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Simultaneous liquid–liquid microextraction and polypropylene microporous membrane solid-phase extraction of organochlorine pesticides in water, tomato and strawberry samples

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ABSTRACT

A procedure involving the simultaneous performance of liquid–liquid microextraction and polypropylene microporous membrane solid-phase extraction was carried out. The applicability of the proposed procedure was evaluated through extraction of several organochlorine pesticides from river water, tomato and strawberry samples. The parameters affecting the extraction efficiency were optimized by multivariable designs, and the analytical features were estimated. Under optimized conditions, analytes were concentrated onto 1.5 cm long microporous membranes placed directly into the sample containing 15 mL of water with 20 μL of 1-octanol. The best extraction conditions were achieved at 59 °C, with 60 min of extraction time and 2.91 g of sodium chloride. The desorption of the analytes was carried out using 30 μL of a mixture of toluene and hexane in the proportion of 60:40% (v/v) for 10 min. Detection limits in the range of 2.7–20.0 ng L^{-1}, 0.50–1.15 μg kg^{-1}, and 1.53–12.77 μg kg^{-1} were obtained for river water, strawberry and tomato samples, respectively. Good repeatability was obtained for all three sample types. The results suggest that the proposed procedure represents a very simple and low-cost microextraction alternative rendering adequate limits of quantification for the determination of organochlorine pesticides in environmental and food samples.

1. Introduction

Despite the fact that in recent decades more attention has been focused on analyte separation and detection, significant advances in sample preparation techniques can be clearly noted. The interest in miniaturization in the area of analytical chemistry has led to the introduction of alternative techniques to substitute the conventional liquid–liquid extraction and solid-phase extraction procedures.

Among these alternative techniques, solid-phase microextraction (SPME), introduced by Pawliszyn et al. [1], basically initiated the miniaturized age in terms of sample preparation procedures in analytical chemistry [2]. Nowadays, SPME is a widely accepted and applied sample preparation technique, as it is a simple, relatively fast extraction and preconcentration procedure and is particularly attractive for the replacement of techniques that use solvents [3]. Another very important sample preparation technique is the liquid-phase microextraction (LPME) introduced firstly by Dasgupta et al. [4] and by Cantwell et al. [5] in 1996. LPME is simply a miniaturized format of liquid–liquid extraction (LLE), but it overcomes many of the disadvantages of LLE as well as some of those of SPME, such as sample carry-over [6].

Among the several possible configurations in which LPME can be performed, the use of a hollow fiber membrane (HF-LPME) to stabilize the extracting phase was introduced by Pedersen-Bjergaard and Rasmussen in 1999 [2]. HF-LPME can be conducted in two-phase or three-phase configurations [7]. The three-phase system consists of immobilizing a water-immiscible organic solvent in the wall pores of the HF while an aqueous acceptor solution is held within its lumen. Thus, analytes are extracted into the intermediary organic phase and subsequently into the aqueous phase. However, when both the wall pores and the HF lumen are filled with an organic solvent, a two-phase configuration is present. In both cases, after the extraction, the acceptor phase is directly injected into the analytical instrument. Recent reviews demonstrate the applicability of the two-phase and three-phase HF-LPME to several classes of compounds and matrices [2,6,8].

Recently, a new configuration for HF-LPME known as hollow fiber microporous liquid–liquid extraction (HF-MMLLE) has been proposed [9]. In this system only the organic solvent immobilized in the membrane pores as the acceptor phase is utilized for non-depleting extraction. After the extraction step, the organic solvent adsorbed in the fiber is desorbed in an adequate solvent prior the instrumental analysis. This alternative method was used to deter-

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mine 4-isobutylacetophenone in river water and sewage samples [9], as well as to determine more than 50 pesticides in alcoholic beverages [10]. Using the same configuration and based on the fact that the network of the microporous membrane can work as an adsorbent of analytes, Montes et al. [11] described the application of dry porous polypropylene membranes for the concentration of off-flavour anisoles in aqueous matrices. The extraction efficiency of the direct and headspace modes of the proposed approach, which has been named microporous membrane solid-phase extraction (MMSPE), was compared with the MMLLE procedure. Under optimized conditions similar precision and limits of quantification and remarkable linear responses were obtained for both techniques. Carpinteiro et al. [12] used the aforementioned techniques to determine poly-halogenated toluene in water samples. In both cases, after the extraction procedure, the analytes were desorbed in an adequate organic solvent prior the chromatographic analysis.

In this study, a simple and low-cost methodology based on the simultaneous application of liquid–liquid microextraction (LLME) and microporous membrane solid-phase extraction (MMSPE) is presented. The proposed procedure was applied to the concentration of organochlorine pesticides in river water, tomato and strawberry samples. Multivariate optimization of several variables potentially affecting the microextraction procedure was performed. The proposed procedure was compared with MMLLE and was found to be more efficient.

2. Experimental

2.1. Instrumentation

Chromatographic analysis was performed with a Shimadzu GC-14B gas chromatograph, equipped with split/splitless injector. An electron capture detector was used for the detection of organochlorine pesticides. Chromatographic separation was carried out in an OV-5 capillary column (30 m × 0.25 mm, 0.25 μm film thickness; OV Specialty Chemical, Marietta, OH). Ultrapure nitrogen was used as the carrier and make-up gas at 1.0 and 35 mL min^{-1}, respectively. Column oven temperature was 80 °C (4 min), 15 °C min^{-1} to 215 °C (1 min), 2 °C min^{-1} to 230 °C (3 min), and 5 °C min^{-1} to 260 °C (2 min). Injector and detector temperatures were fixed at 300 °C.

The target analytes extracted by LPME procedure from water, tomato and strawberry samples were identified using a Shimadzu GC-MS 2010 Plus. A Restek Rtx-5MS (5% diphenyl-95% dimethylpolysiloxane) capillary column (30 m × 0.25 mm × 0.25 μm) was used for the GC separation (Bellefonte, PA). Helium was used as the carrier gas at a flow rate of 1 mL min^{-1}. The oven temperature program and the injector temperature were the same as described previously for the GC. The quadrupole mass detector was operated at 260 °C in the electron impact mode at 70 eV. The ion source temperature was set at 220 °C, and the transfer line was set at 280 °C. The mass acquisition range was 100–500 *m/z*. The peaks were identified on the basis of their fragmentation patterns using the NIST Mass Spectral Search Program 05 (NIST, Washington, D.C.).

Other standard laboratory devices including a 320 Mettler Toledo pH meter, Microquímica MQAMA 301 stirrer, Ultra cleaner 1450 ultrasound bath and FANEM EXCELSA BABY II – 206R centrifuge were used for this study.

2.2. Reagents and solutions

All chemicals were of analytical grade and were used without prior purification. Deionized water from a Milli-Q Millipore® 18.2 MΩ cm^{-1} conductivity purification system (Bedford, MA, USA) was used to prepare all solutions. Prior to use, the laboratory glassware was kept for 24 h in 2% (v/v) Extran Merck (Darmstadt, Germany) and then rinsed with distilled water. It was then transferred to a 20% (v/v) nitric acid solution (Vetec, Rio de Janeiro, Brazil) where it remained for another 48 h followed by 1 h in an ultrasonic bath. Finally, the glassware was washed with deionized water and dried in a dust-free environment.

Organochlorine pesticide standards, including heptachlor, aldrin, heptachlor epoxide, endosulfan I, *p,p′*-DDE, dieldrin, endrin, endosulfan II and *p,p′*-DDD were purchased from Sigma–Aldrich (Milwaukee, WI, USA). Stock standard solutions of the studied pesticides were prepared in methanol and maintained at 4 °C. Working solutions used to optimize the parameters in LPME were prepared at a concentration of 10 ng mL^{-1} every day. Sodium chloride was bought from Nuclear (São Paulo, Brazil). 1-Octanol (analytical-grade from Merck), toluene and hexane (Tedia) were used as extracting solvents. Tomato and strawberry samples were purchased from the local market. River water samples were collected from the Cubatão River (Palhoça) and Araranguá River (Araranguá), both in Santa Catarina State, Brazil.

A Q 3/2 Accurel polypropylene hollow fiber membrane (600 μm id, 200 μm wall thickness and 0.2 μm pore size) was purchased from Membrana GmbH (Wuppertal, Germany). The hollow fiber was cut into segments with 1.5 cm length and was cleaned in acetone and dried before use.

2.3. Sample preparation procedure

Fresh sample (strawberry and tomato) was cut into small pieces and an aliquot of 100.00 g was homogenized using a food processor. One gram of the previously homogenized sample was weighed and placed in a 100 mL flask and spiked with an appropriate amount of the pesticide standard solution. After 3 days, the spiked sample was suspended with 15 mL of deionized water and the pH was adjusted.

The sample was submitted to ultrasonic vibration for 10 min and then centrifuged for 8 min at 4000 rpm. The aqueous phase was placed in a 20 mL glass vial containing 2.91 g of sodium chloride and a PTFE-coated magnetic stir bar. In the case of the river water samples, 15 mL of aqueous sample was added directly to the glass vial containing sodium chloride and the magnetic stirrer.

2.3.1. Simultaneous LLME and MMSPE procedure

After the sample preparation procedure, 20 μL of 1-octanol was placed in the glass vial containing 15 mL of the sample obtained, as described previously. A stainless steel wire was inserted through the silicone septa of the extraction vial and along the 1.5 cm length of the polypropylene hollow fiber membrane lumen. Therefore, only the outlet surface and the pores in the walls of the polypropylene hollow membrane were available for extraction of the analytes. This system was placed in the hole of the polypropylene screw top cap and used to seal the glass vial allowing the extraction phase to pass into the sample. The whole system was kept in a thermostatic bath on a magnetic stirrer, allowing controlled temperature and agitation during the extraction procedure.

Liquid desorption was performed by placing the MMSPE fiber previously removed from the plunger into 30 μL of toluene:hexane (60:40 v/v) contained in a micro-vial of 100 μL for 10 min without agitation after the extraction process had taken place. Finally, an aliquot of 1 μL was injected into the GC.

2.4. Optimization strategies

The optimization of the parameters affecting the pesticide extraction using the MMSPE fiber was performed using multivariate designs. A triangular surface mixture design was used to define the best extracting organic solvent (toluene, 1-octanol and hex-

ane) for the liquid desorption. A Box–Behnken design was applied to study the influence of NaCl addition, extraction time and extraction temperature on the extraction efficiency. The results obtained for these studies were applied to aqueous, tomato and strawberry samples. The amount of strawberry samples (0.250–1.000 g) and methanol added to the sample (0–200 μL), used as a co-solvent, was optimized through a central composite design. A univariate study to determine the influence of sample pH on the efficiency of the pesticide extraction from strawberry samples was evaluated. The best experimental conditions obtained for the strawberry sample were then applied to the tomato samples. In order to maximize the simultaneous pesticide extraction, for the three optimization designs the geometric average of the peak areas for the pesticides were used as the response for optimization in the computer programs, since good levels of detection for all pesticides were obtained. Furthermore, similar results were obtained when the peak areas for each pesticide were used as the response for the optimization. The experimental data were processed using the *Statsoft Statistica 6.0* computer program.

3. Results and discussion

3.1. Adsorption and desorption steps

A solvent suitable for the LPME technique should be easily immobilized in the pores of the polypropylene hollow fiber, have low volatility to prevent solvent loss, be immiscible with water and have a high partition coefficient for the analytes. According to the literature [8,13], 1-octanol is very appropriate for organochlorine pesticide extraction using the LPME technique. Therefore, in this study 1-octanol was selected as the extractor solvent. A preliminary study determined that the addition of 1-octanol in the range of 20–40 μL reached the maximum extraction efficiency. Therefore, a volume of 20 μL of the selected extractor solvent was used to ensure an excess of this solvent in relation to the absorption capacity of the polypropylene membrane.

The use of different solvents for the liquid desorption was studied through a triangular surface mixture design. In this case, toluene, hexane and 1-octanol were evaluated individually, through binary mixtures with 33% of one and 67% of another solvent (v/v), and a ternary mixture containing 33% of each solvent. The results obtained from this design can be seen in Fig. 1. Here, there is a region in which the response reaches high values, corresponding to the use of a mixture of toluene and hexane in the proportion of 60:40% (v/v). This condition was selected to continue the optimization of the proposed method. The effect of desorption time on the efficiency of the system was studied and it did not increase when contact times longer than 10 min were considered (data not shown).

3.2. Effect of extraction conditions for aqueous, tomato and strawberry samples

The factors ionic strength, extraction time and extraction temperature were optimized using the Box–Behnken design. Three levels for each factor were studied: temperatures of 23, 41.5 and 60 °C, extraction times of 20, 40 and 60 min and salt additions (NaCl) of 0, 2.5 and 5 g in 15 mL samples. From the results obtained, the combinations of the three factors were plotted generating three response surfaces (Fig. 2). Thus, quadratic regression equations were obtained for each response surface and the optimum value for each factor was obtained.

For extraction techniques based on the diffusion of analytes, the extraction temperature would be expected to have an important effect on the extraction efficiency. Fig. 2A shows that when

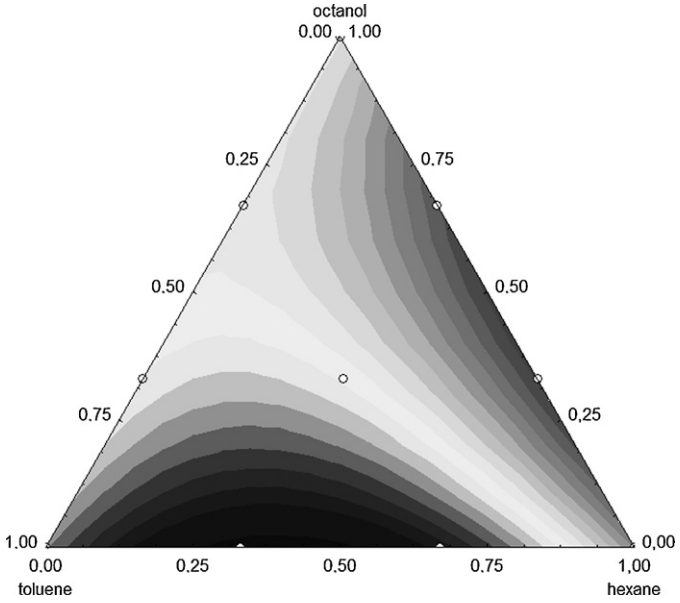

Fig. 1. Effect of different solvents used for liquid desorption (toluene, hexane and 1-octanol) after pesticide extraction by LPME. Experimental conditions: sample volume 20 mL, extraction at room temperature, extraction time 60 min and pesticide concentration 0.5 $\mu g\,L^{-1}$.

the extraction temperature is increased, a higher mass of salt is necessary to improve the extraction efficiency, probably due to an increase in the salt solubility with higher extraction temperatures. A relatively large region of maximum extraction efficiency can be observed around the central point. This behavior can be observed in Fig. 2B and C.

Supported membrane extractions need a long time to reach equilibrium, reducing the analytical frequency. Fig. 2B shows that on increasing the extraction time from 20 to 65 min an enhancement in the analytical signal can be observed until a plateau is reached, where a compromise between extraction efficiency and analytical frequency is established. The extraction temperature effect shows different behaviors. With an increase in the extraction temperature from 20 to 40 °C the analytical signal increases due to greater diffusion of the analytes in the sample. Between 40 and 70 °C the analytical signal remains almost constant and above this range there is a reduction in the extraction efficiency probably because of the reduction in the partition coefficient between the analytes and the membrane.

In Fig. 2C the effect of extraction time and sodium chloride addition on the extraction efficiency can be observed. The behavior of the extraction time is the same as that in Fig. 2B, indicating that the longest extraction time studied gives the highest extraction efficiency. The amount of salt placed in the vial directly influences the viscosity of the sample. Therefore, a higher amount of salt added causes a reduction in the analytical signal (Fig. 2C).

Thus, the region of maximum response corresponds to a temperature of 59 °C, extraction time of 60 min and addition of 2.91 g sodium chloride. These values were then applied in the remainder of the study.

3.3. Effect of sample mass, methanol volume and pH on the extraction efficiency for tomato and strawberry samples

Samples with very complex matrices require a more detailed study to verify variables that could interfere in the analysis. A strawberry matrix, for example, contains macromolecules and dyes that can interfere in the extraction procedure due to the complex interaction between the analytes and the matrix. The use of solvents as

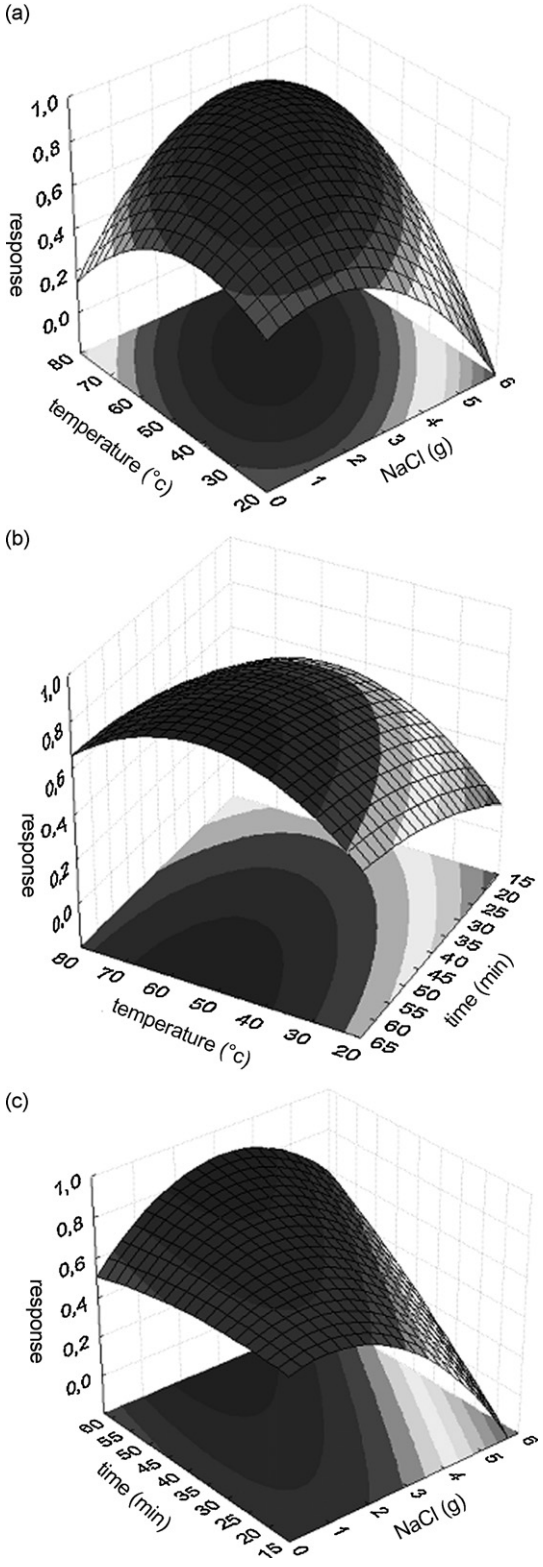

Fig. 2. Surface response to optimize the variables amount of salt added, extraction time and extraction temperature. (A) Effect of extraction temperature and addition of salt on the extraction efficiency. (B) Effect of extraction temperature and time on the extraction efficiency. (C) Effect of extraction time and addition of salt on the extraction efficiency. Experimental conditions: sample volume 15 mL, 1-octanol volume 20 μL, extraction temperature from 23 to 70 °C, extraction time from 20 to 60 min and amount of salt between 0 and 5 g.

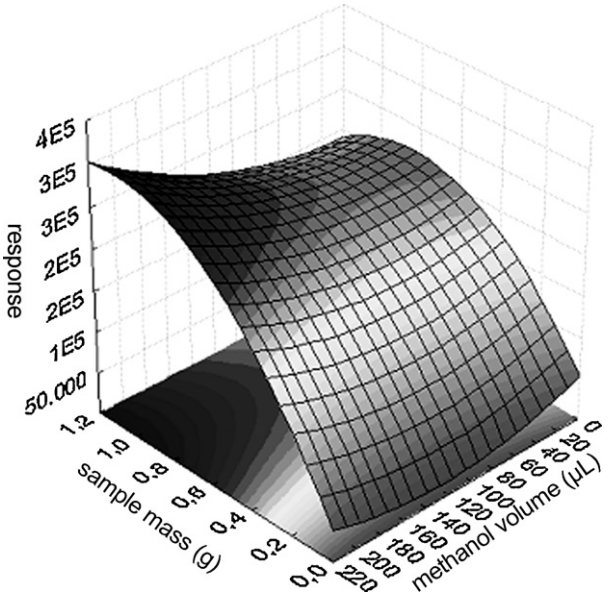

Fig. 3. Surface response to optimize the variables amount of sample and addition of methanol. Experimental conditions: sample mass 0.250–1.000 g, methanol volume 0–200 μL, 0.5 μg kg^{-1} of each pesticide, 1-octanol volume 20 μL, extraction temperature 59 °C, extraction time 60 min and amount of salt 2.91 g.

an auxiliary for the extraction process has been used to facilitate the release of the analytes from the matrix. In this study, the effect of the addition of methanol as a co-solvent on the extraction efficiency before submitting the sample to ultrasonic vibration was studied. The amount of sample submitted to the extraction procedure is another important variable that can affect the extraction efficiency. A central composite design with two factors and five levels was used to verify the effects of the addition of methanol and the amount of sample on the extraction efficiency.

The results obtained can be seen in Fig. 3, which shows an improvement in the extraction efficiency on increasing the amount of sample. On the other hand, the addition of methanol to the extraction system did not lead to significant recovery of the pesticides from the strawberry samples. Therefore, 1.000 g of sample and no addition of methanol was fixed and used throughout.

Due to the presence of natural ionizing dyes in the strawberries, it is possible to avoid the interaction of these species with the solvent/membrane system by adjusting the sample pH. Therefore, the effect of sample pH on the extraction efficiency was study for the strawberry (Fig. 4A) and tomato (Fig. 4B) samples. Fig. 4 shows that with sample pH values close to 2 and 4, respectively, an enhancement in the analytical signal for almost all pesticides studied was obtained. In a suitably acid medium, most of the natural dye content of the fruit and vegetable studied is present in the ionized form, and is not extracted by the membrane. Furthermore, an acid medium can aid the release the analytes from the matrix. Fixed pH values of 2 and 4 were thus used for the strawberry and tomato samples, respectively, in the rest of the study.

3.4. Comparison between LLME–MMSPE and MMLLE procedures

The use of higher extraction temperatures not only improves the extraction efficiency, because of its influence on the analyte diffusion, but also affects the sample viscosity and the solubility of the extractor solvent in the sample, causing the degradation (degeneration) of the membrane in the supported liquid membrane (SLM). This degradation effect can be minimized by adding an adequate amount of solvent to the sample, which constantly renews the liquid membrane. Thus, the solvent introduced directly into

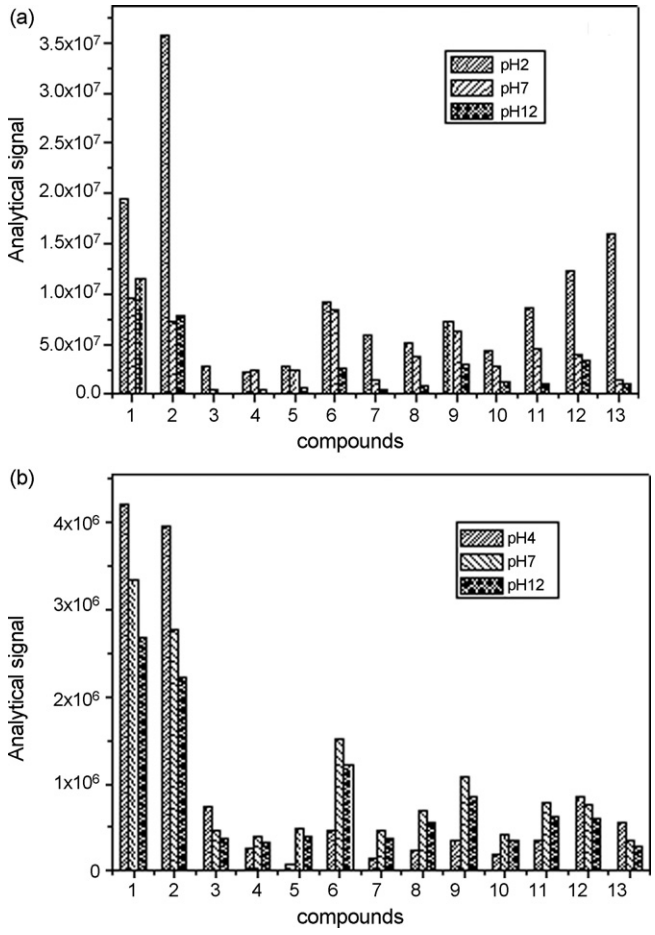

Fig. 4. Effect of sample pH on the analytical signal for (A) strawberry matrix and (B) tomato matrix. (1) α-HCH; (2) β-HCH; (3) Δ-HCH; (4) Heptachlor; (5) Aldrin; (6) Heptachlor epoxide; (7) Endosulfan I; (8) 4,4′-DDE; (9) Dieldrin; (10) Endrin; (11) Endosulfan II; (12) 4,4′-DDD; (13) 4,4′-DDT. Experimental conditions: liquid sample volume 15 mL, 0.5 μg kg^{-1} of each pesticide, extraction time 60 min, extraction temperature 59 °C, NaCl mass 2.91 g and sample mass 1.000 g.

the sample functions not only as an extractor in the liquid–liquid extraction process but also avoids degradation of the membrane. At the same time, the introduction of a dry porous polypropylene membrane into the sample initiates the process of the extraction of this solvent, containing the pesticides, to the pores of the hollow fiber, where it remains bound by capillary forces. The proposed LLME–MMSPE procedure showed higher extraction efficiency than when the extractor solvent was impregnated only into the membrane pores (MMLLE), as can be seen in Fig. 5. The better extraction efficiency when 1-octanol was introduced directly into the sample can be attributed to the formation of fine droplets of the extractor solvent in the bulk of the sample, allowing better interaction between the analytes and 1-octanol. A chromatogram obtained for extraction of the organochlorine pesticides from strawberry (10 μg kg^{-1}) using LLME–MMSPE procedure and the optimized conditions is presented in Fig. 6.

3.5. Analytical figures of merit and accuracy

From the results obtained in the optimization procedure, the analytical figures of merit were investigated for each type of sample (water, strawberry and tomato). Calibration curves were constructed to estimate the linear range, correlation coefficients, and detection and quantification limits for the proposed LLME–MMSPE method. The limits of detection and quantification

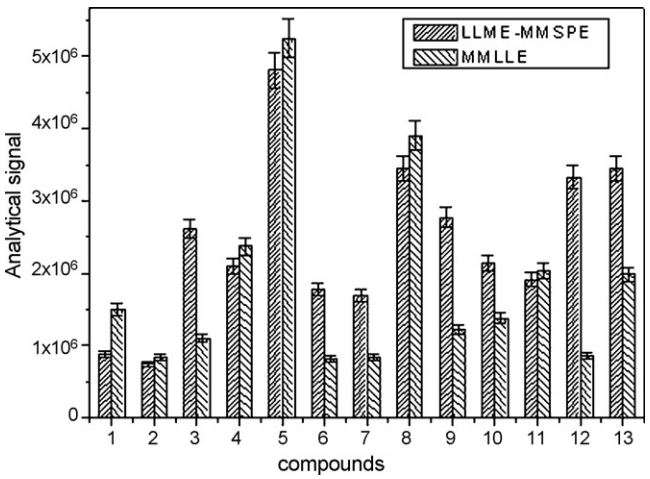

Fig. 5. Effect of an excess of 1-octanol added to the sample on the analytical signal for strawberry matrix. (1) α-HCH; (2) β-HCH; (3) Δ-HCH; (4) Heptachlor; (5) Aldrin; (6) Heptachlor epoxide; (7) Endosulfan I; (8) 4,4′-DDE; (9) Dieldrin; (10) Endrin; (11) Endosulfan II; (12) 4,4′-DDD; (13) 4,4′-DDT. Experimental conditions: liquid sample volume 15 mL, 0.5 μg kg^{-1} of each pesticide, extraction time 60 min, extraction temperature 59 °C, NaCl mass 2.91 g and sample mass 1.000 g.

were calculated as two and ten times the signal to noise ratio, respectively.

The results obtained for water, strawberry and tomato samples are summarized in Tables 1–3, respectively. Good correlation coefficients (R) were obtained for all matrices studied. For the river water samples, the method showed an excellent precision, calculated as the relative standard deviation ($n = 7$) using solutions spiked with 0.4 μg L^{-1} of each pesticide, in the range of 5.1–15.0%. Relative recovery assays were carried out for strawberry and tomato samples using two levels of concentration (20 μg kg^{-1} for strawberry samples and 50 μg kg^{-1} for tomato samples) showing excellent results considering the complexity of the samples. The pesticide 4,4′-DDD cannot be quantified because of its co-elution with some components of the strawberry matrix. The proposed method presented good LOD and LOQ, probably because of the excellent sample clean-up promoted by the membrane, verifying its suitability for the determination of pesticides in vegetables and fruits. The LOD values for the proposed procedure are similar those obtained for methods based on HF-LPME applied to aqueous envi-

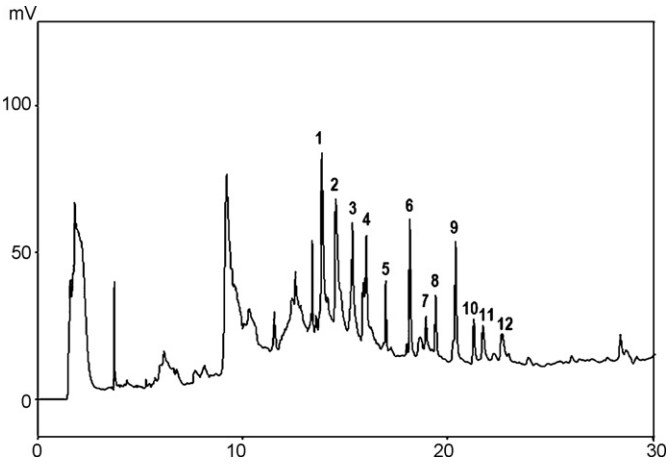

Fig. 6. GC-ECD chromatogram of extracted organochlorine pesticides from strawberry (10 ng/g of cork) using LLME–MMSPE procedure and the optimized conditions (water sample volume 15 mL, extraction time 60 min, extraction temperature 59 °C, NaCl mass 2.91 g and sample mass 1.000 g). (1) α-HCH; (2) β-HCH; (3) Δ-HCH; (4) Heptachlor; (5) Aldrin; (6) Heptachlor epoxide; (7) Endosulfan I; (8) 4,4′-DDE; (9) Dieldrin; (10) Endrin; (11) Endosulfan II; (12) 4,4′-DDD.

Table 1
Linear range, precision, correlation coefficients, and detection and quantification limits obtained for the method proposed to determine pesticides in water samples using the polypropylene membrane.

Compound	Linear range (ng L^{-1})	R^2	LOD[a] (ng L^{-1})	LOQ[b] (ng L^{-1})	RSD
α-HCH	10–600	0.9988	2.7	8.4	10.4
β-HCH	40–600	0.9996	13.0	44.0	14.5
Δ-HCH	60–600	0.9959	20.0	60.0	14.5
Heptachlor	35–600	0.9988	9.2	39.6	9.1
Aldrin	35–600	0.9981	11.0	39.6	10.4
Heptachlor epoxide	30–600	0.9996	9.9	33.0	6.2
Endosulfan I	20–600	0.9998	8.6	28.0	5.1
4,4′-DDE	40–600	0.9978	12.0	42.9	10.7
Dieldrin	40–600	0.9997	13.7	45.0	15.0
Endrin	30–600	0.9996	13.3	31.1	6.6
Endosulfan II	30–600	0.9976	10.0	35.0	5.9
4,4′-DDD	55–600	0.9934	18.0	59.9	8.9
4,4′-DDT	20–600	0.9988	7.9	26.0	9.9

[a] LOD: limit of detection.
[b] LOQ: limit of quantification.

Table 2
Linear range, correlation coefficients, detection and quantification limits, concentration in the sample and recovery tests obtained for the proposed method to determine pesticides in strawberry samples using the polypropylene membrane.

Compound	Linear range (μg kg^{-1})	R^2	LOD (μg kg^{-1})	LOQ (μg kg^{-1})	Found concentration	Relative recovery (%)
α-HCH	2.0–70.0	0.997	0.57	1.90	<LD	83.9 ± 11.9
β-HCH	2.0–70.0	0.992	0.60	2.00	<LD	70.0 ± 6.7
Δ-HCH	2.5–100.0	0.992	0.69	2.30	<LD	94.4 ± 15.0
Heptachlor	2.0–100.0	0.996	0.67	2.24	<LD	123.0 ± 12.9
Aldrin	2.5–100.0	0.993	0.77	2.59	<LD	90.6 ± 11.4
Heptachlor epoxide	2.0–100.0	0.995	0.63	2.10	<LD	101.6 ± 10.9
Endosulfan I	2.0–100.0	0.999	0.50	1.69	<LD	82.7 ± 6.6
4,4′-DDE	2.0–100.0	0.995	0.73	2.46	3.7 ± 0.1	74.4 ± 9.4
Dieldrin	3.0–100.0	0.999	1.10	3.30	<LD	105.9 ± 14.5
Endrin	3.5–100.0	0.999	1.15	3.85	<LD	72.4 ± 8.5
Endosulfan II	3.0–100.0	0.999	1.03	3.44	<LD	99.0 ± 9.0
4,4′-DDT	3.0–50.0	0.998	1.06	3.50	<LD	104.9 ± 6.7

ronmental matrices, which are between 0.001 and 0.047 μg L^{-1} [14–16]. The determination of the same pesticides in aqueous samples using the SDME technique gave LOD values between 0.01 and 20 μg L^{-1} [17,18]. In summary, the proposed method offers extraction efficiencies similar to or better than others described in the literature.

3.6. Application of the methodology to river water samples

The proposed method was also applied in the analysis of two river water samples collected from the Cubatão River (Palhoça) and Araranguá River (Araranguá), both in Santa Catarina, Brazil, close to tomato and corn plantations. The analytes were quantified using the addition calibration technique and recovery tests were performed spiking each sample with 0.2 μg L^{-1} of each pesticide. This procedure was carried out in triplicate and the results can be observed in Table 4.

As can be observed in the table, for the samples from the Cubatão River and Araranguá River it was possible to quantify five of the nine pesticides investigated. Furthermore, the data shown in Table 4 indicate that the accuracy of the method can be considered satisfactory; especially taking into consideration that no previous sample preparation was carried out before the preconcentration procedure.

Table 3
Linear range, correlation coefficients, detection and quantification limits, concentration in the sample and recovery tests obtained for the proposed method to determine pesticides in tomato samples using the polypropylene membrane.

Compound	Linear range (μg kg^{-1})	R^2	LOD (μg kg^{-1})	LOQ (μg kg^{-1})	Relative recovery (%)
α-HCH	2–80	0.999	1.53	5.09	103.8 ± 10.3
β-HCH	10–230	0.999	2.90	9.66	75.3 ± 7.2
Δ-HCH	20–230	0.996	6.46	21.52	59.3 ± 2.5
Heptachlor	20–230	0.997	6.49	21.63	69.6 ± 8.6
Aldrin	30–230	0.992	9.93	33.06	82.6 ± 12.3
Heptachlor epoxide	10–230	0.998	3.60	12.03	96.3 ± 9.6
Endosulfan I	5–230	0.999	1.65	5.49	90.8 ± 8.8
4,4′-DDE	10–230	0.997	4.06	13.53	95.8 ± 8.1
Dieldrin	10–230	0.998	3.78	12.60	109.7 ± 10.9
Endrin	25–230	0.988	8.12	27.05	114.4 ± 7.7
Endosulfan II	40–230	0.986	12.77	42.53	91.9 ± 12.4
4,4′-DDD	20–230	0.995	6.20	20.66	100.1 ± 5.0
4,4′-DDT	10–230	0.997	3.88	12.93	116.9 ± 11.6

Table 4
Results obtained for the determination of the target pesticides using the proposed method and recovery tests.

Compound	Cubatão River		Araranguá River	
	Found concentration ($ng\,L^{-1}$)	Relative recovery (%)	Found concentration ($ng\,L^{-1}$)	Relative recovery (%)
α-HCH	<LD	103.5 ± 3.7	<LD	108.8 ± 8.9
β-HCH	<LD	90.0 ± 1.4	<LD	118.3 ± 8.3
Δ-HCH	<LD	115.0 ± 2.3	73.8 ± 13.6	96.6 ± 8.7
Heptachlor	11.8 ± 3.5	86.3 ± 11.6	<LD	109.4 ± 11.2
Aldrin	161.0 ± 25.1	106.1 ± 10.0	<LD	116.6 ± 12.7
Heptachlor epoxide	<LD	94.9 ± 8.5	<LD	86.3 ± 10.1
Endosulfan I	<LD	104.9 ± 9.8	<LD	81.2 ± 7.9
4,4′-DDE	76.0 ± 25.5	105.6 ± 4.5	<LD	103.3 ± 14.7
Dieldrin	44.0 ± 19.9	107.5 ± 4.8	<LD	110.0 ± 9.2
Endrin	<LD	105.7 ± 1.3	<LD	119.1 ± 8.2
Endosulfan II	<LD	107.5 ± 1.5	<LD	118.7 ± 10.8
4,4′-DDD	<LD	115.0 ± 3.5	<LD	113.0 ± 10.2
4,4′-DDT	<LD	96.3 ± 6.5	<LD	112.5 ± 15.0

4. Conclusions

The simultaneous use of LLME and MMSPE procedures to determine organochlorine pesticides in diverse matrices provided low limits of detection ($ng\,L^{-1}$) and good precision and linearity. The proposed method presents the advantages and drawbacks of the HF-LPME, as it is simple, effective, low cost, uses microliters of organic solvents, is almost free of matrix effects, and completely avoids problems associated with carry-over. On the other hand, HF-LPME is relatively inefficient for the most polar substances. However, the proposed combination between LLME and MMSPE leads to an improvement in the extraction efficiency. To the best of our knowledge, this constitutes the first study that applies simultaneous LLME and polypropylene MMSPE as a preconcentration procedure.

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References

[1] C.L. Arthur, J. Pawliszyn, Anal. Chem. 62 (1990) 2145.
[2] S. Pedersen-Bjergaard, K.E. Rasmussen, J. Chromatogr. A 1184 (2008) 132.
[3] T.P. Gbatu, K.L. Sutton, J.A. Caruso, Anal. Chim. Acta 402 (1999) 67.
[4] H. Liu, P.K. Dasgupta, Anal. Chem. 68 (1996) 1817.
[5] M.A. Jeannot, F.F. Cantwell, Anal. Chem. 68 (1996) 2236.
[6] J. Lee, H.K. Lee, K.E. Rasmussen, S. Pedersen-Bjergaard, Anal. Chim. Acta 624 (2008) 253.
[7] Y. Tao, J.-F. Liu, T. Wang, G.-B. Jiang, J. Chromatogr. A 1216 (2009) 756.
[8] D.A. Lambropoulou, T.A. Albanis, J. Biochem. Biophys. Methods 70 (2007) 195.
[9] S. Zorita, T. Barri, L. Mathiasson, J. Chromatogr. A 1157 (2007) 30.
[10] P.P. Bolanos, R. Romero-Gonzáles, A.G. Garrido Frenich, J.L. Martínez Vidal, J. Chromatogr. A 1208 (2008) 16.
[11] R. Montes, I. Rodríguez, E. Rubi, M. Ramil, R. Cela, J. Chromatogr. A 1198 (2008) 21.
[12] M.I. Carpinteiro, I. Rodríguez, R. Cela, M. Ramil, J. Chromatogr. A 1216 (2009) 2825.
[13] H. Pan, W. Ho, Anal. Chim. Acta 527 (2004) 61.
[14] C. Basheer, V. Suresh, R. Renu, H.K. Lee, J. Chromatogr. A 1033 (2004) 213.
[15] C. Basheer, R. Balasubramanian, H.K. Lee, J. Chromatogr. A 1016 (2003) 11.
[16] C. Basheer, H.K. Lee, J.P. Obbard, J. Chromatogr. A 968 (2002) 191.
[17] T. Barri, S. Bergström, A. Hussen, J. Norberg, J.Å. Jönsson, J. Chromatogr. A 1111 (2006) 11.
[18] L. Zhao, H.K. Lee, J. Chromatogr. A 919 (2001) 381.