Pestic. Phytomed. (Belgrade), 23 (2008) 265-271
Pestic. fitomed. (Beograd), 23 (2008) 265-271

UDC: 632.95:543.393
Scientific paper * Naučni rad

Determination of Atrazine, Acetochlor, Clomazone, Pendimethalin and Oxyfluorfen in Soil by a Solid Phase Microextraction Method

Rada Đurović, Jelena Gajić-Umiljendić and Tijana Đorđević

Institute of Pesticides and Environmental Protection, 11080 Belgrade, Banatska 31b, Serbia (radda@ptt.rs)

SUMMARY

A solid phase microextraction (SPME) method for simultaneous determination of atrazine, acetochlor, clomazone, pendimethalin and oxyfluorfen in soil samples was developed. The method is based on a combination of conventional liquid-solid procedure and a following SPME determination of the selected pesticides. Initially, various microextraction conditions, such as the fibre type, desorption temperature and time, extraction time and NaCl content, were investigated and optimized. Then, extraction efficiencies of several solvents (water, hexane, acetonitrile, acetone and methanol) and the optimum number of extraction steps within the sample preparation step were optimized.

According to the results obtained in these two sets of experiments, two successive extractions with methanol as the extraction solvent were the optimal sample preparation procedure, while the following conditions were found to be most efficient for SPME measurements: 100 µm PDMS fibre, desorption for 7 min at 270^{0}C, 30 min extraction time and 5% NaCl content (w/v).

Detection and quantification were done by gas chromatography-mass spectrometry (GC/MS). Relative standard deviation (RSD) values for multiple analysis of soil samples fortified at 30 µg/kg of each pesticide were below 19%. Limits of detection (LOD) for all the compounds studied were less than 2 µg/kg.

Keywords: Solid phase microextraction; Pesticides; Soil

INTRODUCTION

One of the most time-consuming and difficult tasks in pesticide residue chemical analysis is the extraction and purification of target analytes from sample matrices, particularly from such highly complex ones as soil. Generally, routine procedures such as liquid-liquid extraction (LLE), soxhlet extraction and solid phase extraction (SPE) are time-consuming, tedious, require large quantities of organic solvents and are often relatively expensive. Therefore, recent trends in sample preparation have focused on developing simpler, fast-

er, more reliable and cost-efficient methods by reducing analysis time and solvent consumption. Solid phase microextraction (SPME), as a technique combining the extraction and concentration processes into one step, is an example of such development.

SPME is a simple, selective and efficient sorption/desorption method, based on the analytes' distibution between the sample matrix and extraction medium. Extraction is performed in a thin polymer film coating of a fused silica fibre, which is either immersed in a sample (DM/SPME) or exposed to a headspace above the sample (HS/SPME). After extraction, the fibre carrying the sorbed analytes is introduced into a gas chromatograph injector for thermal desorption (GC), while in case of liquid chromatography (LC) the analytes are desorbed by solvent elution.

So far, there have been scarce references to SPME application for determining pesticides in soil. Most of them are based on preparation of soil mixtures with distilled water and subsequent immersion of the SPME fibre in the slurry (Magdic et al., 1996) or its exposure to the gas phase above slurry (Ng et al., 1999; Castro et al., 2001; Doong and Liao, 2001; Navalon et al., 2002; Zhao et al., 2006; Fernandez-Alvarez et al., 2008). Some researchers have suggested that the DM/SPME of a soil organic extract obtained by solid-liquid extraction and diluted with an appropriate amount of water is the most reliable soil SPME method (Prosen and Zupancic-Kralj, 1998; Bouaid et al., 2001; Lambropoulou and Albanis, 2004). Their results indicate that this approach is more sensitive and provides both higher recoveries and better linearity. Most of these proposed methods, however, focus on simultaneous determination of pesticides belonging to only one or two pesticide groups. To our knowledge, there is actually only one report on SPME determination of pesticides that belong to several pesticide groups (chloroacetanilide, pyrethroid, organochlorine and organophosphorus compounds) (Fernandez-Alvarez et al., 2008). This method is based on headspace analysis of soil samples wetted with ultrapure water (50%, v/w).

As no previous studies are known to us dealing with the DM/SPME determination of pesticides of different pesticide groups, the intent of this study was to develop a rapid and simple DM/SPME method for simultaneous determination of 5 compounds having distinct chemical structures and belonging to different pesticide groups. The main parameters affecting DM/SPME procedures, such as the fibre type, temperature and time of desorption, extraction time and NaCl content, as well as the extraction efficiencies of several solvents (water, hexane, acetonitrile, acetone and methanol) and the optimum number of extraction steps within the sample preparation step were investigated and optimized.

MATERIAL AND METHODS

Reagents and materials

The pesticides chosen for this study were: clomazone, acetochlor, oxyfluorfen and pendimethalin (Dr Ehrenstorfer), and atrazine (Syngenta) (Table 1). Stock solutions (1 g/L) of each pesticide standard were prepared by dissolving the weighed amount in acetone (J.T. Baker, Deventer, Holland). The solutions were stored at -18°C. Working standard mixed solutions (10 mg/L and 1 mg/L of each compound) were pre-

Table 1. Pesticides studied and some of their physico-chemical properties[a]
Tabela 1. Pesticidi i neke njihove fizičko-hemijske osobine[a]

Pesticide Pesticid	Chemical group Hemijska grupa	M_r[b] (g/mol)	Water solubility Rastvorljivost u vodi (mg/L)	Log K_{ow}[c]	H[d] (Pam^3/mol)
Atrazine	Triazine	215.7	33	2.5	1.5×10^{-4}
Acetochlor	Chloroacetamide	269.8	223	4.14	3.83×10^{-1}
Oxyfluorfen	Diphenyl ether	361.7	0.116	4.47	9.40×10^{-2}
Pendimethalin	Dinitroaniline	281.3	0.3	5.18	2.73×10^{-3}
Clomazone	Isoxayolidinone	239.7	1100	2.5	4.19×10^{-3}

[a]Data cited from refs. – Podaci preuzeti iz referenci (Pesticide Manual, 2000-2001; www.sitem.herts.ac.uk/aeru/footprint/en/index.htm, 2008)
[b]Molecular weight – Molekulska masa
[c]Partition coefficient between n-octanol and water (as the log value) – Particioni koeficijent između n-oktanola i vode (predstavljen preko logaritamske vrednosti)
[d]Henry's constant – Henrijeva konstanta

pared weakly by diluting individual stock solutions with acetone and storing at 4°C. Water standard solutions (25 µg/L) were used for optimizing the SPME method. Highly purified deionized water (Purelab Option – R7, Elga, UK) was used to dilute the mixed acetone solutions. Sodium chloride (99.5% purity) was purchased from Merck (Darmstadt, Germany) and hexane, acetonitrile and methanol from J.T. Baker (Deventer, Holland).

The fibres used (Supelco, Bellefonte, PA, USA) were: 100 µm polydimethyl-siloxane (PDMS) and 85 µm polyacrilate (PA). Before use, the fibres were conditioned in the gas chromatograph injection port as recommended by the manufacturer. A magnetic stirrer (Roth RCT Basic, Germany) and 8 x 3 mm stirring bars were used to mix the samples during extraction. Extraction was performed in 4 ml vials (Supelco).

An uncontaminated soil sample originating from Kikinda was used in the study. The main physico-chemical properties of this soil were: pH (H_2O) = 8.39; organic matter content = 3.17%; sand content = 73.96%; silt content = 22.60%; clay content = 3.44%. The soil was air dried and sieved (2 mm) before using.

Polypropylene centrifuge tubes with caps (50 ml) (Sarstedt, Germany), filter papers 1PS, 150 mm diameter (Watman Int. Ltd., Maidstone, UK) and a centrifuge (UZ 4, Iskra, Slovenia) were used in the soil extraction procedure.

Instrumentation

A gas chromatograph-mass spectrometer (GC/MS) as a detection device (CP–3800/Saturn 2200, Varian, Australia) with 30 m x 0.25 mm x 0.25 µm VF-5ms column (Varian) was used. The GC was programmed as follows: initial temperature was 120°C, then increased to 170°C at 8°C/min and held for 4.5 minutes, increased to 280°C at 9°C/min and held for 5.5 minutes. Helium was used as the carrier gas and its flow rate was 1.1 ml/min.

The ion trap mass spectrometer was operated in the electron impact/selected ion monitoring (EI/SIM) mode. The ion trap and transferline temperatures were set to 220°C and 250°C, respectively. One specific pesticide ion was selected for detection and quantification, while a second one was used for confirmation. The ions inspected were as follows: 200 (215) for atrazine, 204 (125) for clomazone, 223 (146) for acetochlor, 252 (317) for oxyfluorfen and 252 (191) for pendimethalin.

Optimization of DM/SPME analysis

DM/SPME conditions, such as the fibre type, desorption temperature and time, extraction time and NaCl content, were investigated and optimized using 4 ml of aqueous solution containing 25 µg/L of each pesticide.

The following SPME conditions were found to be the most efficient for simultaneous extraction of the selected pesticides: 100 µm PDMS fibre, desorption for 7 min at 270°C, extraction time of 30 min, and 5% NaCl content (w/v).

Soil extraction optimization

Efficiency of the method optimized for SPME of aqueous solutions was tested in the analysis of soil samples. In that part of the study, sub-samples of 8 g were placed in polypropylene centrifuge tubes and fortified at 30 µg/kg level of each pesticide using 1 mg/L mixed standard solution. The spiked samples were homogenized for 15 min using a mehanical stirrer and left for 24 hours prior to further analysis.

The extraction efficiencies of various solvents (water, hexane, acetonitrile, acetone and methanol) and the optimum number of extraction steps were determined by the following procedure: soil samples were extracted with 15 ml of solvent for 30 min using a mehanical stirrer and then centrifuged for 15 min at 4000 rpm. The extract was filtered and evaporated to dryness at 35°C using a rotary evaporator (Devarot, Elektromedicina, Slovenia). The residues were redissolved in 1 ml of acetone, and 0.2 ml of these solutions were each diluted with water to a final volume of 10 ml for DM/SPME measurements. The presence of organic solvent (2%) was so prevented from affecting SPME measurements and fibre life (Eisert and Levsen, 1995a, 1995b; Urruty and Montury, 1996; Hernandez et al., 2000; Lambropoulou and Albanis, 2004).

RESULTS AND DISCUSSION

DM/SPME optimization

Different experimental parameters that affect SPME measurements were optimized using spiked water samples. Optimization was done by a well-structured step-by-step approach including choice of a most suitable SPME fibre, determination of optimal desorption temperature and time, extraction time and NaCl content.

Because polydimethyl-siloxane (PDMS) and polyacrylate (PA) fibres have been most throughly studied, and frequently described as the most efficient for pesticide extraction (Lambropoulou et al., 2000; Doong and Liao, 2001; Sakamoto and Tsutsumi, 2004; Fernandez-Alvarez et al., 2008), these fibres were chosen for our study. In order to determine the optimum desorption temperature and time, half-hour extraction procedures were performed at ambient temperature. In the first set of experiments, desorption temperature was varied from 265 to 285°C with 5 min desorption time. After that, desorption time was varied from 5 to 9 min at the chosen optimal desorption temperature. Between two measurements, desorption of a blank fibre was done each time to ensure that no residual compounds were present on the fibre. For the PA fibre, desorption for 7 min at 280°C was found to be optimal, while the corresponding optimum for the PDMS fibre was 7 min at 270°C. Finally, the PDMS fibre was found to be more appropriate for the mixture of selected pesticides and was therefore chosen for further work.

Time dependence of the amount of analytes extracted by the fibre was investigated at intervals ranging from 10 to 60 min. The results indicate that 30 min extraction time was enough to reach sorption equilibrium for atrazine, 45 min for acetochlor and 50 min for clomazone. On the other side, the 60 min interval was insufficient for pendimethalin and oxyfluorfen to overtake the sorption equilibrium. This is in line with the established fact that high molecular weight compounds, due to their low diffusion, and compounds that have low water solubility (higher affinity toward the SPME fibre) need longer extraction times to overtake equilibrium (Pawliszyn, 1997; Valor et al., 2001). Considering the pesticides' molecular weights, water solubility and log K_{ow} shown in Table 1, it is evident that our results are in accordance with the rules mentioned.

Although extraction using equilibrium time is recommended, some proposed theoretical models for explanation of the SPME process have indicated that quantification is possible before a sorption equilibrium is reached (Ai, 1997, 1998; Đurović et al., 2007), so that a 30 min extraction time, for practical reasons, was chosen in the following experiments. The time period of 30 min has been found enough time to provide sufficient analytical sensitivity for all compounds studied. Additionally, this interval was in accordance with the chromatographic run time (in our case 28.47 min), which ensured a maximum sample throughput when manual extraction was applied.

An addition of salt to a sample would decrease the solubility of some analytes in the aqueous phase, which stimulates their movement into the fibre coating (Pawliszyn, 1997). For that reason, the effect of ionic strength on the SPME process was studied by adding different amounts of NaCl to the water mixed standard solutions (0, 2.5, 5, 10 and 15% (w/v)).

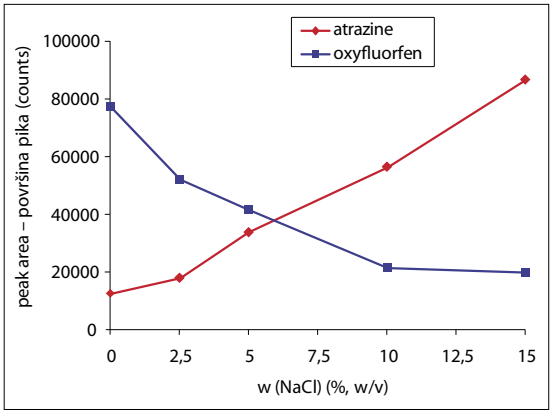

Figure 1. Effect of ionic strength on the analytical signal of atrazine and oxyfluorfen
Slika 1. Uticaj jonske jačine rastvora na analitički signal kod atrazina i oksifluorfena

The results (Figure 1) indicate that ionic strenght affects SPME efficiency in different ways and that the yield of SPME depends on the nature of each pesticide. Thus, based on compounds behaviour, considering their logK_{ow} values and solubility (Table 1), they can be classified into two groups. The first group includes compounds whose extraction efficiencies decrease as the percentage of NaCl added to the solution increases. This group consists of the more hydrophobic pesticides, such as pendimethalin and oxyfluorfen, which have high log K_{ow} values (5.18 and 4.47, respectively) and low water solubility (0.3 and 0.116 mg/L, respectively) (Table 1). The second group of compounds is made up of pesticides whose extraction yields increased with the increase of NaCl content. These compounds are characterized by high solubility in water and/or lower log K_{ow} values, as in the case of atrazine, clomazone and acetochlor (Table 1). Figure 1 shows the effect of ionic strength on analytical signals for atrazine and oxyfluorfen as the representative pesticides of each group.

Finally, considering the results obtained for all pesticides in this study, a 5% NaCl content was chosen as optimal (Figure 1).

Soil extraction optimization

Efficiency of the optimized SPME method was tested by analysing soil samples. As mentioned before, the DM/SPME of a soil organic extract obtained by conventional solid-liquid extraction diluted with an appropriate amount of water was shown to be a more efficient method than immersion of the SPME fibre in the slurry of soil sample and distilled water (Prosen and Zupancic-Kralj, 1998; Bouaid et al., 2001; Lambropoulou and Albanis, 2004). Therefore the former approach was chosen in the sample preparation step.

Extraction efficiencies of various solvents (water, hexane, acetonitrile, acetone and methanol) and the optimum number of extraction steps were determined by a well-structured step-by-step approach. At first, the most efficient solvent was chosen by applying a single extraction procedure as described in MATERIALS AND METHODS / Soil extraction optimization. In general, for most of the selected pesticides, the recoveries obtained with methanol were higher than those with other solvents, and methanol was therefore chosen for further work. The next step was to determine optimum extraction steps. Hence, the extraction of spiked soil samples with methanol was repeated up to four times under the same procedure. For most pesticides studied, the best recoveries were achieved after two extraction steps.

Finally, according to the results acquired in these two sets of experiments (Figure 2), two successive extractions with methanol as the extraction solvent were chosen as the optimal sample preparation procedure.

Validation of proposed method

Linearity of the developed method was tested in a concentration range from 2 to 600 μg/kg. The obtained arrangements and correlation coefficients (R) for all pesticides under study are presented in Table 2. It shows that the acquired correlation coefficients exceeded 0.996 for all compounds.

The limit of detection was computed as three times the base line noise (S/N = 3) at the lowest detectable concentration. LODs for all pesticides studied were equal or less than 1.52 μg/kg (Table 2).

Precision and confidence of the developed method were determined by performing four consecutive measurements of soil samples fortified at 30 μg/kg level. Both relative standard deviation (RSD) and recovery values are presented in Table 2. The table shows that RSDs for all pesticides under study were below 19%. For most of the analyzed pesticides, the recovery values were higher than 68%. An explanation for the lower recoveries of pendimethalin may be the strong influence of soil matrix on the pesticide and/or an insufficient power of methanol as an extraction solvent in the sample preparation step (Sparks, 1995).

The results presented suggest that the SPME method can be used for efficient and selective extraction of pesticides from complex matrix samples such as soil.

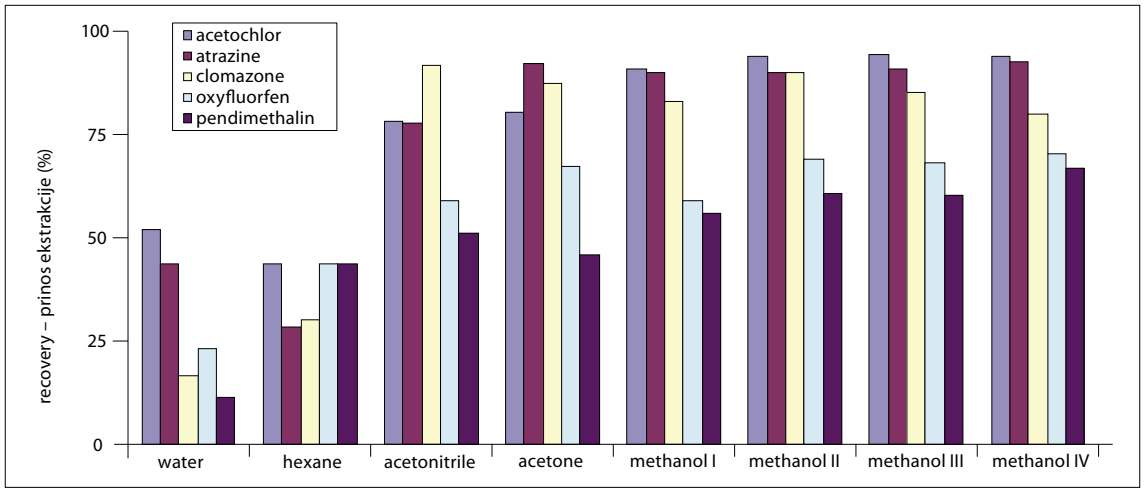

Figure 2. Dependence of extraction efficiency on: A) type of organic solvent and B) number of extraction steps, using the most efficient solvent

Slika 2. Zavisnost efikasnosti ekstrakcije od: A) vrste organskog rastvarača i B) broja ekstrakcionih koraka pri korišćenju najefikasnijeg rastvarača

Table 2. Linearity ranges (μg/kg), correlation coefficients (R), recoveries (%, n = 4), relative standard deviations (RSDs, %) and limits of detection (LODs, μg/kg) obtained for pesticides under study
Tabela 2. Opsezi linearnosti (μg/kg), korelacioni koeficijenti (R), prinosi ekstrakcije (%, n = 4), relativne standardne devijacije (RSDs, %) i granice detekcije (LODs, μg/kg) ispitivanih pesticida

Pesticide Pesticid	Concentration range Koncentracioni opsezi (μg/kg)	R	Recovery Prinosi ekstrakcije (%)	RSD (%)	LOD (μg/kg)
Atrazine	2 – 600	0.996	89.93	9.45	0.67
Acetochlor	2 – 600	0.998	93.77	7.79	0.18
Oxyfluorfen	2 – 600	0.999	68.82	15.20	0.27
Pendimethalin	2 – 600	0.999	60.66	18.56	1.52
Clomazone	2 – 600	0.999	89.87	4.31	0.12

ACKNOWLEDGEMENTS

This work was realized within Project No TR20041, supported by the Ministry of Science and Enviromental Protection of the Republic of Serbia.

REFERENCES

Ai, J.: Solid phase microextraction for quantitative analysis in non-equilibrium situations. Anal. Chem., 69: 1230-1236, 1997.

Ai, J.: Solid phase microextraction in headspace analysis. Dynamics in non-steady-state mass transfer. Anal. Chem., 70: 4822-4826, 1998.

Bouaid, A., Ramos, L., Gonzalez, M., Fernandez, P. and Camara, C.: Solid-phase microextraction method for the determination of atrazine and four organophosphorus pesticides in soil samples by gas chromatography. J. Chromatogr. A, 939: 13-21, 2001.

Castro, J., Perez, R.A., Sanchez-Brunete, C. and Tadeo, J.L.: Analysis of pesticides volatilized from plants and soil by headspace solid-phase microextraction and gas chromatography. Chromatographia, 53**:** S-361 – S-365, 2001.

Đurovic, R., Marković, M. and Marković, D.: Headspace solid phase microextraction in the analysis of pesticide residues – kinetics and quatification prior to the attainment of partition equilibrium. J. Serb. Chem. Soc., 72: 879-887, 2007.

Doong, R.A. and Liao, P.L.: Determination of organochlorine pesticides and their metabolites in soil samples using headspace solid-phase microextraction. J. Chromatogr. A, 918: 177-188, 2001.

Eisert, R. and Levsen, K.: Determination of organophosphorus, triazines and 2,6-dinitroaniline pesticides in aqueous samples via solid-phase microextraction (SPME) and gas chromatography with nitrogen-phosphorus detection. Fresenius J. Anal. Chem., 351: 555-562, 1995a.

Eisert, R. and Levsen, K.: Determination of pesticides in aqueous samples by solid-phase microextraction in-line coupled to gas chromatography-mass spectrometry. J. Am. Soc. Mass Spectrom., 6: 1119-1130, 1995b.

Fernandez-Alvarez, M., Llompart, M., Pablo Lamas, J., Lores, M., Garsia-Jares, C., Cela, R. and Dagnac, T.: Simultaneous determination of traces of pyrethroids, organochlorines and other main plant protection agents in agricultural soils by headspace solid-phase microextraction-gas chromatography. J. Chromatogr. A, 1188: 154-163, 2008.

Hernandez, F., Beltran, J., Lopez, F.J. and Gaspar, J.V.: Use of solid-phase microextraction for the quantitative determination of herbicides in soil and water samples. Anal. Chem., 72: 2313-2322, 2000.

Lambropoulou, D.A. and Albanis, T.A.: Determination of the fungicides vinclozolin and dicloran in soils using ultrasonic extraction coupled with solid-phase microextraction. Anal. Chim. Acta, 514: 125-130, 2004.

Lambropoulou, D.A., Konstantinou, I.K. and Albanis, T.A.: Determination of fungicides in natural waters using solid-phase microextraction and gas chromatography coupled with electron-capture and mass spectrometric detection. J. Chromatogr. A, 893: 143-156, 2000.

Magdic, S., Boyd-Boland, A., Jinno, K. and Pawliszyn, J.: Analysis of organophosphorus insecticides from environmental samples using solid-phase microextraction. J. Chromatogr. A, 736: 219-228, 1996.

Navalon, A., Prieto, A., Araujo, L. and Vilchez, J.L.: Determination of oxadiazon residues by headspace solid-phase microextraction and gas chromatography-mass spectrometry. J. Chromatogr. A, 946: 239-245, 2002.

Ng, W.F., Teo, M.J. and Lakso, H.A.: Determination of organophosphorus pesticides in soil by headspace solid-phase

microextraction. Fresenius J. Anal. Chem., 363: 673-679, 1999.

Pawliszyn, J.: Solid Phase Microextraction – Theory and Practice. Wiley-VCH, New York, USA, 1997.

Pesticide Manual: British Crop Protection Council – C D S Tomlin (12th ed.), 2000-2001.

Prosen, H. and Zupancic-Kralj, L.: Use of solid-phase microextraction in analysis of pesticides in soil. Acta Chim. Slov., 45: 1-17, 1998.

Sakamoto, M. and Tsutsumi, T.: Applicability of headspace solid-phase microextraction to the determination of multi-class pesticides in waters. J. Chromatogr. A, 1028: 63-74, 2004.

Sparks, D.L.: Environmental Soil Chemistry. Academic Press, Inc., San Diego, 1995.

Urruty, L. and Montury, M.: Influence of ethanol on pesticide extraction in aqueous solutions by solid-phase microextraction. J. Agric. Food Chem., 44: 3871-3877, 1996.

Valor, I., Perez, M., Cortada, C., Apraiz, D., Molto, J. C. and Font, G.: SPME of 52 pesticides and polychlorinated biphenyls: Extraction efficiencies of the SPME coatings polydimethylsiloxane, polyacrylate, polydimethylsiloxane-divinylbenzene, carboxen-polydimethylsiloxane and carbowax-divinylbenzene. J. Sep. Sci., 24: 39-48, 2001.

www.sitem.herts.ac.uk/aeru/footprint/en/index.htm. Accession date: June 21, 2008.

Zhao, R., Wang, X., Fu, S., Yuan, J., Jiang, T. and Xu, X.: A novel headspace solid-phase microextraction method for the exact determination of organochlorine pesticides in environmental soil samples. Anal. Bioanal. Chem., 384: 1584-1589, 2006.

Određivanje atrazina, acetohlora, klomazona, pendimetalina i oksifluorfena u zemljištu metodom mikroekstrakcije u čvrstoj fazi

REZIME

Za istovremeno određivanje atrazina, acetohlora, klomazona, pendimetalina i oksifluorfena u zemljištu ustanovljena je metoda mikroekstrakcije u čvrstoj fazi (SPME). Metoda je zasnovana na kombinaciji klasične tečno-čvrste pripreme uzorka i SPME određivanja. Izvršena je optimizacija najvažnijih mikroekstrakcionih parametara, kao što su izbor ekstrakcionog vlakna, desorpciona temperatura i vreme, ekstrakciono vreme i sadržaj natrijumhlorida (NaCl). Ispitivani su, takođe, ekstrakciona efikasnost različitih rastvarača (voda, heksan, acetonitril, aceton i metanol), kao i optimalan broj ekstrakcionih koraka u toku pripreme uzoraka zemljišta. Na osnovu rezultata dobijenih iz ova dva seta eksperimenata izabrani su sledeći radni uslovi: dve sukcesivne ekstrakcije sa metanolom kao ekstrakcionim rastvaračem u stupnju pripreme uzorka, dok su za mikroekstrakcione uslove uzeti: 100 µm PDMS vlakno, desorpcija u trajanju od 7 min na 270°C, ekstrakciono vreme – 30 min, uz dodatak NaCl (5%, w/v).

Detekcija i kvantifikacija analita je vršena gasno-masenim spektrometrom (GC/MS). Vrednosti relativnih standardnih devijacija (RSDs) višestruko ponovljenog merenja uzorka obogaćenog do 30 µg/kg svakog pesticida su ispod 19%. Granice detekcije (LODs) za sve pesticide uključene u ispitivanje su ispod 2 µg/kg.

Ključne reči: Mikroekstrakcija u čvrstoj fazi; pesticidi; zemljište