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The Application of Solid Phase Microextraction in the Analysis of Organophosphorus Pesticides in a Food Plant

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A SPME method for the determination of organophosphorus pesticide residues in a local food plant, *Chrysanthemum coronarium*, was established. Pesticide residues were extracted by SPME fiber (with 100 μm poly(dimethylsiloxane) coating) from a biphasic water/plant tissues mixture and determined by GC-FPD. An equilibrium model was derived for the system and revealed that pesticide recoveries were related to the water:plant tissues ratio, f, and the partition coefficient, K_{WV}, for the distribution of pesticides between the aqueous phase and the plant tissues. The model was verified by four organophosphorus pesticides, namely phorate, diazinon, methyl parathion, and ethion. The best fitted K_{WV} values ($P < 0.05$) for the pesticides were found to be $4.73 \pm 0.32 \times 10^{-2}$ (phorate); $1.11 \pm 0.10 \times 10^{-1}$ (diazinon); $9.18 \pm 0.95 \times 10^{-2}$ (methyl parathion); and $8.21 \pm 1.28 \times 10^{-3}$ (ethion). Remarkable correlation between the K_{WV} of the organophosphorus pesticides and their octanol/water partition coefficients, K_{OW}, was also established.

Introduction

The presence of pesticide residues in fruits and vegetables has aroused growing public concerns. Cases of intoxication due to consumption of contaminated agricultural food products happen from time to time. New analytical techniques that enable determination of pesticide residues in complex sample matrices with shorter turnaround time, improved sensitivity, and/or reliability are in constant demand. At present, most analytical methods in the literature involve extraction of pesticide residues from plant tissues by organic solvents (*1*−*4*), surfactants (*5*, *6*), supercritical fluids (*7*), or solid-phase extractants (*8*−*10*) followed by GC or HPLC determination (*11*−*13*). These extraction and cleanup procedures are usually tedious and time-consuming. Also, usage of environmental "unfriendly" organic solvents in most of these procedures imposes health hazardous to laboratory personnel and extra operational costs for waste treatment.

Solid phase microextraction (SPME) is a new and rapidly developing "solvent-less" solid−liquid extraction method (*14*−*25*). It involves extraction of analytes from the sample matrix (liquid or gaseous phase) onto an immobilized stationary phase. These extracted analytes may then be determined by GC via thermal desorption at the injector port or by HPLC via special injector interface. The advantages of SPME include true solvent-free extraction, high sensitivity, no need of cleanup procedures, and simple instrumentation. These make SPME an ideal tool for pesticide residue determination. In fact, there are already reports on the applications of SPME for the sampling of organophosphorus pesticides in surface and groundwaters (*25*−*27*). However, literature on the feasibility of SPME determination of pesticide residues in agricultural products remains scarce.

Extraction efficiency of any liquid−solid extraction depends very much on the partitioning behavior of the analytes between the solution phase and the sorbent. Previous studies by Johnson et al. (*28*) have already shown that recoveries in the solid phase extraction of various nitrogen-containing and organophosphorus pesticides from natural waters containing humic materials were lower than those obtained from pure water. In the application of SPME to determine pesticide residues in agricultural products, knowledge on the sorption properties of the analytes on vegetation is, therefore, important. In this work, a novel method of measuring the partition coefficient for the distribution of organophosphorus pesticide between water and plant tissues is developed using SPME. Four organophosphorus pesticides, namely phorate, diazinon, methyl parathion, and ethion, were selected as model pesticides, and a local commercial vegetable species, *Chrysanthemum coronarium* (Chinese name, "*Tong-ho*"; Japanese name, "*Shungiku*"), was used for the study. *Chrysanthemum coronarium* is a very popular food plant in the southern part of China and is mainly produced in the Guangdong Province. There are numerous food poisoning cases in the region due to the consumption of organophosphorus pesticide contaminated *Chrysanthemum coronarium* every year.

Experimental Section

Authentic phorate, diazinon, methyl parathion, and ethion standards were obtained from Supelco, Inc. and were used without further purification. SPME device (with 100 μm poly(dimethylsiloxane) coating) was obtained from Supelco Inc. The SPME fiber was preconditioned at 270 °C under helium overnight prior to its first extraction and was conditioned at 270 °C under helium for 30 min before each subsequent extraction. Tissue blender (National) was thoroughly rinsed with deionized water and methanol (AR grade, RDH) before each use. A Hewlett-Packard HP-5890 GC-FPD (phosphorus mode) with a 10 m × 0.53 m × 2.0 μm HP-17 column was used for the determination of the organophosphorus pesticides. A local food plant species, *Chrysanthemum coronarium*, was used for the study.

In the determination of optimum extraction time and detection limits, known amounts of organophosphorus pesticide standards were spiked into 50.0 g of fresh *Chrysanthemum coronarium* which had been mechanically cut into small pieces and was suspended in minimum amount of pure water. The spiked sample was allowed to stand for ~1 h before mixing and blending with a final volume of 150 mL of deionized water in a tissue blender for at least 5 min. Five grams of the resulting well-mixed paste was transferred into a clean 10 mL vial. The mouth of the vial was sealed with paraffin so that a headspace of about 5 mL was maintained during SPME extraction. SPME extraction was carried out by inserting the SPME fiber into the well stirred

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sample at room temperature. The sampled SPME fiber was rinsed with minimum amount of deionized water before GC-FPD analysis. A signal-to-noise ratio of 3:1 was used as the criteria for detection limit determination.

In the determination of water:plant tissues partition coefficients of the pesticides, a series of samples containing 50.0 g of fresh vegetables and various amounts of deionized water were blended into well-mixed pastes. Five grams of each of the resulting pastes were transferred into clean 10 mL vials. Equal amounts of pesticide standards were spiked into each sample and the samples were then sealed with paraffin and were allowed to stand for half an hour at room temperature with stirring before SPME extraction. The extraction time was 90 min, and the sampled SPME fiber was rinsed with a minimum amount of deionized water before GC-FPD analysis. Recoveries of the various organophosphorus pesticides at different amounts of aqueous extractant were determined by comparing the peak areas of the pesticides obtained above to those obtained by spiking the same amount of pesticide standards into 5.0 g of deionized water.

Thermal desorption of the extracted organophosphorus pesticides was effected by inserting the SPME fiber into the GC injector port under splitless mode, kept at 270 °C, for 5 min. The GC column temperature was first held at 35 °C for 6 min, then increased to 200 °C at a rate of 30 °C/min, and held at 200 °C for 3 min before further increased to 280 °C at a rate of 10 °C/min. The flame photometric detector was set at phosphorus mode. After each extraction and desorption, the SPME fiber was reconditioned by keeping in the GC injection port for 30 min at 270 °C. Fiber blank was checked by putting the conditioned SPME fiber in clean deionized water for 15 min followed by thermal desorption and GC checking before each extraction. Throughout the study, no pesticide residues were found left on the SPME fiber after each reconditioning.

Results and Discussion

The dried/wet weight ratio of fresh *Chrysanthemum coronarium* is ca. 7% and its lipid content is ca. 0.3% (wet weight) (*29*). Phorate, diazinon, methyl parathion, and ethion spiked onto fresh vegetable samples were leached by blending with deionized water (pH 7) at room temperature. In the determination of optimum extraction time and detection limits, the spiked samples were allowed to stand at room temperature for about 1 h before subsequent leaching and extraction. The purpose of suspending the sample in minimum amount of water is to ensure homogeneous spiking. A longer "aging" period was not attempted in view of possible hydrolytic loss of the spiked pesticides. For the same reason, only a half an hour "equilibration" period was allowed during the determination of water:plant tissues partition coefficients of the pesticides. With the intention to simplify the sample preparation procedures, plant tissues were not separated from the aqueous extract before solid phase microextraction. In essence, a biphasic system was created with the pesticides distributed between the aqueous phase and the plant tissues, and the SPME fiber was in equilibrium with the those species partitioned into the aqueous phase. The absorption−time profiles for the four organophosphorus pesticides (water:plant tissues ratio = 3:1) are shown in Figure 1. With the exception of ethion, the amount of pesticides extracted by the SPME fiber surpassed 90% of their corresponding equilibrium values within 60 min. In the case of ethion, equilibrium is not reached after 4 h. These results were consistent with those obtained by Valor et al. (*27*) who attributed the slow SPME equilibration of ethion to its high molecular mass and, hence, small diffusion coefficient. The absorption−time profiles with different water:plant tissues ratios have also been examined, but no

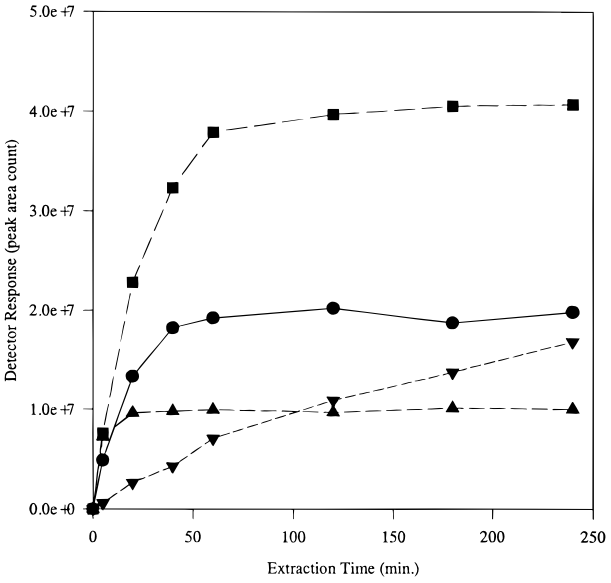

FIGURE 1. SPME absorption−time profile for the organophosphorus pesticide residues (60−140 ppb) in *Chrysanthemum coronarium*: (■) diazinon; (●) phorate; (▲) methyl parathion; and (▼) ethion. Water:plant tissues ratio = 3:1.

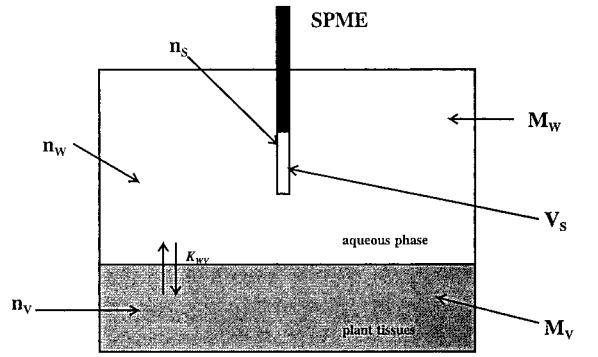

FIGURE 2. The biphasic model representing the solid-phase microextraction of organophosphorus pesticides from the aqueous plant tissues extract.

significant difference was revealed. Nevertheless, extraction equilibration may not be a necessity in solid phase microextraction as long as the extraction duration is fixed, and mixing conditions and extraction volumes are kept constant (*20*, *30*). A 90 min extraction duration was adopted throughout the experiment.

As demonstrated by Porschmann et al. (*24*), the removal of analytes from the aqueous phase of a multiple component phases system by SPME should introduce minimal disturbance to the system, especially for organophosphorus pesticides which possess much lower octanol/water partition coefficients, K_{OW}, than other nonpolar analytes. The equilibrium processes in the present biphasic system can be represented by the model shown in Figure 2. The distribution of the pesticides among the plant tissues, aqueous medium, and the SPME coating can be summarized by two partition coefficients

$$K_{WV} = \frac{C_W}{C_V} = \left(\frac{n_W}{n_V}\right)\cdot\left(\frac{M_V}{M_W}\right) \quad (1)$$

$$K_S = \frac{C_S}{C_W} = \left(\frac{n_S}{n_W}\right)\cdot\left(\frac{M_W}{V_S}\right) \quad (2)$$

and

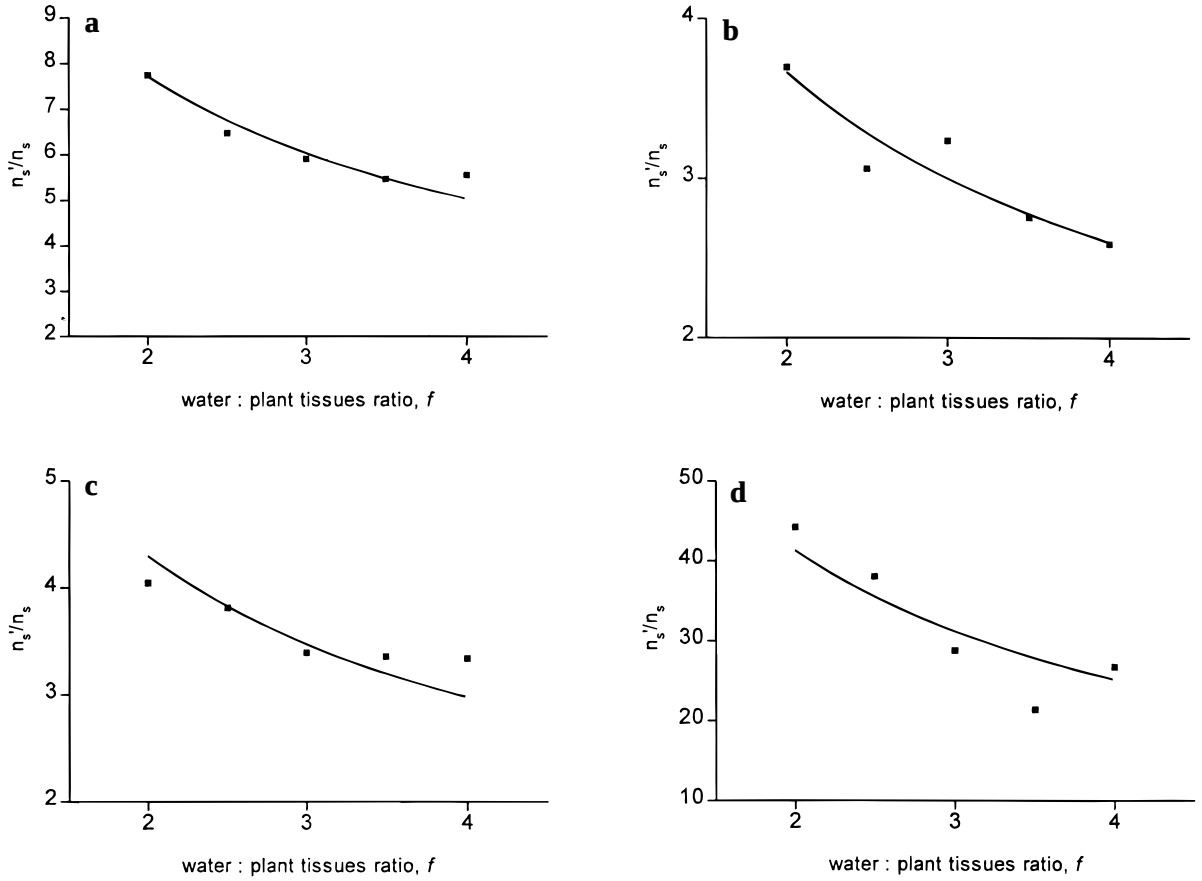

FIGURE 3. Relation between the reciprocal of SPME recovery and the water:plant tissues ratio, *f*, for (a) phorate; (b) diazinon; (c) methyl parathion; and (d) ethion (extraction time = 90 min). The dots represent observed values and the lines are the modeled best fit curves generated in accordance with eq 9.

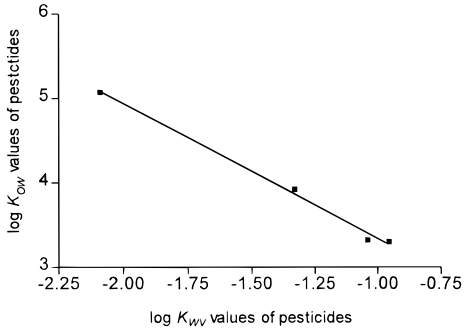

FIGURE 4. The relation between the estimated water/plant tissues partition coefficients, K_{WV}, of the organophosphorus pesticides and their octanol/water partition coefficients, K_{OW}.

$$n_T = n_S + n_W + n_V \tag{3}$$

where K_{WV} and K_S are the partition coefficients for the distribution of pesticide species between the aqueous phase and the plant tissues component and between the SPME coating and the aqueous phase respectively; C_W and C_V are the concentrations of the pesticide species in the aqueous phase and in the plant tissues component, in terms of amount of pesticide per unit mass of the corresponding component phase, respectively; C_S is the concentration of pesticide species on the SPME coating in terms of amount of pesticide per unit volume of coating; n_T, n_W, n_V, and n_S are the total amount of pesticide in the system and the amounts of pesticide species distributed in the aqueous phase, plant tissues, and SPME coating, respectively; M_W and M_V are the masses of the aqueous and the plant tissues component phases respectively; and V_S is the volume of the SPME coating.

From eq 3, the relation between n_S and n_T can be obtained

$$n_W + n_V = n_T - n_S$$

$$n_W\left(1 + \frac{M_V}{K_{WV}M_W}\right) = n_W k = n_T - n_S \tag{4}$$

where $k = 1 + M_V/K_{WV}M_W$. Substituting (4) into (2), we have

$$K_S = \frac{n_S}{\left(\frac{n_T - n_S}{k}\right)}\cdot\frac{M_W}{V_S} = \frac{n_S k M_w}{V_S(n_T - n_S)}$$

$$K_S V_S n_T - K_S V_S n_S = n_S k M_W$$

$$n_S = \frac{K_S V_S n_T}{K_S V_S + kM_W} = \frac{K_S V_S n_T}{K_S V_S + M_W\left(1 + \frac{M_V}{K_{WV}M_W}\right)} \tag{5}$$

$$n_S = \frac{K_S K_{WV} V_S n_T}{K_S K_{WV} V_S + K_{WV} M_W + M_V} \tag{6}$$

Similar to n_S, a new term, n_S', can be defined to represent the amount of pesticide absorbed onto the SPME coating when a similar amount of pesticide, n_T, is spiked into an "equivalent" monophasic system. When $K_{WV} \ll 1$ (as expected for most organic pollutants), such an equivalent monophasic system can be achieved by using $(M_W + M_V)$ amount of pure water. Similar to eq 6, the relation between n_S' and n_T can be expressed as follows

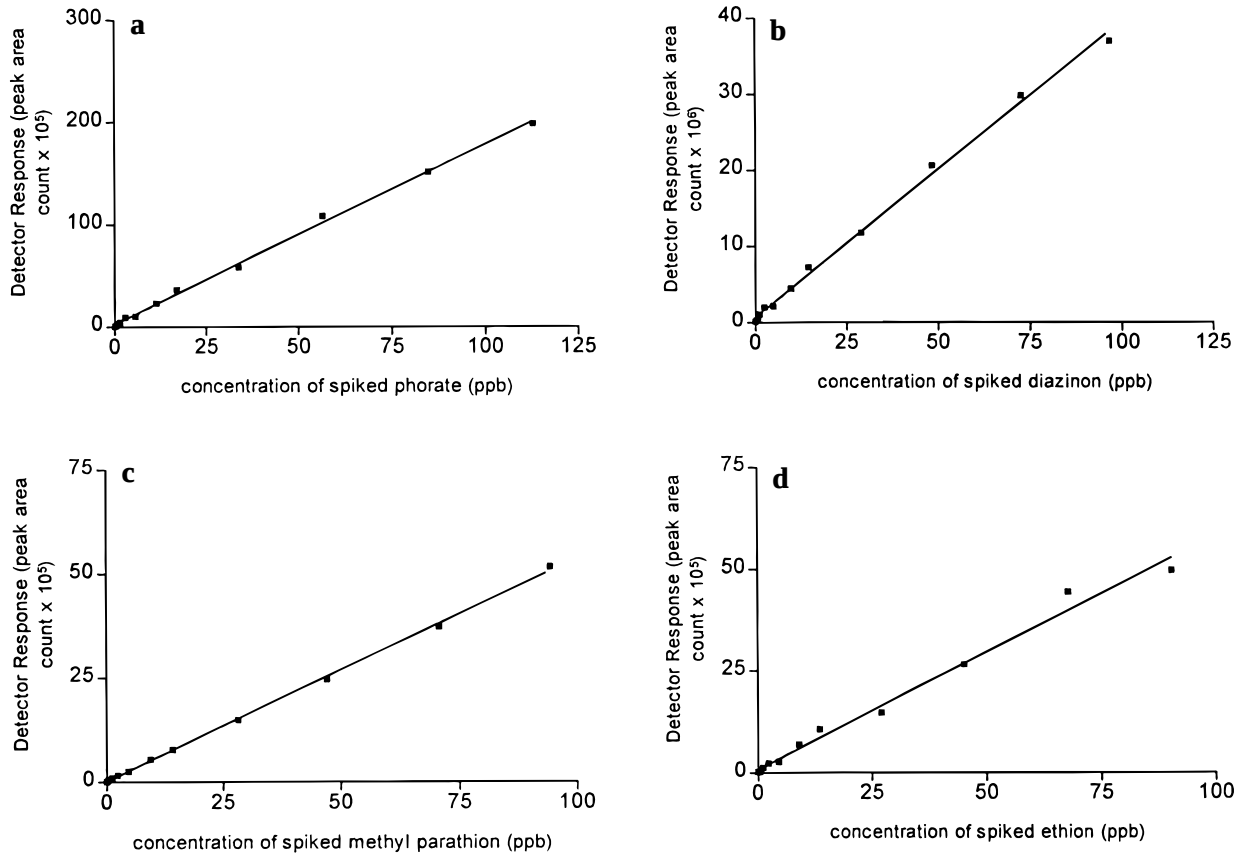

FIGURE 5. Relation between the SPME/GC-FPD response and pesticide spike level for the four organophosphorus pesticides (a) phorate; (b) diazinon; (c) methyl parathion; and (d) ethion. The spike levels are based on fresh vegetable weight.

$$n_S' = \frac{K_S V_S n_T}{K_S V_S + (M_W + M_V)} \quad (7)$$

From eqs 6 and 7, SPME analyte recovery can be expressed as

$$\frac{n_S}{n_S'} = \frac{K_S K_{WV} V_S + K_{WV}(M_W + M_V)}{K_S K_{WV} V_S + K_{WV} M_W + M_V} \quad (8)$$

If the water:plant tissues ratio is f:1 (i.e., $M_W = f \cdot M_V$), the reciprocal of recovery, n_S'/n_S, becomes

$$\frac{n_S'}{n_S} = \frac{K_S K_{WV} V_S + M_V(fK_{WV} + 1)}{K_{WV}[K_S V_S + M_V(1 + f)]} \quad (9)$$

The term $M_V \cdot (1 + f)$ must be greater than $K_S \cdot V_S$ because of the large sample size, M_V, and excess aqueous extractant used (i.e., $f > 1$). As a result, eq 9 can be simplified to

$$\frac{n_S'}{n_S} \approx \frac{f}{1 + f} + \frac{1}{K_{WV}(1 + f)} \approx \frac{K_{WV} f + 1}{K_{WV}(1 + f)} \quad (10)$$

From the above equilibrium considerations, the SPME analyte recovery is controlled by the partition coefficient, K_{WV}, for the distribution of the organophosphorus pesticide between aqueous phase and the plant tissues as well as the amount of water used for the extraction. Fitting the reciprocal of the SPME recoveries, n_S'/n_S, of the four organophosphorus pesticides at different water:plant tissues ratios, f, into the above equilibrium model reveals the K_{WV} values of the pesticides ($P < 0.05$) to be $4.73 \pm 0.32 \times 10^{-2}$ (phorate); $1.11 \pm 0.10 \times 10^{-1}$ (diazinon); $9.18 \pm 0.95 \times 10^{-2}$ (methyl parathion); and $8.21 \pm 1.28 \times 10^{-3}$ (ethion) (Figure 3). The equilibrium model derived above should also be applicable to the distribution of chemical species in any biphasic system. Thus, eq 10 represents a new and more convenient way to measure partition behavior of chemical compounds using SPME.

TABLE 1. Relationship between the K_{WV} and K_{OW} Values of the Organophosphorus Pesticides

pesticide	estimated log (K_{WV})	log (K_{OW})
phorate	−1.33	3.92[a]
diazinon	−0.96	3.30[a]
methyl parathion	−1.04	3.32[b]
ethion	−2.09	5.07[b]

[a] Data from ref 31. [b] Data from ref 32.

As the lipophilic pesticide species are likely to be bound to the lipid fraction of the plant tissues, their water:plant tissues partition properties should be correlated with their K_{OW}. Figure 4 shows the relation between the experimentally determined log K_{WV} of the organophosphorus pesticides and their log K_{OW} values (data tabulated in Table 1). The two partition coefficients are found to follow the Collander equation (*33*)

$$\log K_{OW} = -1.60(\pm 0.08) \log K_{WV} + 1.73(\pm 0.12) \quad (11)$$

$$(r^2 = 0.9945;\ P < 0.05)$$

The remarkable correlation between K_{WV} and K_{OW} provides a basis for the estimation of SPME efficiency based on the octanol/water partitioning properties of the organophosphorus pesticides. In general, the larger the K_{OW} value, the lower will be the SPME recovery at fixed water:sorbent ratio,

TABLE 2. Typical SPME Recoveries of the Four Organophosphorus Pesticides

pesticide[a]	recovery of SPME determination[b] (%)	RSD[c] (%)
phorate	12.83	6.1
diazinon	25.66	19.8
methyl parathion	23.49	17.3
ethion	2.41	1.5

[a] Concentrations of the pecticides range from 45 to 57 ppb. [b] Water: plant tissues ratio = 3:1, extraction time = 90 min. [c] $n = 15$.

TABLE 3. Estimated Method Detection Limits of the SPME Method for the Determination of Four Organophosphorus Pesticides in Food Plant *Chrysanthemum coronarium*

pesticide[a]	limit of detection[b,c] (ng/g)
phorate	9.4
diazinon	4.7
methyl parathion	5.1
ethion	75.0

[a] Water:plant tissues ratio at 3:1, extraction time = 90 min. [b] SPME recoveries of the pesticides in Table 2 are taken into account. [c] Based on fresh vegetable weight.

f. To improve SPME recovery, larger *f* value is required. As a rough estimation according to eq 10, $f \geq 18$ for diazinon and $f \geq 279$ for ethion are needed in order to achieve greater than 70% recovery. While handling of large amount of aqueous extract may not be practicable in route analysis, it is considered that lower recoveries may also be acceptable as long as good precision of recovery is maintained. Table 2 tabulates the typical recoveries for the SPME determination of the four organophosphorus pesticides at water:plant tissues ratio of 3:1. Relative standard deviations of SPME recoveries of all pesticides were within 20%, and this demonstrated the reproducibility of the SPME method. Figure 5 shows the relationship between SPME/GC-FPD response and the pesticide spike level. All four organophosphorus pesticides show good linearity up to the 100 ppb which should practically cover the working range of pesticide residue analysis in fruits and vegetables. The inferred detection limits are tabulated in Table 3. In general, detection limits down to part per billion level based on fresh vegetation can be obtained. Much lower detection limits should be able to be achieved using dried sample.

In conclusion, poly(dimethylsiloxane) SPME technique is feasible for the determination of organophosphorus pesticide residues in food plants. Good reproducibility and sensitivity are achieved with very simple analytical procedures. However, problem of discrimination due to solubility of pesticides exists. Although this can partly be alleviated by using a large excess of water for extraction (large *f* value), such dependence of SPME recovery on the K_{WV} value of the analyte may pose some uncertainties on its application as a quantification method for pesticide residues in agricultural food products. Nevertheless, the convenience, simplicity, and reliability of the SPME technique should undoubtedly make it a valuable semiquantitative tool for screening purposes. Also, the equilibrium model developed in this work may be extended to study distribution properties of chemical species in other biphasic systems, e.g., air/water and water/sediment partitioning systems.

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Literature Cited

(1) Sharp, G. J.; Brayan, J. G.; Dilli, S.; Haddad, P. R.; Desmarchellier, J. M. *Analyst* **1988**, *113*, 1493−1507.
(2) Muir, D. C. G. *J. Agric. Food Chem*. **1980**, *28*, 714−719.
(3) Barceló, D.; Porte, C.; Cid, J.; Albaigés, J. *Int. J. Environ. Anal. Chem*. **1990**, *38*, 199−209.
(4) Bellar, T. A.; Budde, W. L. *Anal. Chem*. **1988**, *60*, 2076−2083.
(5) Pinto, C. G.; Pavón, J. L. P.; Cordero, B. M. *Anal. Chem*. **1992**, *64*, 2334−2338.
(6) Pinto, C. G.; Pavón, J. L. P.; Cordero, B. M. *Anal. Chem*. **1995**, *67*, 2606−2612.
(7) Seidel, V.; Lindner, W. *Anal. Chem*. **1993**, *65*, 3677−3683.
(8) Junk, G. A.; Richard, J. J. *Anal. Chem*. **1988**, *60*, 451−454.
(9) Hinckley, D. A.; Bidleman, T. F. *Environ. Sci. Technol*. **1989**, *23*, 995−1000.
(10) Wells, M. J. M.; Michael, J. L. *Anal. Chem*. **1987**, *59*, 1739−1742.
(11) *Manual of Analytical Methods for the Analysis of Pesticides in Humans and Environmental Samples*; Watts, R. R., Ed.; EPA-600/8-80-038; U.S. Environmental Protection Agency, Environmental Toxicology Division, Health Effects Research Laboratory; Research Triangle Park, NC, 1980.
(12) Barceló, D. *Analyst* **1991**, *116*, 681−689.
(13) Sherman, J. *Anal. Chem*. **1993**, *59*, 40R−54R.
(14) Arthur, C. L.; Pawliszyn, J. *Anal. Chem*. **1990**, *62*, 2145.
(15) Arthur, C. L.; Killam, L. M.; Buchholz, K. D.; Pawliszyn, J. *Anal. Chem*. **1992**, *64*, 1960−1966.
(16) Louch, D. S.; Motlagh, S.; Pawliszyn, J. *Anal. Chem*. **1992**, *64*, 1187−1199.
(17) Arthur, C. L.; Killam, L.; Motlagh, S.; Potter, D. W.; Pawliszyn, J. *Environ. Sci. Technol*. **1992**, *26*, 979.
(18) Zhang, Z.; Pawliszyn. J. *Anal. Chem*. **1993**, *65*, 1843−1852.
(19) Zhang, Z.; Yang, M.; Pawliszyn, J. *Anal. Chem*. **1994**, *66*, 844A−853A.
(20) Potter, D. W.; Pawliszyn, J. *Environ. Sci. Technol*. **1994**, *28*, 298−305.
(21) Chen, J.; Pawliszyn. J. *Anal. Chem*. **1995**, *67*, 2530−2533.
(22) Steffen, A.; Pawliszyn. J. *J. Agric. Food Chem*. **1996**, *44*, 2187−2193.
(23) Górecki, T.; Pawliszyn, J. *Anal. Chem*. **1996**, *68*, 3008−3014.
(24) Poerschmann, J.; Zhang, Z.; Kopinke, F.-D.; Pawliszyn, J. *Anal. Chem*. **1997**, *69*, 597−600.
(25) Magdic, S.; Boyd-Boland, A.; Jinno, K.; Pawliszyn, J. *J. Chromatogr. A* **1996**, *736*, 219−228.
(26) Boyd-Boland, A. A.; Magdic, S.; Pawliszyn, J. *Analyst* **1996**, *121*, 929−937.
(27) Valor, I.; Moltó, J. C.; Apraiz, D.; Font, G. *J. Chromatogr. A* **1997**, *767*, 195−203.
(28) Johnson, W. E.; Fendinger, N. J. Plimmer, J. R. *Anal. Chem*. **1991**, *83*, 1510−1513
(29) Zee, S. Y.; Hui, L. H. *Hong Kong Food Plants*; The Urban Council: Hong Kong, 1981.
(30) Buchholz, K. D.; Pawliszyn, J. *Environ. Sci. Technol*. **1993**, *27*, 2844−2848.
(31) *The Pesticides Manual*, 10th ed.; Tomlin, C., Ed.; British Crop Protection Council: Faenham, Surrey, UK, 1994.
(32) Hansch, C.; Leo, A. *Substituent Constants for Correlation Analysis in Chemistry and Biology*; John Wiley & Son: New York, 1979.
(33) Collander, R. *Acta Chem. Scand*. **1950**, *4*, 1085−1098.

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