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Modified QuEChERS Method for the Determination of Multiclass Pesticide Residues in Fruit Samples Utilizing High-Performance Liquid Chromatography

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Abstract A modified quick, easy, cheap, effective, rugged, and safe method (QuEChERS) followed by high-performance liquid chromatography (HPLC) with variable wavelength detector (VWD) has been developed for the quantitative determination of six multiclass pesticide residues including atrazine, ametryn, and terbutryn among herbicides; methidathion and carbaryl among insecticides; and chlorothalonil which is a fungicide. The QuEChERS extraction method developed was aimed to extract and preconcentrate the target analytes from selected fruits such as tomato, watermelon, and papaya samples. Various experimental parameters affecting the extraction efficiency of the method including the use of dispersive solid-phase extraction (d-SPE) cleanup, types and amount of salts, sample size, and composition as well as volume of the extraction solvent, acetonitrile, were optimized. Under the optimum experimental conditions, matrix-matched calibration curves were constructed using the tomato sample as the representative matrix and good linearity, over wide concentration ranges, was obtained with a coefficient of determination (r^2) of 0.990 or better. The limits of detection (LOD) and quantification (LOQ) of the proposed method were in the ranges of 1.7–3.3 and 5.8–11.1 μg kg^{-1}, lower than the maximum residue limits set by the European Union for the raw fruits, such as tomato, watermelon, and papaya. The relative standard deviations (RSDs) of the intra- and inter-day precision studies were varied over the range of 0.2–11.7 %. The proposed method was successfully applied to different fruit samples, and satisfactory recoveries, ranging from 78 to 118 %, were obtained.

Keywords QuEChERS · Fruit samples · Multiclass pesticide residues · High-performance liquid chromatography

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Introduction

The use of chemical pesticides in agricultural crops is becoming vital for controlling pests that greatly affect the yields, in addition to improving the quantity and quality of the products that reach the consumer (Ortelli et al. 2004). Intensive and widespread uses of pesticides, on the other hand, are known to cause undesired contamination of the atmosphere, environmental waters, soils, and agricultural products, and subsequently identified to have toxic effects on human health and biological systems. As a consequence, raw fruits and vegetables as well as their processed products such as juices could also be contaminated by pesticide residues directly and/or indirectly from polluted soils or surface and ground waters and hence could give rise to serious risks of health and safety (Tadeo et al. 2000), which may create great concerns among the legislative bodies. For instance, the European Union (EU) has set the maximum residue limits (MRLs) for pesticides in various agricultural products including fruits and vegetables. Specifically, for the target pesticides, the MRLs in tomato, papaya, and watermelon fruits are in the range of 10–20, 000 μg kg^{-1} (EU Pesticides Database).

The analysis of pesticide residues is commonly carried out in a sequence of several steps, including extraction of the

target analytes from the sample matrix, extract cleanup, and preconcentration prior to their determination (Romero-González et al. 2008). Regardless of their enormous disadvantages, such as time consuming, labor intensive, use of large sample size, and high volume of expensive and/or hazardous organic solvents, traditional sample preparation techniques including liquid–liquid extraction (LLE) (Kolbe and Andersson 2006; Sannino 2007) and solid-phase extraction (SPE) (Topuz et al. 2005; Yang et al. 2011) have commonly been used for the analysis of pesticide residues from fruits and vegetables.

During the last couple of decades, great efforts have been made to introduce new, miniaturized, and simplified methodologies in order to overcome the drawbacks of these classical sample preparation methods, LLE and SPE. As a consequence, several sample preparation techniques including solid-phase microextraction (SPME) (Zambonin et al. 2004; Sagratini et al. 2007; Cortes-Aguado et al. 2008), matrix solid-phase dispersion (MSPD) (Albero et al. 2003; Albero et al. 2004; Chu et al. 2005), single-drop microextraction (SDME) (Xiao et al. 2006; Zhao et al. 2006), and supported liquid membrane (SLM) (Khrolenko and Wieczorek 2005) have been developed and applied to the analysis of multiclass pesticide residues in different food samples. However, these techniques involve multistep sample pretreatment procedures. Recently, another environmentally benign technique, called quick, easy, cheap, effective, rugged, and safe (QuEChERS) methodology, has been developed for the analysis of pesticide residues in complex samples (Anastassiades and Lehotay 2003; Lehotay et al. 2005; Lehotay 2007). The technique involves two steps: extraction/partitioning step using acetonitrile as extraction solvent and cleanup step using dispersive solid-phase extraction (d-SPE). Since its introduction, QuEChERS has received significant popularity as a method of choice for the analysis of multiclass pesticide residues in food samples such as fruits and vegetables (Melo et al. 2013; Romero-González et al. 2008; Wilkowska and Biziuk 2011; Fernandes et al. 2011; Bruzzoniti et al. 2014). Moreover, due to its enormous advantages in terms of simplicity, rapidity, selectivity, and flexibility, the technique has been accepted and registered as an official method by the Association of Official Analytical Chemists (AOAC) International, AOAC official method 2007.01, and European Committee for Standardization (CEN) standard method, CEN standard method EN 15662, of course with minor modification of the original version, for the analysis of pesticide residues in fruits and vegetables (Lehotay 2007; González-Curbelo et al. 2011).

The QuEChERS procedure involves the use of an equal proportion of the sample and the extraction solvent, i.e., 1 mL acetonitrile per 1 g sample, during the first liquid–liquid partitioning step (Anastassiades and Lehotay 2003; Lehotay et al. 2010; Cieślik et al. 2011; Wilkowska and Biziuk 2011; Sinha et al. 2012). As a result, the method does not involve preconcentration (enrichment) of the target analytes (Melo et al. 2013). Though their aims were not for preconcentration, some works that made use of acetonitrile to the sample in 1:2 ratios have also been reported (Arroyo-Manzanares et al. 2013; Sampaio et al. 2012). The analysis of the final extract of the sample has usually been carried out using sensitive analytical techniques such as gas and/or liquid chromatography coupled with mass or tandem mass spectrometry detectors (Arroyo-Manzanares et al. 2013; Carneiro et al. 2013; Sinha et al. 2012; Cieślik et al. 2011; Pareja et al 2011; Cortes-Aguado et al, 2008).

Moreover, the QuEChERS methodology has also been used in combination with the traditional dispersive liquid–liquid microextraction (DLLME) (Chen et al. 2013; Melo et al. 2012; Cunha and Fernandes 2011) and ultrasound-assisted dispersive liquid–liquid microextraction based on solidification of floating organic droplet method (UA-DLLME-SFO) (You et al. 2013) for the analysis of pesticide residues in different food samples. The purpose of combining the QuEChERS methodology with DLLME was to increase the enrichment factor of the extraction processes. However, a procedure that could increase the enrichment factor by using the QuEChERS procedure alone has significant advantages including minimization of organic solvent volume, saving the time required for sample preparation, and the possibility of combining the extraction method with readily available instruments such as high-performance liquid chromatography (HPLC)–variable wavelength detector (VWD).

In the present study, a modified QuEChERS methodology that is aimed to greatly decrease the volume of acetonitrile while enriching the selected pesticides in fruit samples in combination with HPLC–VWD has been proposed. The pesticides studied comprised of atrazine (atraz), ametryn (amet), and terbutryn (terb) among herbicides; methidathion (meth) and carbaryl (carb) among insecticides; and chlorothalonil (chlor) which is a fungicide, and these pesticides have widely been used in Ethiopia for the control of pests. The fruit samples selected for the study include tomato, watermelon, and papaya which were purchased from local markets in Addis Ababa, Ethiopia.

Materials and Methods

Chemicals and Reagents

All chemicals and reagents used in this study were of analytical grade, and the solvents were of HPLC grade. Acetonitrile purchased from Ashland Chemical (S. Giuliano MI, Italy) and ultrapure water obtained after purification with double distiller A8000 Aquatron water still (Bibby Scientific Ltd, Staffordshire, UK) were used throughout the study. Anhydrous magnesium sulfate ($MgSO_4$) purchased from

Fisher Scientific Company (USA), sodium chloride (NaCl), glacial acetic acid (CH_3COOH, 100 %), and anhydrous sodium acetate (CH_3COONa, 99 %) obtained from BDH Laboratory Supplies (Poole, England) were used during the routine experiments.

Analytical pesticide standards of carbaryl (99.5 %), methidathion (95.2 %), and chlorothalonil (99.3 %) were obtained from Sigma-Aldrich (St. Louis, MO, USA). Atrazine (99.4 %), ametryn (99.3 %), and terbutryn (99.5 %) were purchased from Dr. Ehrenstorfer (Augsburg, Germany). Stock standard solutions containing 1000 mg L^{-1} of each compound were prepared by dissolving appropriate quantities of each standard in acetonitrile and stored in the dark at 4 °C. An intermediate working solution containing 20 mg L^{-1} of each analyte was also prepared in acetonitrile for use during optimization of the extraction parameters.

Instruments and Equipment

Chromatographic analyses were performed using Agilent Technologies 1200 infinity series HPLC, equipped with a quaternary pump, an Agilent 1200 Series Vacuum Degasser, an Agilent 1200 Series Autosampler, and an Agilent 1200 Series UV–Vis Variable Wavelength Detector, all purchased from Agilent Technologies (Germany). Data acquisition and processing were accomplished with LC ChemStation software (Agilent Technologies).

Chromatographic separation was performed using an Eclipse plus C_{18} column (100×4.6 mm I.D., 3.5 μm particle size) obtained from Agilent Technologies. The d-SPE tube used for cleanup was a Supel QuE PSA (EN) tube (containing 150 mg Supelclean™ PSA, 150 mg Discovery® DSC-18, and 900 mg $MgSO_4$) purchased from Sigma-Aldrich (St. Louis, MO, USA). The ultrasonic cleaner Decon®, from Decon Laboratories Limited (Hove, East Sussex); Xcentrifuge, Centurion Scientific Limited (Ford, Arundel, West Sussex); a centrifuge, model 800, Jiangsu Zhenji Instruments Co., Ltd. (Jiangsu, China); 50 mL centrifuge tube (Corning Inc., NY, USA); and a chopper (TangFa, China) were used for sample preparation.

Chromatographic Conditions

The reverse phase separation of the analytes was performed with isocratic elution comprising of 45 % water (solvent A) and 55 % acetonitrile (solvent B) throughout the analysis. Prior to the sample/extract injection, the HPLC column was washed and conditioned with the mobile phase for 15 min. Analysis was performed with a flow rate of 0.5 mL min^{-1}, a column temperature set at 30 °C, an injection volume of 15 μL, and a monitoring wavelength of 224 nm.

QuEChERS Extraction Procedure

Fresh tomato, papaya, and watermelon samples were obtained from a local market in Addis Ababa, Ethiopia. Each sample was washed with tap water to remove any dust particles from the surface. Then, they were cut into pieces with a knife and chopped into fine pieces using a chopper. Ten grams of the chopped samples was weighed in a centrifuge tube and subsequently spiked with appropriate concentrations of the target analyte standard mixtures. The content was then shaken for few seconds and kept to stand for about 15 min to establish equilibration. After the addition of 2 mL acetonitrile, the content was shaken again for few more seconds. This was followed by addition of 4 g $MgSO_4$ and 1 g NaCl to the sample mixture and further shaken vigorously for 1 min (Melo et al. 2013). The content was then centrifuged at 3000 rpm for 10 min. Finally, 1.5 mL acetonitrile extract was transferred to the d-SPE tube containing 75 mg PSA, 75 mg C_{18}, and 450 mg $MgSO_4$. The d-SPE tubes were sealed, shaken for 30 s, and centrifuged at 3000 rpm for 3 min. The resulting extract was taken with a 3-mL syringe and filtered using 0.2-μm nylon filters into a 1.5-mL amber autosampler vial in order to inject 15 μL of it into the HPLC system. Utilizing this procedure, approximately 6 to 12 samples were treated in an hour, with a preconcentration factor of 5.

Results and Discussion

Optimization of HPLC Conditions

In the present study, various compositions of the binary mobile phase including water (solvent A) and acetonitrile (solvent B) were studied in isocratic mode, at a flow rate of 0.5 mL min^{-1}. As a compromise between adequate retention times for the target analytes and efficient separation of the peaks, the mixture of the mobile phase composed of 45 % water and 55 % acetonitrile was found to exhibit the desired separation for all the compounds in less than 20 min.

The effect of the mobile phase flow rate was also studied in the range of 0.3–0.8 mL min^{-1}. It was observed that for all the target analytes, both the retention times and peak widths were lowered with increasing flow rates, in addition to the lowered resolution between carbaryl and atrazine at higher flow rates. Thus, as a compromise, a flow rate of 0.5 mL min^{-1} was chosen as optimum throughout the study. The effect of the injection volume was also investigated over the range of 10–30 μL. It was observed that the peak areas of the target analytes increased with the injection volume; however, above 15 μL, some of the peaks including that of carbaryl and atrazine were relatively broadened and the resolutions between them were found unsatisfactory. Thus, an injection volume

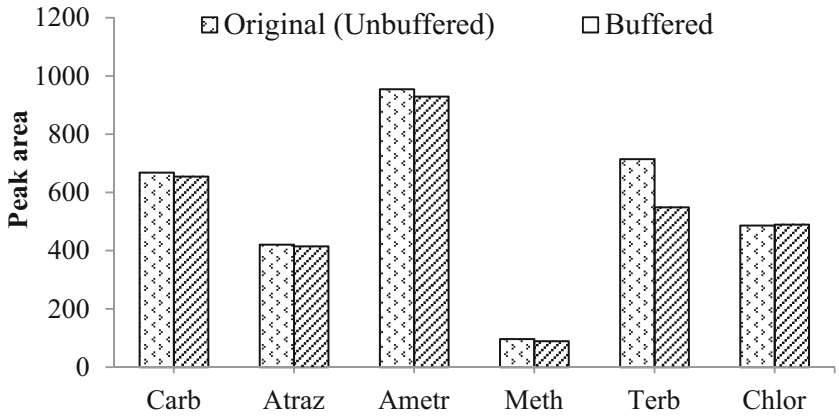

Fig. 1 Comparison of the original (unbuffered) and buffered QuEChERS methods. Experimental conditions: **a** for the original (unbuffered) QuEChERS, 10 g tomato sample extracted with 10 mL acetonitrile, 4 g $MgSO_4$, and 1 g sodium chloride; **b** for the buffered approach, 10 g tomato sample extracted with 10 mL acetonitrile containing 1 % acetic acid, 4 g $MgSO_4$, and 1 g sodium acetate

of 15 μL was selected as a compromise between the sensitivity and adequate peak resolution. The column temperature was also evaluated in the range of 25–35 °C. However, no significant change was observed in the studied temperature range. Thus, the column temperature was set at 30 °C for all the target analytes at 224 nm and VWD monitoring wavelength throughout this work. Under these optimum conditions, the retention times of the analytes were found to be 7.03 min (carb), 7.71 min (atraz), 10.1 min (amet), 11.7 min (meth), 15.4 min (terb), and 18.1 min (chlor).

Optimization of the QuEChERS Procedure

In this study, the QuEChERS method combined with HPLC–VWD has been proposed for extraction and analysis of six multiclass pesticide residues in tomato, watermelon, and papaya samples. In order to obtain the optimal QuEChERS conditions, various parameters affecting the extraction performance of the method such as use of d-SPE for cleanup, type and amount of salts, sample size, and composition and volume of the extraction solvent (acetonitrile) were investigated. All experiments were performed in triplicate by spiking tomato samples with 500 μg kg^{-1} of atrazine, ametryn, terbutryn, carbaryl, and chlorothalonil as well as 1000 μg kg^{-1} of methidathion. The average peak areas of the replicate analysis were considered to evaluate the influence of the experimental parameters on the extraction efficiency of the method.

It is known that the QuEChERS method involves two basic steps: extraction with acetonitrile and partitioning between acetonitrile and the aqueous phase after the addition of NaCl and $MgSO_4$, and d-SPE cleanup procedure utilizing small quantities of SPE sorbents such as primary secondary amine (PSA), C_{18}, and graphitized carbon black (GCB). d-SPE is used as a "chemical filter" to remove matrix interferences without retaining the target analytes (Zhao et al. 2012; Melo et al. 2013). In the current work, the significance of the d-SPE cleanup step was investigated by analyzing the acetonitrile extracts of tomato samples without cleanup and with application of the d-SPE cleanup procedure, utilizing a combination of 75 mg PSA, 75 mg C_{18}, and 450 mg $MgSO_4$. The obtained results demonstrated that the use of d-SPE removes several co-extracts to the extent that they could not be measured in the extracts, and thus, d-SPE cleanup, employing the combination of PSA and C_{18}, was used for further experiments.

During the preliminary experiments, the extraction performance of two QuEChERS methodologies: the original (unbuffered) (Anastassiades and Lehotay 2003) and the buffered (Lehotay et al. 2005) were evaluated. As can be seen in Fig. 1, though the obtained results were similar, utilizing both methodologies for the target analytes such as atrazine, methidathion, and chlorothalonil, relatively higher peak areas

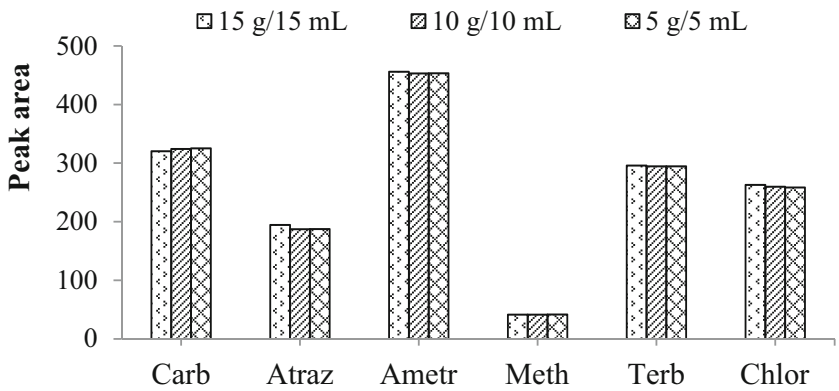

Fig. 2 Effect of the sample size. Experimental conditions: 10 mL acetonitrile, 4 g $MgSO_4$, and 1 g sodium chloride; 2 mL of the supernatant was cleaned up with d-SPE containing 75 mg PSA, 75 mg C_{18}, and 450 mg $MgSO_4$

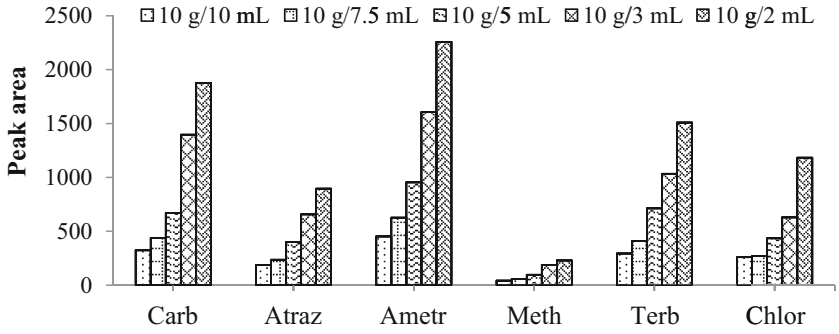

Fig. 3 Effect of the extraction solvent volume. Experimental conditions: 10 g tomato sample, 4 g $MgSO_4$, and 1 g sodium chloride; 1.5 mL of the supernatant was cleaned up with d-SPE containing 75 mg PSA, 75 mg C_{18}, and 450 mg $MgSO_4$

were observed with the original (unbuffered) QuEChERS approach for carbaryl, ametryn, and terbutryn. In the buffered approach, after the addition of the buffering reagents, i.e., 1 % acetic acid and 1 g sodium acetate, the pH of the content was lowered (Machado et al. 2013) and, thus, it was found inconvenient for extraction of basic analytes since they could be ionized at a lower pH. Therefore, the unbuffered QuEChERS version was selected for the further experiments.

The sample size commonly used in QuEChERS is generally 10 g (Anastassiades and Lehotay 2003) or 15 g (Lehotay et al. 2005; Lehotay 2007). However, it is reasonable to decrease the sample size since the amount of extraction solvent and the salt are proportionally minimized. Accordingly, in this study, the effect of different sample sizes with proportional amount of the salt and acetonitrile on the peak area of analytes were investigated. It was found that minimizing the sample size does not affect the peak areas of the target analytes (Fig. 2). Thus, whenever required, any of the sample size can be used without affecting the extraction efficiency of the method. Thus, a sample size of 10 g was selected for subsequent experiments.

Use of a relatively higher volume of acetonitrile is one of the basic drawbacks of QuEChERS. In this study, the effect of the volumes of extracting solvent, acetonitrile, was investigated using different volumes of acetonitrile with a constant mass of tomato samples. As can be seen from Fig. 3, peak areas of the target analytes increased as the volume of acetonitrile decreased and the highest peak areas were observed with 2 mL acetonitrile. The increase in peak areas with decreasing acetonitrile volume may be attributed to the preconcentration of the target analytes. But, further reduction of the volume of the acetonitrile, i.e., below 2 mL, was found inconvenient, since the volume of the extract collected after the first extraction step was too small for the subsequent d-SPE cleanup procedure. Therefore, 2 mL acetonitrile was chosen as the optimum volume for further studies.

On the other hand, use of a lower volume of acetonitrile has the advantage of preconcentrating the analytes of interest, without the application of an additional drying step, which may also cause loss of some target analytes (Melo et al. 2013). Compared to the original QuEChERS approach, the proposed modified procedure has exhibited a better preconcentration (enrichment) factor, and the extract obtained was also directly injected to the HPLC–VWD system without the need for further pretreatment steps.

Analytical Method Validation

Analytical Performance Characteristics

The performance of the proposed QuEChERS extraction technique combined with HPLC-DAD was evaluated utilizing matrix-matched calibration curves established employing the target pesticide-free tomato sample as a representative matrix.

Table 1 Statistical and performance characteristics of the proposed method

Analytes	Linear range (μg kg^{-1})	r^2	LOD (μg kg^{-1})	LOQ (μg kg^{-1})	MRLs (μg kg^{-1})		
					Tomato	Papaya	Watermelon
Carb	6–400	0.998	1.8	5.8	10	10	10
Atraz	11–400	0.995	3.3	11.1	50	50	50
Amet	6–400	0.998	1.7	5.7	–[a]	–[a]	–[a]
Meth	110–1200	0.991	33	110	20	20	20
Terb	11–400	0.999	3.2	10.8	–[a]	–[a]	–[a]
Chlor	10–400	0.998	2.9	9.8	2000	20,000	1000

[a] Not indicated in the EU Pesticides Database

Table 2 Intra- and inter-day precision of the proposed method (% RSD) for the spiked tomato samples

Analytes	Intra-day; RSD (n=6)		Inter-day; RSD (n=9)	
	Level 1	Level 2	Level 1	Level 2
Carb	0.3	0.7	2.9	6.5
Atraz	0.5	0.9	5.7	6.7
Amet	0.2	0.5	10.0	3.7
Meth	3.0	1.4	6.8	9.7
Terb	0.5	0.3	11.2	5.6
Chlor	1.5	0.7	11.7	9.01

Level 1=100 μg kg^{-1} for carb, atraz, amet, terb, and chlor; 300 μg kg^{-1} for meth; level 2=300 μg kg^{-1} for carb, atraz, amet, terb, and chlor; 900 μg kg^{-1} for meth

The calibration curves were constructed by spiking the tomato samples with a mixture of the analytes at six concentration levels: 20, 40, 100, 200, 300, and 400 μg kg^{-1} for carb, atraz, amet, terb, and chlor as well as five concentration levels: 120, 300, 600, 900, and 1200 μg kg^{-1} for meth. Each level was extracted in duplicate (experimental replicates), under the optimum procedure, and each extract was also injected in duplicate (instrumental replicates). Then, calibration curves were obtained by considering the peak areas as the instrumental response versus the analyte concentrations. The coefficients of determinations (r^2) of the analytes were 0.990 or better, which confirmed a good linearity of the analytical method over the concentration range studied. The limits of detection (LOD) and quantification (LOQ) were considered as the minimum analyte concentrations yielding 3 and 10 times the signal-to-noise (S/N) ratio, respectively, and found to be far below the European maximum residue limits (MRLs) set for these analytes in tomato, watermelon, and papaya samples, with the exception of methidathion (EU Pesticides Database). The performance characteristics of the proposed method in the tomato sample are shown in Table 1.

Precision Study

The precision of the proposed method was investigated in terms of repeatability (intra-day precision) and reproducibility (inter-day precision). Repeatability was assessed by spiking the tomato samples at two concentration levels: level 1: 100 μg L^{-1} for carb, atraz, amet, terb, and chlor and 300 μg L^{-1} for meth as well as level 2: 300 μg L^{-1} for carb, atraz, amet, terb, and chlor and 900 μg L^{-1} for meth. Each concentration level was prepared in duplicate (experimental replicates) and was then injected in triplicate (instrumental replicates) on the same day, under the same experimental conditions. Reproducibility was also assayed by spiking tomato samples at the same concentration levels indicated herein for intra-day precision study, during three consecutive days, and each concentration level was injected in triplicate. The results of both intra- and inter-day precision, expressed as relative standard deviations (RSDs) of peak areas, are shown in Table 2. It was found that acceptable precision, i.e., RSD less than 12 %, was obtained in both cases (European Commission 2013, SANCO/12571/2013).

Applications and Recovery Studies

The practical applicability of the proposed method was evaluated by performing recovery studies in three different kinds of fruits including tomato, watermelon, and papaya samples. None of the target analytes was detected in any of these samples. To investigate the applicability of the proposed method to the selected fruit samples, recovery studies were performed at two concentration levels, similar to those earlier used for precision study. Each concentration level was extracted in duplicate, and each was injected in triplicate. Recoveries were calculated by comparing the concentration of the extracted analytes with the initial concentration of the target analytes, spiked to the fruit samples (Carneiro et al. 2013; Lopes et al. 2012; Xiao et al. 2006). Recoveries and the corresponding

Table 3 Percentage recoveries (% R, n=6) of the method for tomato, watermelon, and papaya samples

Analyte	Tomato, % R (RSD)		Watermelon, % R (RSD)		Papaya, % R (RSD)	
	Level 1	Level 2	Level 1	Level 2	Level 1	Level 2
Carb	110 (0.3)	108 (0.7)	109 (7.3)	115 (1.4)	99 (4.5)	118 (4.0)
Atraz	107 (0.5)	81 (0.9)	112 (10.1)	87 (2.2)	114 (5.2)	115 (1.8)
Amet	101 (0.2)	109 (0.5)	101 (3.9)	80 (2.4)	113 (1.1)	113 (0.5)
Meth	102 (3.0)	90 (1.4)	104 (7.3)	105 (4.1)	111 (3.4)	112 (3.4)
Terb	107 (0.5)	98 (0.3)	99 (5.8)	80 (2.1)	117 (3.3)	114 (0.8)
Chlor	85 (1.5)	108 (0.7)	78 (7.7)	84 (3.1)	85 (1.7)	115 (1.6)

Level 1=100 μg kg^{-1} for carb, atraz, amet, terb, and chlor; 300 μg kg^{-1} for meth; level 2=300 μg kg^{-1} for carb, atraz, amet, terb, and chlor; 900 μg kg^{-1} for meth

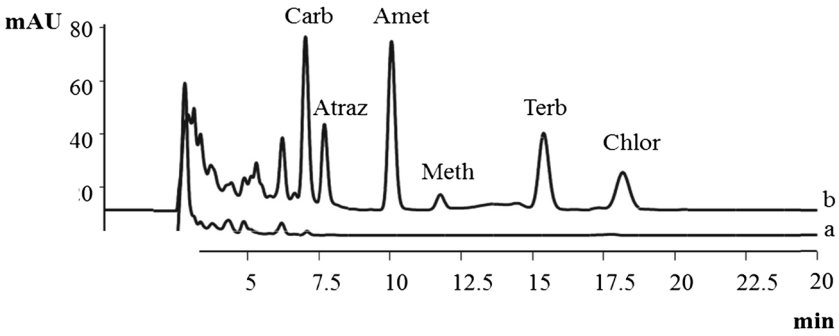

Fig. 4 Typical chromatograms of **a** the blank of tomato sample and **b** the spiked tomato sample with 500 μg kg^{-1} for carb, atraz, amet, terb, and chlor; 1000 μg kg^{-1} for meth

RSD (n=6) of each target analyte in tomato, watermelon, and papaya samples are shown in Table 3. The observed recoveries were in the range of 78–118 % in all the samples. These results were in good agreement with the acceptable recovery range (i.e., from 70 to 120 %) established by the European Commission for pesticide residue analysis in food and feed samples (European Commission 2013, SANCO/12571/2013).

Representative chromatograms of the blank tomato and of the sample spiked with 500 μg kg^{-1} for carb, atraz, amet, terb, and chlor, and 1000 μg kg^{-1} for meth were analyzed by the proposed QuEChERS–HPLC–VWD method under the optimum conditions, shown in Fig. 4.

Conclusions

In this study, a novel analytical method has been proposed for the analysis of six multiclass pesticide residues including three herbicides, two insecticides, and one fungicide in tomato, watermelon, and papaya samples, utilizing QuEChERS methodology in combination with HPLC–VWD. Various experimental parameters affecting the chromatographic separation and the extraction efficiencies of the target analytes were studied, and the optimum conditions were established. Under the optimum conditions, except for methidathion, the proposed method demonstrated its usefulness for the determination of the analytes, with LODs and LOQs far below MRLs set by the EU for these pesticides in these samples. The method has also provided acceptable precisions, wide linearity ranges, and satisfactory recoveries for all the analytes in the selected fruit samples. The method has the advantages of simplicity, easy operation, and short analysis time with consumption of a low volume of the less hazardous organic solvent, acetonitrile. Moreover, the proposed modified QuEChERS procedure has also provided a better enrichment factor compared with the earlier reported versions. Therefore, it could be successfully utilized as an attractive alternative for the analysis of multiclass pesticide residues in fruits and other similar complex matrices of different origins for routine quality control.

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Compliance with Ethics Requirements

Conflict of Interest Tesfa Bedassa declares that he has no conflict of interest. Abera Gure declares that he has no conflict of interest. Negussie Megersa declares that he has no conflict of interest. This article does not contain any studies with human or animal subjects.

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