A rapid method for analysis of multi-OPPs residues in wheat by HS-SPME-GC-FPD/MSD

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Abstract

The rapid detection of pesticides residues in wheat has become a top food security priority. A solvent free headspace solid-phase microextraction (HS-SPME) has been evaluated for rapid screening of Organophosphorus pesticides (OPPs) residues in wheat with high sensitivity. The wheat sample (35 g) was placed in a 100mL sealed amber glass vial and heated at 60°C for 45 min. The 8 OPP residues were extracted with a 50µm/30µm DVB/CAR/PDMS fiber. In the HS-SPME procedures, the fiber was exposed in the headspace above sample for 45 min extraction, and then removed from the HS and injected in the GC injection port at 250°C for desorption of extracted chemicals. The multi residues were determined with a GC flame photometric detector GC-FPD and qualified by a GC mass spectrometer detector (GC-MSD). Seven fortification levels of 8 organophosphorus pesticides on wheat were analyzed. The GC responses for 50µm/30µm DVB/CAR/PDMS fiber were increased with increasing the fortification levels and there were significant (r2 > 0.985) linear regressions. The Lowest of Detections (LODs) of the multi pesticide standards were evaluated under the conditions of the validation study in a range of levels from 0 (control) to 100 ng (pesticides residue) g-1 of wheat that separated on a low-polar GC capillary column (Agilent DB-35UI). The result of HS-SPME method was compared with QuEChERS AOAC 2007.3 method.

Keywords: organophosphorus pesticides residue, Solid Phase Microextraction (SPME), GC-FPD/MSD, wheat

1. Introduction

Pesticides are the most common toxic chemicals used widely br the grain industry for protection of grain from insect damage. The well-known pesticides include organochlorine pesticides (OCPs), organophosphorus pesticides (OPPs) and nitrogen-containing herbicides. They can accumulate in the food chain during preharvest growing and postharvest storage. Therefore, there are food security concerns in regard to these compounds in food. As a result, maximum residue levels (MRLs) have been established by the international organizations (FAO/WHO) and government authorities (European Commission, 2008; APVMA, 2012). The introduction should provide sufficient background and state clearly the importance of the work being reported as well as its focused objective(s).

No	Name (IPUAC No.)	BP	MW	FAO/WHO’s MRL(mg/Kg)			APVMA’s MRL (mg/Kg)		
				Grain/Rape seed	Meat	Fruit	Grain/Rape seed	Meat	Fruit
1	Chlorpyrifos (2921-88-2)	200°C	350.59	0.5W	-/0.01P C9H11CL3NO3PS	0.5G	0.1	0.5	1G
2	Chlorpyrifos methyl (5598-13-0)	N/A	322.53	10W	0.1/0.01P	1G	10	0.05	N/A
3	Diazinon (333-41-5)	306°C	304.35	0.02M	2	0.5T	0.1	0.7/0.05P	2G/0.7V
4	Etrimphos (38260-54-7)	N/A	292.29	N/A	C10H17N2O4PS	N/A	N/A	N/A	N/A
5	Fenitrothion (122-14-5)	118°C	277.23	6	0.05	0.5Apple	10/0.1	0.05	1/0.5V
6	Malathion (121-75-5)	157°C	330.36	10W	N/A	0.5A,5G	8/10	1	2(8G)/2
7	Phosphamidon (13171-21-6)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
8	Pirimiphos-methyl (29232-93-7)	N/A	305.33	7	0.01 C11H20N3O3PS	N/A	N/A	N/A	N/A

The traditional pesticide residue analytical technologies are liquid-liquid extraction (LLE) and solid-phase extraction (SPE) (Ballesteros and Parrado, 2004). Currently, the most popular multi-residue determination approach is QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) method, which was developed by Anastassiades et al. (2003). The QuEChERS method was based on an extraction with MeCN followed by a partitioning step and then cleaning up by dispersive solid-phase extraction (d-SPE). This method was originally designed for analysis of pesticide residues in high-moisture (>75%) and low fat food matrices (Lehotay et al., 2005), the approach has been modified to suit for dry and fatty matrices by adding water and using modified clean-up step (Díez, et al., 2006; Lehotay et al., 2005). For barley samples, the QuEChERS method only achieved an overall recovery value of 60–70% of non-, medium- and polar pesticides (Díez, et al., 2006). For wheat grains, flour and bran samples, Diana, et al., (2011) used a modified QuEChERS method and GC-MS to analyse 24 pesticides with 2.5-5 μg kg^{-1} of Limit of Detection of Method (LODm) and 5-10 μg kg^{-1} of Limit of Quantity of Method (LOQm) respectively. Although the QuEChERS method for sample preparation and multi-residue determination of pesticides has been achieved, but it is difficult to avoid use of organic solvent, the simplicity of sample preparation and the complexity of different matrices.

The headspace solid-phase microextraction (HS-SPME) is a solvent-free extraction technique that has the potential to alter the current conventional extraction methods for on-site routine extraction chemicals. Since Pawliszyn and Liu (1987) reported the SPME technology (the solid-phase microextraction has been widely applied to analyse pesticide residues in environmental water and soil samples with DI-SPME (Direct Immersion) (Arthur and Pawliszyn, 1990; Doong and Liao, 2001; Dimitra and Triantafyllos, 2003; SIGMA-ALDRICH, 2013) , and in food such as strawberries and cherries (Yang et al., 1999), orange and carrot juices (Ana and Luís, 1999.), pears and apple or peach nectars (Fytianos et al., 2007$^{)}$, vegetable (Ren and Padovan ,2012). Ren and Pardovn (2012) reported that phosphine residue in wheat can be analysed by HS-SPME combined with GC-FPD (AOAC Official Method 2007). However, there was no report on analysis of organophosphorus esticide residues in wheat by HS-SPME-GC-FPD/MSD. Therefore, grain industry need to employ simple and sensitive trace analysis methods for the measurement and identification of these pesticides at the nanogram per kilogram level.

This paper will report a new Headspace-Solid Phase Micro Extraction method to determine multi-organophosphorus pesticide residue in wheat. The results will be compared between the HS-SPME and QuEChERS method.

2. Materials and Methods

2.1. Standards and reagents

Individual stock standard solutions of Chlorpyrifos-methyl, Fenitrothion and Pirimiphos-methyl were purchased from AccuStandard, Inc., CT, USA and stored at -16°C.

Series 9 OP Pesticide Mix #1 (OPP # 9) containing 100 µg OPPs mL^{-1} were supplied by RESTEK (Bellefonte, PA, USA).

Parathion-d10 and alpha-BHC-d6 (1000 µg mL^{-1}) were used for GC Internal Standard (AOAC Method 2007.01) from ULTRA Scientific, Agilent Technologies, USA.

Toluene and acetonitrile used were analytical grade provided by Aldrich Chemical Company Inc. Australia.

2.2. Wheat sample

Wheat used was 2012/2013 harvested Australia standard white wheat with 11.8% (w/w, wet basis) moisture content which was determined by using a Graintec HE 50 electronic moisture meter (Graintec Pty Ltd, Toowoomba, Australia). The results obtained were calculated from four replicate measurements.

2.3. Equipment and apparatus

The SPME fiber was performed in a manual holder (Supelco, Inc., Bellefonte, PA, USA). Eight different types of SPME fibre were compared to evaluate their effectiveness on the extraction of multi-organophosphorus pesticide compounds. Based on the polarity and size of fibers, eight SPME fibers selected were 100µm polydimethylsiloxane (PDMS), 7µm polydimethylsiloxane (PDMS), 85µm carboxen/polydimethylsiloxane (CAR/PDMS), 65µm PDMS/divinylbenzene (DVB), 60µm CarboWAX/Polyethylene Glycol (PEG), 50µm/30µm DVB/CAR/PDMS, 65µm CarboWAX/DVB and 85µm polyacrylate(PA). All selected fibers were purchased from Supelco (Bellefonte, PA, USA). The fibers were conditioned in accordance with the manufacture's recommendations prior to use, and cleaned between each extraction by exposing the fibres into the GC injection port for 15 min at 260ºC.

Thermo/Pierce Reacti-Therm Heating/Stirring Module (Model No. 18940, Waltham, MA, USA) for heating and homogenising grain samples.

For quantity analysis of organophosphorus pesticides, a HP 5890II GC-FPD system with phosphorus filter (HP, Inc., USA) was used. The GC-FPD was equipped with a split/splitless injector and a SPME inlet (Supelco, Bellefonte, PA, USA) which operated under splitless mode at 250°C during the analytic running. The injection was splitless and the GC split valve was set to switch on at 1 min after injection. Ten organophosphorus pesticides were separated on a 30m x 0.25mm ID x 0.25µm DB-35 Ultra capillary column (J&W Scientific, USA) and a 5m x 0.25mm ID (J&W Scientific, USA) as guard column. Helium was used as carrier gas at constant pressure of 35psi. The fuel and make up gases for FPD were hydrogen at 75 mL min^{-1} and air at 100 mL min^{-1} and nitrogen at 15 mL min^{-1}. The oven temperature was programed from initial temperature of 100°C raised to 150°C at 25°C min^{-1}; from 150°C to 193°C at 3°C min^{-1}; from 193 to 290°C at 35°C min^{-1} and stay for 5 min at 290°C.

For qualification analysis of organophosphorus pesticides, a HP GC 5890II-5972 mass selective detector (MSD) (HP, Inc., USA) was used. The GC conditions were identified from the GC-FPD as described previously. The mass spectrometer detector was operated at transfer line temperature 300°C; ion source temperature 170°C and EI ionization mode electron impact at 70eV.

The mass spectra of unknown compounds were de-convoluted and Identified by AMDIS_32 with the NIST MS database 2008. Identification of OPP was completed by comparing mass spectrum with GC-FPD retention time of each individual organophosphorus pesticide standard.

2.4. Preparation dilute standard and certificated organophosphorus pesticide standards

The diluted standard was prepared by adding calculated volume of standard into the acetonitrile solvent to make 10 mL solution containing 40 µg mL^{-1}.

The diluted Certificated Organophosphorus Pesticide Standards (OPP#9) was prepared by adding calculated volume of OPP#9 into the acetonitrile solvent to make 1 mL diluted OPP#9 solution containing 10 µg OPP#9 mL^{-1} and stored at 4°C.

2.5. Optimisation of HS-SPME extraction procedures

The HS-SPME procedure involved three steps. Firstly, choose of fiber, extraction temperature and period, and fiber desorption temperature and period in GC injector were optimized over a range of conditions to determine which can generate the highest values of most of pesticides residue we interested. Secondly, wheat was fortified at different levels and the peak areas were used to calculate for each level of fortification. This level of fortification became the calibration standards for calculation of the unknown pesticides residue as to the equation (Eq. 1):

$$Residue\ level = Pu + \frac{Fk}{Pk} \qquad \text{Eq. 1}$$

Where: F_k is the known fortification level

P_k is the peak area of the known fortification

P_u is the peak area of the unknown sample

2.5.1. Selection of fibers.

For selection of high efficient fibres on extraction of organophosphorus pesticides, eight fibers coated with different types of coatings, including 100µm polydimethylsiloxane (PDMS), 7µm polydimethylsiloxane (PDMS), 85µm carboxen/polydimethylsiloxane (CAR/PDMS), 65µm PDMS/divinylbenzene (DVB), 60µm CarboWAX(Polyethylene Glycol, PEG), 50µm/30µm DVB/CAR/PDMS, 65µm CarboWAX/DVB and 85µm polyacrylate (PA) were exposed in the sealed 4 mL amber vial containing Diluted OPP#9 standard and a magnetic stirring bar at 60°C for 1 hour extraction time. After 1 hour extraction, the fibre was retracted and exposed in a GC-FPD injection port at 250°C for holding 10 min.

2.5.2. Estimation of GC injector temperature for desorption of OPPs from fibre.

For optimisation of desorption temperature and time, a selected 50µm/30µm DVB/CAR/PDMS fiber was used to evaluate efficient thermal desorption of the OPPs in a GC injection port depend on both the GC injector temperature and exposure time. The selected fibre was inserted into the headspace of the 4 mL amber vial with a magnetic stirring bar at 60ºC for 1 hour exposure. The fibre was carefully injected into the GC at different injector temperatures (100,150, 200 and 250ºC) and desorbed for 1, 2, 3 and 5, 10 minutes respectively. The time for desorption of OPPs from the HS-SPME fiber was also decided by re-injection of the same fiber completed desorption process at 250°C in 1, 2, and 3 min. Results are the mean of duplicate samples and duplicate injection of each sample.

2.5.3. Estimation of temperature and time for extraction of OPPs.

For optimisation of extraction temperature, wheat samples (35 g) spiked with ten different OPPs in a 100 mL amber vial equipped with a PTFE septum lid was immersed in a water bath at 25, 40, 60, 80, and 100°C respectively. The pesticide residues were extracted with a selected 50µm/30µm DVB/CAR/PDMS fibre for 30, 45 and 60 min. After exposure, the fibre was retracted and then exposed in a GC injection port (250°C) for holding 3 min. All results are the mean values from two replicate samples and duplicate injection of each sample.

2.5.4. Spiking studies and lowest of detection (LODs)

The method validation was conducted by analysing spiked standards, including the spiking study and evaluation of the Limit of Detections (LODs). For preparation of spiked standards, wheat samples at 0 (untreated blank), 0.05, 0.025, 1, 5, 25 and 100 ng (pesticides) g^{-1} of wheat were prepared by adding and mixing appropriate volumes of OPPs standard into 100 mL flask containing 50 g untreated blank wheat. The fiber 50µm/30µm DVB/CAR/PDMS was pierced the flask and exposed in the headspace over the wheat sample. Then, the flask was placed in a water bath at 60°C for 45 min. At the end of extraction, the fiber was withdraw and inserted into the GC-FPD/MS injection port at 250°C to desorb for 3 min. Peak areas recorded in the figures are the mean of duplicate samples.

2.6. Compare QuEChERS procedures and HS-SPME method for analysis of OPPs

AOAC official method 2007.01 was adapted to analyse OPP residues in wheat by GC-FPD. Spiked wheat samples (5 g) were grinded and taken instead of 15 g (sample amount in the AOAC official method 2007.01 procedure) being the rest 10 g filled up with cool DI water for adding the necessary moisture (70%-75%) in a 50 mL PTFE Tubes (Part No.5982-5755, Agilent, US). Then 15 mL of acetonitrile with 1% acetic acid plus 75 µL diluted QuEChERS AOAC Standard solution (Part No.5190-0502, Agilent, US) were added to the sample. The mixture was shaken for 1 min. Afterwards, a packet of QuEChERS AOAC Pouch (6 g of anhydrous $MgSO_4$ and 1.5 g of NaAcetate, Part No.5982-0755, Agilent, US) was added, and the mixture was hand-shaken for another minute after 1 min of centrifugation at 2000 r.p.m. 8 mL of extraction solution was cleaned-up within a 15 mL dispersive SPE tube (400 mg PSA and 1200 mg $MgSO_4$, Part No.5982-5058, Agilent, US) by hand-shaken for 1 min again. After 1 min centrifugation at 2000 r.p.m, 4 ml extract and the addition of 400 µL of a diluted AOAC Method QC solution (Triphenyl phosphate, Part No.5190-0503, Agilent, US) plus 1 mL Toluene were evaporated in the Thermo/Pierce Reacti - Therm Heating/Stirring Module (Model No. 18940, Waltham, MA, USA) at 50°C under sufficient Nitrogen flow protection until volume was lower than 0.5 mL. The concentrated extract was filled up to 1 mL by adding toluene and dried by adding anhydrous $MgSO_4$. The extract was centrifuged and then transferred to a 2 mL AS vial for GC–FPD analysis.

Finally, after the optimization of HS-SPME procedures, the results were compared with those from the QuEChERS methodology. At the end of the optimized extraction time, the fiber was withdrawn from the HS and injected into the GC injection port at optimized desorption temperature conditions. For the conventional QuEChERS analysis, a 2µL aliquot of final QuEChERS sample extraction solution was injected into the GC instrument.

2.7. Statistical analysis

All samples were analysed by two fibres and each had two injections (*n=4*). The GC peak areas were averaged. The variation of GC peak areas, pesticides levels, and SDs were analysed with Microsoft Excel Software and Statistic software OriginPro 7.5 SR1 (v7.5776).

3. Results and Discussion

3.1. Effect of fiber selection of HS-SPME extraction

The eight fibers coated with different types of coatings, including 100µm polydimethylsiloxane (PDMS), 7µm polydimethylsiloxane (PDMS), 85µm carboxen/polydimethylsiloxane (CAR/PDMS), 65µm PDMS/divinylbenzene (DVB), 60µm CarboWAX(Polyethylene Glycol, PEG), 50µm/30µm DVB/CAR/PDMS, 65µm CarboWAX/DVB and 85µm polyacrylate (PA) were compared. The 50µm/30µm DVB/CAR/PDMS extracted all 10 OPP compounds with higher intensities while the fewest compounds were extracted by 85µm carboxen (CAR)/PDMS in the headspace (Figure 1). The observation proved the OPP residues with high polarity, boiling point from 74.1°C to 306°C and high molecular weight from 220.98 to 350.59 were not extracted by 85µm carboxen (CAR)/PDMS which is suitable for gases and low molecular weight compounds (MW 30-225). In the other hand, the carbowax (PEG), divinylbenzene (DVB), and thinner polydimethylsiloxane (PDMS) were observed to extract more OPP residues than other fiber coatings. The result matched the different fibers' characters, the carbowax (PEG) coating is applied to extract polar compounds with MW 40-275, the divinylbenzene (DVB) coating can extract volatiles, amines and nitro-aromatic compounds with MW 50-300, and thinner polydimethylsiloxane (PDMS) coating can extend the molecular weight range of extraction to 600 as maximum

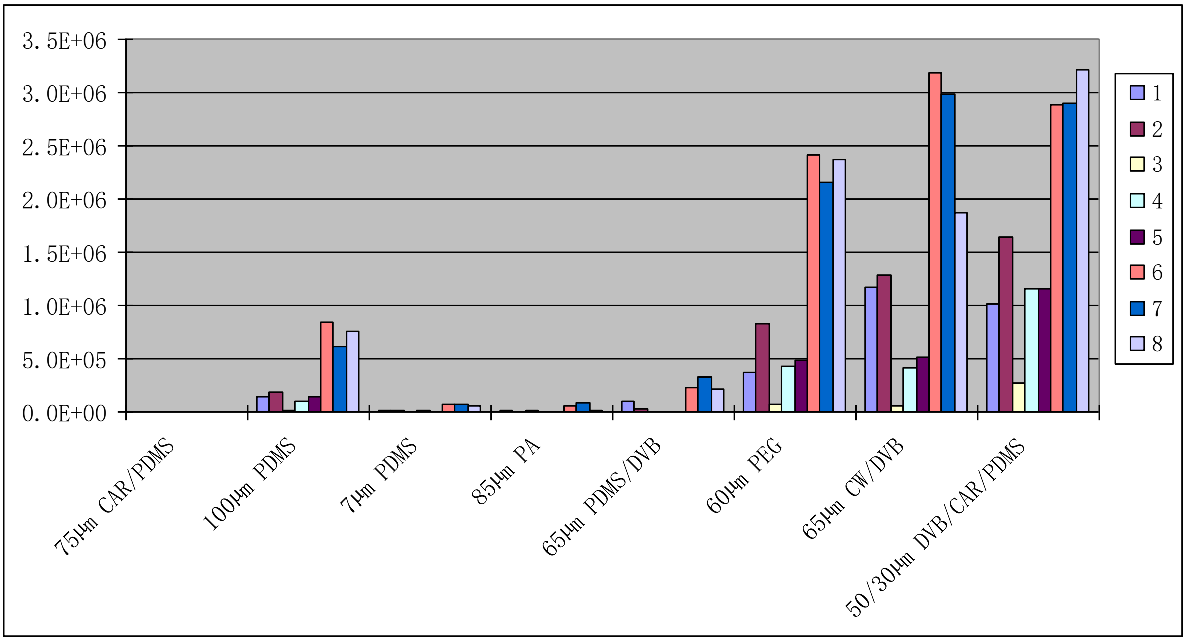

Figure 1 GC readings for 8 OPP residues extracted by eight fibers after 45 min extraction at 60°C. 1, Diazinon; 2, Etrimfos; 3, Phosphamidon; 4, Chloropyriphos-Methyl; 5, Pirimiphos-Methyl; 6, Malathion; 7, Chloropyriphos; 8, Fenitrothion.

3.2. Effect of temperature and period on extraction of pesticides residues

The extraction temperatures (25°C, 40°C, 60°C, 80°C and 100°C) were evaluated in the HS-SPME parameter screening experiment. The chromatograph peak areas of the most pesticides residue extracted from spiked wheat increased quickly from 40°C to 60°C, then the extraction efficiencies for all pesticides residues were dropped dramatically after 60°C (Figure 2). This phenomenon is probably partly due to the degradation of OPP compounds at higher temperature, because extra peaks came out and grew up were observed not reported. Hence, 60°C was chosen as the optimal temperature for the HS-SPME extraction of pesticides residues. At the different HS-SPME extraction temperatures, the 50µm/30µm DVB/CAR/PDMS had more sensitivity on broad extraction of low-boiling point compounds than the 65µm CarboWAX/DVB fiber. While the CarboWAX/DVB fiber absorbed more high-boiling point OPPs than the DVB/CAR/PDMS fiber, e.g. the GC-FPD response of Malathion was 1.55 times higher in the CarboWAX/DVB fiber. In summary, the DVB/CAR/PDMS fiber had wider extraction range of organophosphorus pesticide residues with comparable GC response signals.

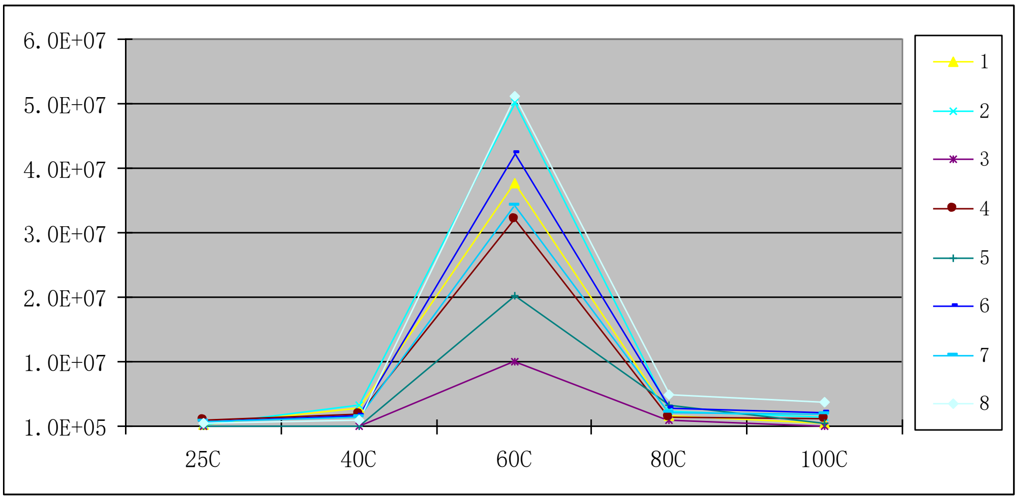

Figure 2 Effect of heating temperature on HS-SPME procedure of 8 OPP residues detected 50µm/30µm DVB/CAR/PDMS after 45 min extraction. 1, Diazinon; 2, Etrimfos; 3, Phosphamidon; 4, Chloropyriphos-Methyl; 5, Pirimiphos-Methyl; 6, Malathion; 7, Chloropyriphos; 8, Fenitrothion.

On the basis of the previous optimization experiment results, the 50µm/30µm DVB/CAR/PDMS fiber was selected for optimization of the HS-SPME extraction time. 3.4 µL of Series 9 OP Pesticide Mix #1 was extracted from a 4 mL amber vial by the two DVB/CAR/PDMS fibers separately for 10, 30, 45, and 60 min (Figure 3). It was observed that the concentration of polar OPPs with lower boiling point and lower molecular weight compounds had been absorbed suturally at the first 10 min, such as DDVP and methacriphos, and dropped as increasing the extraction time. While it was necessary to have longer extraction time for OPP compounds with higher molecular weight and higher boiling point, such as Chlorpyrifos (2921-88-2) and Malathion (121-75-5). The most OPP compounds had their peak value within 30 min, but for phosphamidon and pirimiphos-methyl, the SPME extraction time 45 min was the top point.

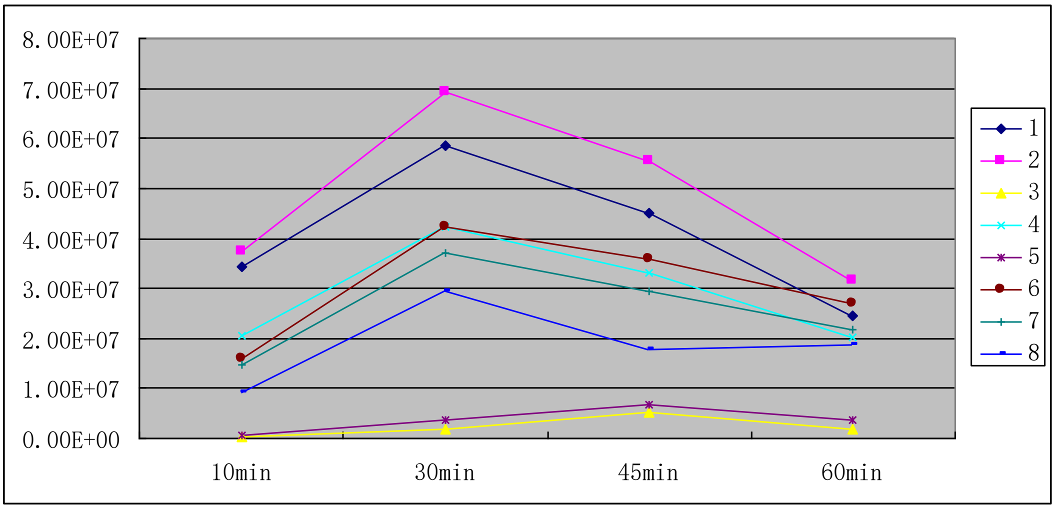

Figure 3 Effects of time of HS-SPME extraction for 8 OPP residues with 50µm/30µm DVB/CAR/PDMS fiber at 60°C. 1, Diazinon; 2, Etrimfos; 3, Phosphamidon; 4, Chloropyriphos-Methyl; 5, Pirimiphos-Methyl; 6, Malathion; 7, Chloropyriphos; 8, Fenitrothion.

3.3. Effect of Gas Chromatograph Injector temperature and time on desorption of pesticides from the fiber

The chromatograph peak areas for the most tested 8 pesticides residue extracted from spiked wheat varied from 150°C to 250°C (Figure 4). The HS-SPME extraction condition was at 60°C for 45 min, the sample was 0.1 mg Kg-1 spiked sample. The peak areas of detected pesticides residues rose up with increasing injection port temperature until it was at 250°C, which was the middle range of the recommend operation temperature for the 50µm/30µm DVB/CAR/PDMS fiber. The desorption time of the HS-SPME fiber was decided by re-injection of the same fiber completed desorption process at 250°C in 1, 2, and 3 min. The results showed the OPPs residues absorbed on the 50µm/30µm DVB/CAR/PDMS fiber were desorbed quickly at 250°C, 0.3% of absorbed 8 tested OPP residues were left after 1 min desorption time. There was no OPP residue peaks detected in the other two experiments for re-injection after 2, and 3 min desorption. Therefore, 2 min SPME fiber desorption time was chosen.

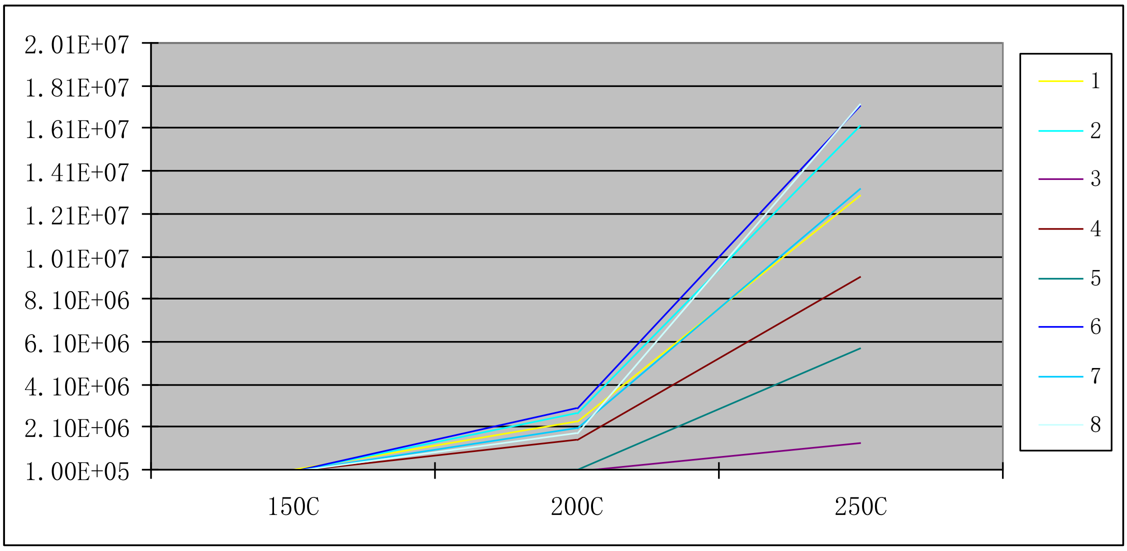

Figure 4 Effects of Gas Chromatograph injector temperature on desoprtion of 8 OPP residues from the 50µm/30µm DVB/CAR/PDMS fiber after 45 min extraction at 60°C. 1, Diazinon; 2, Etrimfos; 3, Phosphamidon; 4, Chloropyriphos-Methyl; 5, Pirimiphos-Methyl; 6, Malathion; 7, Chloropyriphos; 8, Fenitrothion.

3.4. Linearity and lowest of detection (LODs)

The seven fortification levels used were 0 (untreated blank), 0.05, 0.025, 1, 5, 25 and 100 ng g^{-1} were analysed under the optimised HS-SPME experimental conditions. The 50µm/30µm DVB/CAR/PDMS fiber was used for extraction of pesticides at 60°C for 45 min, the fiber desorption happened at 250°C for 2 min. The GC response had an approximate linear relationship within the fortification levels, the regression equation and correlation coefficient for each of OPP compounds were calculated and shows in Table 1. The values of the correlation coefficient were > 0.98 for all OPPs analysed by GC-FPD.

The LOD was defined as the lowest concentration that the analytical process could be separated reliably from background levels. The estimation of LOD was based on 3 times signal-to noise ratio. The linear range was determined by analyzing seven fortification level standards. The results of linearity and LODs were shown in Table 1 and Figure 5.

Table 1 Linearship and Limit of Detection (LOD) of 8 OPP compounds extracted by HS-SPME (50µm/30µm DVB/CAR/PDMS fiber at 60°C for 45 min) and detected by GC-FPD.

No.	Name	Linear range (ng g^{-1})	LODs (ng g^{-1})	Correlation Coefficient (R^2)
1	Diazinon	0.05-100	0.021	0.9909
2	Etrimfos	0.25-100	0.022	0.9925
3	Phosphamidon	5.0-100	4.493	0.9808
4	Chloropyriphos-Methyl	1-100	0.057	0.9965
5	Pirimiphos-Methyl	25.0-100	7.664	0.9884
6	Malathion	1-100	0.068	0.9979
7	Chloropyriphos	1-100	0.096	0.9977
8	Fenitrothion	5-100	0.174	0.9993

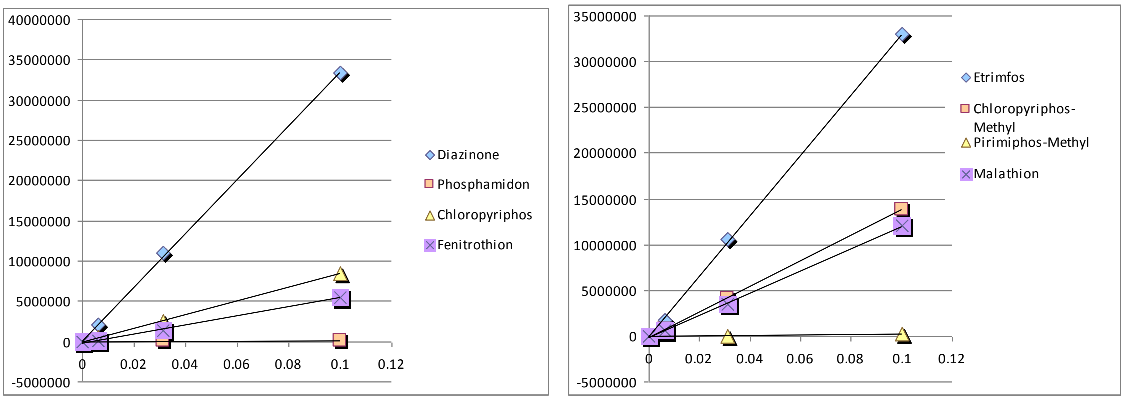

Figure 5 Linearship graphs for 8 OPPs residue extracted from spiked wheat samples with HS-SPME method.

3.5. Comparison of QuEChERS extraction method and HS-SPME method

The three levels of spiking fortification standards were analysed and compared by QuEChERS AOAC extraction method and HS-SPME method in the same GC-FPD condition as mentioned in 2.2. The result showed the HS-SPME method had higher sensitivity and selectivity than QuEChERS AOAC method 2007.3 in the Table 2. For example, HS-SPME could detect 8 compounds at 5 ng g^{-1} level, but QuEChERS not. At all three levels of spiking fortification, the sensitivity of HS-SPME was higher than the QuEChERS AOAC 2007.3 for most of OPP compounds.

Table 2 Gas Chromatograph Peak Areas for the three spiking level wheat samples extracted with the QuEChERS AOAC 2007.3 method and HS-SPME method.

No.	Compound	5 (ng g^{-1})		25 (ng g^{-1})		100 (ng g^{-1})	
		HS-SPME	QuEChERS	HS-SPME	QuEChERS	HS-SPME	QuEChERS
1	Diazinon	2.08E+06	N/A	1.09E+07	9.43E+04	3.34E+07	4.13E+05
2	Etrimfos	1.79E+06	N/A	1.06E+07	1.18E+05	3.30E+07	4.92E+05
3	Phosphamidon	4.51E+03	N/A	4.27E+04	N/A	1.19E+05	N/A
4	Chloropyriphos-Methyl	7.20E+05	N/A	4.14E+06	N/A	1.39E+07	3.45E+05
5	Pirimiphos-Methyl	N/A	N/A	3.06E+04	N/A	1.84E+05	1.24E+05
6	Malathion	6.11E+05	N/A	3.46E+06	1.08E+05	1.20E+07	4.47E+05
7	Chloropyriphos	4.65E+05	N/A	2.44E+06	8.63E+04	8.43E+06	3.66E+05
8	Fenitrothion	1.56E+05	N/A	1.35E+06	1.12E+05	5.49E+06	4.64E+05

Except for the higher sensitivity and selectivity, the advantages of HS-SPME method were observed when using 50µm/30µm DVB/CAR/PDMS fiber without solvent consuming, comparing with QuEChERS AOAC 2007.3, included decreasing the sample preparation procedure steps from 17 steps to 5 steps, lower matrix effect (no huge solvent peak) and less chemical consuming. Although, the scientific literature contains well-established methods, like QuEChERS for the determination of pesticides residues in wheat at low levels. However, further quantitative analysis and identifications of unknown degradation compounds are needed to clarify the mechanisms involved in the HS-SPME extraction process.

4. Conclusions

A multi-residue method based on HS-SPME sampling technology with GC-FPD analysis was established for the detection of organophosphorus pesticide residues in wheat. The HS-SPME method using the 50µm/30µm DVB/CAR/PDMS fiber is an innovative solvent-free fast screening method with high sensitivity by being quick, easy, cheap and safe. In this study, 8 typical organophosphorous pesticide residues, from low to high-boiling point, and low to high molecular weight, were identified by GC-MSD and quantified by GC-FPD. The further step will be extending this method to different pesticides residue on different grain matrix, e.g. canola.

Acknowledgements

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