrices.

4. Conclusion

The increasing human population has led to greater crop production, so there is a significant increase in pesticide application worldwide [30]. The results presented in this study support the viability of an alternative approach for identifying polar pesticides. The validation results show that the method is satisfactory in terms of sensitivity (LODs below 6.07 ng/g), accuracy (with relative recoveries of 85% to 118% in all cases), and precision (RSD below 20%). Method validation parameters, including linearity, matrix effect, precision and accuracy, LOD, and LOQ, showed that the developed method met the requirements for pesticide residue analysis.

The modified QuPPe method followed by LC-MS/MS was successfully validated for detecting and accurately quantifying 13 polar pesticide residues in tomato, wheat, olive, sunflower, and herbal tea. The LOQs were well below the EU MRL for all the polar analytes. The resulting procedure was suitable for detecting polar pesticides in various food samples. In this study, we observed that the matrix effect did not negatively affect the method. The proposed modified method meets the European Union criteria and maximum residue levels.

Although the described methodology has similar sensitivity to previously published LC-MS based methods, the use of different mobile phase combination, solvent amount, and extraction time approach is a clear advantage over them. The presented methodology allows the evaluation of a wide range of pesticides used for different purposes and belonging to different classes. The list of target analytes can be expanded with additional pesticides from current validated chemical classes. In this context, it is always advisable to evaluate the method performance of newly introduced pesticides. This method has many advantages, such as being sensitive, fast, simple, inexpensive, and does not require derivatization.

Data Availability

The data supporting the current study are available from corresponding author upon request.

Conflicts of Interest

The authors declare that there are no conflicts of interest.

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