



A SDME/GC–MS methodology for determination of organophosphate and pyrethroid pesticides in water

Anselmo de Souza Pinheiro, Gisele Olimpio da Rocha, Jailson Bittencourt de Andrade *

Instituto de Química-Universidade Federal da Bahia, Campus de Ondina, 40170-290, Salvador-BA, Brazil

Centro Interdisciplinar de Energia e Ambiente-CIEnAm, Universidade Federal da Bahia, Canela, 40110-040, Salvador-BA, Brazil

INCT de Energia e Ambiente, UFBA, 40170-290, Salvador, BA, Brazil

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ABSTRACT

A rapid and simultaneous method for identification and quantification of pesticides residues in water samples have been developed and applied to the analysis of real samples. Tap and San Francisco River water samples were collected from Propria town and Aracaju city in the state of Sergipe, Brazil. A new single-drop microextraction (SDME) followed by gas chromatography–mass spectrometry techniques were used to determine the dimethoate, methyl parathion, ethion (organophosphates) and permethrin (pyrethroid) pesticides in water samples. The parameters linearity, linear range, precision, accuracy, sensitivity and robustness were studied for validation of the SDME/GC–MS method. An important point to this study is that plots of relative response and logarithmic concentrations were used to verify that the measurements were within the linear dynamic range of the method. In order to enhance high linearity of analytical curve, points that do not belong to 95 to 105% of linear range were excluded. Recovery tests of pesticides in different water samples (tap water and river water) were between 76.2 and 107% and this evaluation was used to demonstrate the reliability of the method. For all pesticides the method showed the limits of detection (LOD) in a range between 0.05 and 0.38 $\mu\text{g L}^{-1}$ and the limit of quantification (LOQ) between 0.15 and 1.1 $\mu\text{g L}^{-1}$. All these parameters demonstrate high sensitivity of the developed method and the capability for detecting and quantifying of low levels of pesticides in water samples.

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1. Introduction

The generic term “pesticide” identifies a wide spectrum of synthetic compounds with biocide activity used to remove weeds, fungi and insects to increase agricultural productivity [1]. The toxicity of pesticides and their corresponding harmful environmental effects are increasingly evident and have aroused interest in the identification and quantification of such compounds in various matrices, such as juices [2], vegetables [3], fruits [4], milk [5], soils [6] and water [1,6,7]. Thus, it is of paramount importance to develop faster and more selective analytical methodologies with lower cost-benefit ratios that are less harmful to the environment and more sensitive to trace levels of pesticide residues in various matrices including natural and drinking waters [7,8].

The single drop microextraction (SDME), is a relatively new technique that has been increasingly used in the analysis of pesticides because it is simple, cheap, fast, effective and virtually free of organic solvents [9] and currently has become a powerful tool to analyze different groups of analytes in various matrices [10–13], with an

emphasis on the determination of pesticides in water [7,9,14–27]. The SDME technique is based on the principle of a distribution of analytes between an organic solvent microdrop and an aqueous phase. The analytes with high partition coefficient can reach high concentrations, once they are transferred by diffusion from a significant volume of sample (1–5 mL) to a microdrop of organic solvent (5–50 μL), thus obtaining a high enrichment factor [28]. The SDME procedure uses a microsyringe with the needle immersed into a water sample containing the analytes. The needle hangs a drop of the solvent while the sample is stirred. After extraction, the drop is aspirated back into the microsyringe and then injected into a gas chromatograph (GC) or liquid chromatograph (LC) [7,21,25,28–30]. The SDME procedure has the advantage of combining both a pre-concentration and sample introduction steps into a single-step extraction [31] which can widely be used in the determination of organic [32,33] and inorganic analytes [34–36]. However some problems such as microdrop instability and solvent loss during extraction may reduce the repeatability of the method [28,37].

Due to the environmental potential and human toxicity of pesticides and pyrethroids, development and optimization of sensitive extraction methods for these compounds are required. When using SDME procedure, it is necessary to optimize parameters that control mass transfer and hence improve the efficiency of the

* Corresponding author at: Instituto de Química-Universidade Federal da Bahia, Campus de Ondina, 40170-290, Salvador-BA, Brazil. Tel.: +55 7132375524; fax: +557132636435.

E-mail address: jailson@ufba.br (J.B. de Andrade).

procedure [9]. Usually the parameters to be optimized are type of extraction and time, solvent, stirring speed, ionic strength, pH, temperature effect of the aqueous phase and the drop volume of extraction solvent. Therefore, the aim of this study was to develop and validate a sensitive SDME/GC–MS method to determine some widespread used organophosphate and pyrethroid pesticides in water samples.

2. Experimental

2.1. Reagents and solutions

Chromatographic grade methanol was purchased from Merck (Darmstadt, Germany) and pesticide standards of dimethoate (99.6%), methyl parathion (99.6%), permethrin (99%), and ethion (99.6%) were purchased from AccuStandard (New Haven, USA). Stock standard solutions were prepared in methanol at concentration of $200\text{ }\mu\text{g mL}^{-1}$. Analytical standard solutions of each pesticide were prepared at a concentration of $5\text{ }\mu\text{g mL}^{-1}$.

2.2. Sampling

In the present study a total of 16 water samples were collected from two sites. First, 9 river water samples were collected from an irrigation project field “Perimetro Irrigado Propriá”, in the town of Propriá (State of Sergipe, Brazil: $10^{\circ}12'40''\text{S}$, $36^{\circ}50'25''\text{W}$). The site is located at the right border of the San Francisco River where rice is the main crop cultivated. Second, tap water samples from two cities, Propriá (5 water samples) and Aracaju (State of Sergipe, Brazil: $10^{\circ}55'56''\text{S}$, $37^{\circ}04'23''\text{W}$) (2 samples of tap water) were also collected.

2.3. The SDME procedure

The SDME standard procedure published by Pinheiro and de Andrade [7] was adopted in this study. In this procedure a $10\text{ }\mu\text{L}$ microsyringe was used for measuring $1\text{ }\mu\text{L}$ of extraction solvent and then introduced in the glass vial. Firstly, needle of the microsyringe was inserted through the septum and directly immersed into the water sample (10 mL) containing the analytes. Secondly, the micro-syringe plunger was depressed and then the microdrop was formed. Thirdly, we allowed the microdrop to get in contact to water sample for 30 min under agitation (300 rpm). After that, microdrop ($1\text{ }\mu\text{L}$) was drawn back into syringe and finally, the syringe was removed from the vial and immediately injected in a GC–MS. The GC–MS running was 24.0 min and the whole SDME/GC–MS determination took 54.0 min.

2.4. GC–MS analysis

The analysis of pesticides was performed by using Shimadzu GC-2010 system coupled with QP2010 mass spectrometer detector (Kyoto, Japan). The fused-silica capillary column ($30\text{ m} \times 0.32\text{ mm}$) coated with $0.25\text{-}\mu\text{m}$ bonded film of DB-1 (J & W Scientific, Folsom, CA) was used. The GC column temperature program used was as follows: 60°C for 1 min, then ramped to 150°C at a rate of $20^{\circ}\text{C min}^{-1}$ (held for 4 min) and then ramped to 290°C at a rate of $15^{\circ}\text{C min}^{-1}$ (held at this temperature for 5.17 min). The injector and detector temperatures were 250°C , and all injections were made in the split/splitless mode (splitless time: 0.75 min). Helium was used as carrier gas at a flow rate of 1.2 mL min^{-1} . A $1\text{ }\mu\text{L}$ sample volume was injected at 280°C . The mass spectrometer was operated in the electron impact mode with an ion source temperature of 250°C and the electron impact energy was set at 70 eV. The MS scanned mass range m/z 40 and 300 was used for quantitative determinations of the studied pesticides. For quantitative determination using selective ion monitoring (SIM), pesticides were identified by ions with the

following m/z values and quantified by the ions 87, 93 and 125 for dimethoate (retention time: 13.41 min); 109, 125 and 263 for methyl parathion (retention time: 14.95 min); 97, 153 and 231 for ethion (retention time: 17.63 min); 163 and 183 for permethrin (retention time: 20.15 min). Quantification was performed by calculating the absolute peak areas. Fig. 1 illustrates a standard chromatogram obtained under optimized conditions for all investigated pesticides.

3. Results and discussion

3.1. SDME procedure optimization

The optimized parameters of the SDME/GC–MS were: extraction time – 30 min, solvent – toluene, drop volume – $1\text{ }\mu\text{L}$, stirring speed – 300 rpm [7]. Additionally, multivariate optimization was performed by a 2^2 full factorial design to identify the influence of salt addition (% NaCl) and pH on the SDME procedure. Two replicates were performed at the central point to estimate the experimental error [38,39], which were minimal in the six experiments. The dependent variable (response) was the sum of the analyte peak areas in each experiment. Table 1 shows the studied parameters and their sampling levels.

From the planning matrix built from the Pareto chart, shown in Fig. 2, it appears that the pH and salt effects are not significant at the 95% confidence level. Therefore, the pH was set at 5.0 (pH of the water) and no salt was added in the following steps (Section 2.3).

3.2. Validation

The parameters used for the validation of the SDME/GC–MS analytical method were linearity and linear range, precision, accuracy, limit of detection (LOD), limit of quantification (LOQ), (Table 2) and robustness (Table 3).

The linearity was studied by analysis of water samples extracted from the matrix using a pre-concentration of 10 mL ultra-pure water spiked with a standard solution of pesticides in the linear concentration ranges of $0.15\text{--}60\text{ }\mu\text{g L}^{-1}$ for dimethoate, $0.15\text{--}40\text{ }\mu\text{g L}^{-1}$ for methyl parathion, $0.19\text{--}30\text{ }\mu\text{g L}^{-1}$ for ethion and $1.1\text{--}60\text{ }\mu\text{g L}^{-1}$ for permethrin. The plots were linear for all compounds with r values higher than 0.90, which indicated good linearity. Similar results for r (0.9930–0.9994) were obtained in a study using SDME/GC–MS for the determination of organochlorine pesticides in vegetables [32]. The r values were better than those reported elsewhere [20,25], thus demonstrating the high linearity and sensitivity of the method. LOD and LOQ were established for the method by the analysis of ultra-pure water enriched with decreasing concentrations of the analyte and then extracted by SDME until the lowest detectable level (concentration equal to three times the signal/noise) and the lowest determinable level (concentration equal to ten times the signal/noise), respectively. The results for the limit of detection ranged from 0.05 to $0.38\text{ }\mu\text{g L}^{-1}$ and limit of quantification from 0.15 to $1.1\text{ }\mu\text{g L}^{-1}$ (Table 2). These values are typically a factor of 6–8 lower than those reported by Pinheiro and de Andrade (2009) [7] for organophosphate and pyrethroid pesticides in water samples collected from the irrigation project “Platô de Neópolis,” site located in Neópolis town, Sergipe (Brazil). Also the LOD ($10\text{--}75\text{ }\mu\text{g L}^{-1}$) reported by Palit et al. (2005) [9] in the determination of the chemical warfare agents and related compound in water samples using SDME/GC–MS are much higher than our study. When compared to other studies that used the same method in the determination of organochlorine pesticides in vegetables and chlorobenzene in water samples, it appears that the LOD values ($0.02\text{--}0.2\text{ }\mu\text{g L}^{-1}$) [32,40] are substantially similar to the results obtained in the present study. These comparisons show that the LOD and LOQ obtained from this study demonstrate higher sensitivity of the developed SDME/GC–MS method and therefore, a powerful tool for detection and quantification low concentrations of

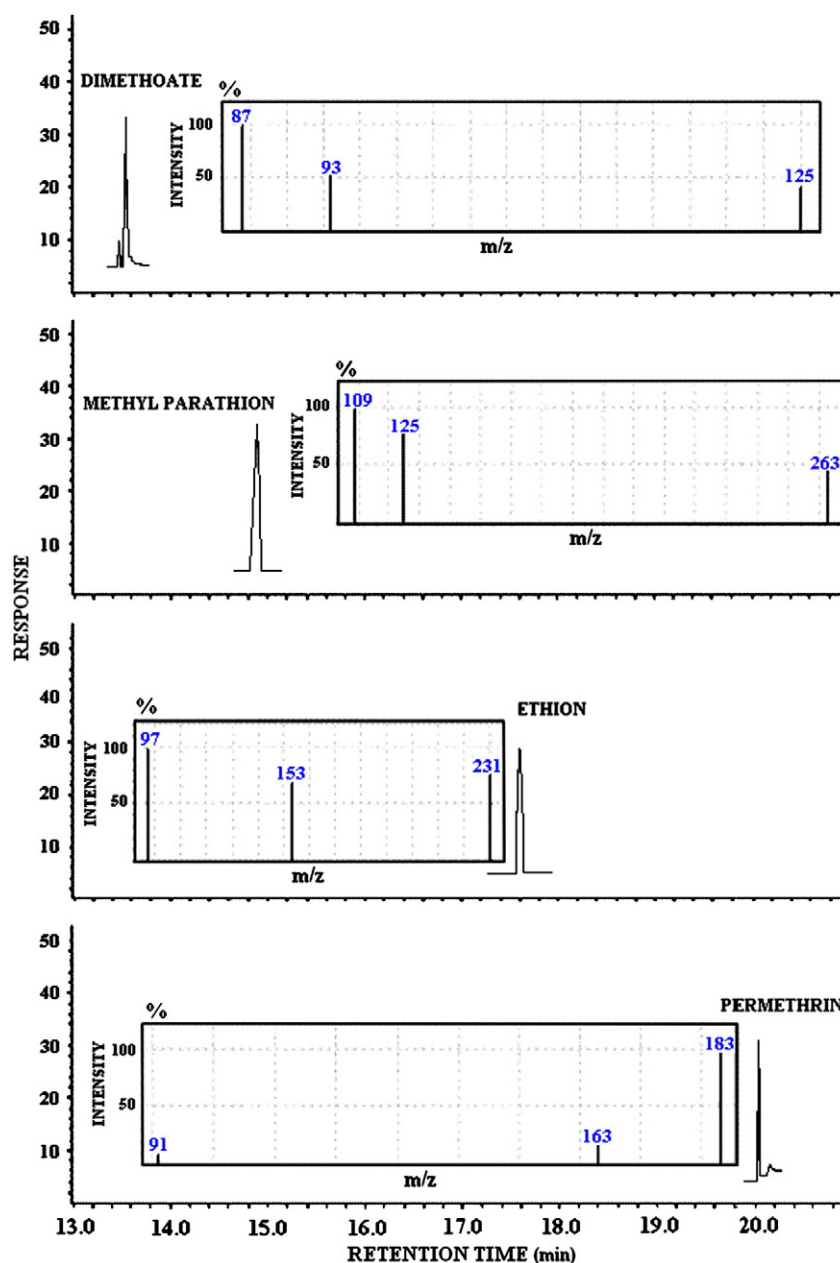


Fig. 1. Chromatograms of the standards used to identify peaks shown with the associated retention times and respective mass spectra (GC–MS).

pesticides in different samples such as water and vegetables samples [41].

Graphs were constructed with the relative response on the y axis and the corresponding concentrations in logarithmic scale on the x axis, as suggested Huber [42] to verify that all used points are within the corresponding linear dynamic range. These graphs (Figs. 3–6) showed that the method is linear within the response range of 95 to 105% and it is within the linear range for all pesticides studied. The points found to be outside the linear range of 95 to 105% were excluded from the analytical curve.

Table 1

Scores of sampling used in the 2^2 full factorial design.

Factors	Levels of sampling		
	–1	Center point	1
pH	1	3	5
Salt addition (% NaCl)	0	5	10

The precision of the method was evaluated using coefficient of variation (CV) from replicates ($n=9$), taken at three concentration levels (low, medium and high) as suggested by the ANVISA [43]. The deviation should not exceed 20%, in accordance with the methods of validation of pesticide residues and should be in a range similar to those obtained in other studies using SDME/GC–MS (4.8–11.4%) [44] and SDME/GC–FPD (7.9–13.6%) [23] for the determination of pesticides in water.

Recovery tests were performed by the enrichment of Milli-Q water and tap water (both 10 mL) with standard solution composed of $2.5 \mu\text{g L}^{-1}$ of methyl parathion, dimethoate and permethrin and $1.25 \mu\text{g L}^{-1}$ of ethion. Recoveries range from 76.2 to 107% that shows that the present method is accurate. The recoveries from this study are similar to other reported recovery levels (63.3–120%) [25,26,32].

In order to assess possible matrix effects, both tap water and river water (10 mL) were also enriched with standard solution ($2.5 \mu\text{g L}^{-1}$ – methyl parathion, dimethoate and permethrin, $1.25 \mu\text{g L}^{-1}$ – ethion). The relative recoveries of methyl parathion, dimethoate, ethion and

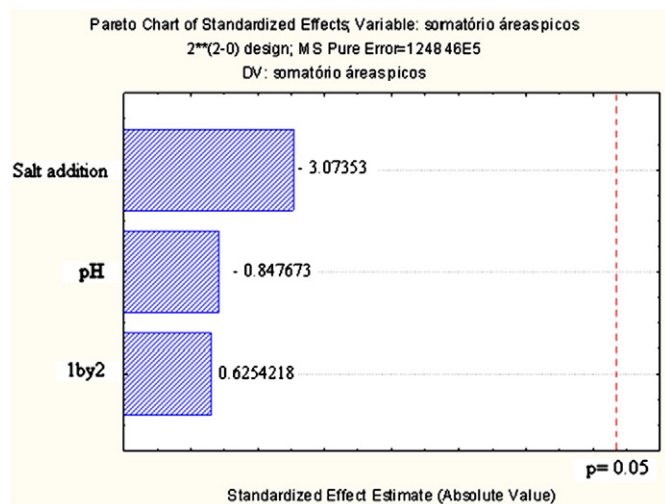


Fig. 2. Pareto chart.

Table 2
Parameters for validation of SDME/GC–MS analytical method.

Pesticide	Linear range ($\mu\text{g L}^{-1}$)	r^a (n^b)	R^c (%)	LOD ($\mu\text{g L}^{-1}$)	LOQ ($\mu\text{g L}^{-1}$)	CV ^d
Dimethoate	0.15–60	0.9969 (7)	78.6	0.05	0.15	10.2
Methyl parathion	0.15–40	0.9998 (7)	76.2	0.05	0.15	8.19
Ethion	0.1875–30	0.9950 (6)	106.8	0.0625	0.1875	7.39
Permethrin	1.125–60	0.9823 (6)	99.4	0.375	1.125	14.35

^a r = linear correlation coefficient.

^b n = replicates.

^c R = relative recovery.

^d CV = coefficient of variation ($n=9$).

Table 3
Factors used in the determination of robustness.

Parameters	Levels	
	Nominal	Variation
pH	5.0	1.0

permethrin from tap water were 76.2, 78.6, 107 and 99.4%, respectively, and from river water were 78.0, 82.2, 110 and 103%, respectively. The recovery results obtained show that the matrix has little effect on the extraction of pesticides by SDME.

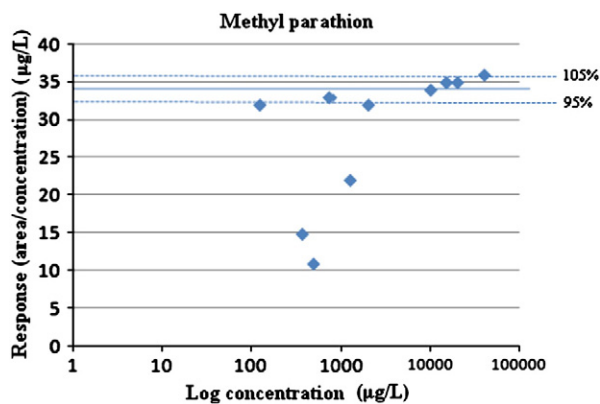


Fig. 3. Plot of response as a function of logarithmic concentration (Methyl parathion – GC–MS).

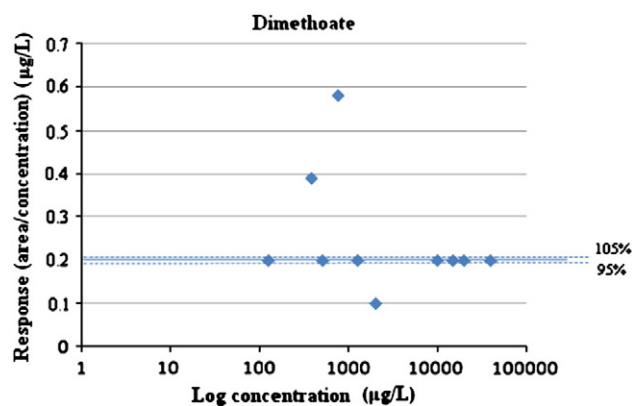


Fig. 4. Plot of response as a function of logarithmic concentration (Dimethoate – GC–MS).

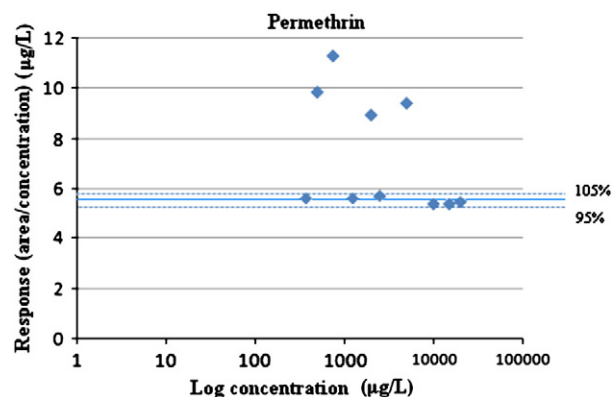


Fig. 5. Plot of response as a function of logarithmic concentration (Permethrin – GC–MS).

To check the robustness of the method the pH and sampling levels are shown in Table 3. For the pH=1 levels the responses were obtained in terms of total area of the peaks of the four pesticides and the CV was calculated (Table 4).

The experimental design showed that the pH is not significant at the 95% confidence level when varied from 5.0 to 1.0 (Fig. 2). The degree of result reproducibility (Table 4) demonstrates the robustness of the method.

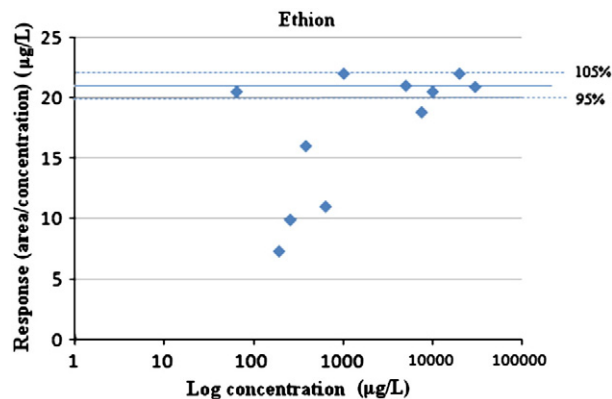


Fig. 6. Plot of response as a function of logarithmic concentration (Ethion – GC–MS).

Table 4

Comparison of CV (method and robustness).

Pesticide	Precision (CV) ^a	Precision (CV) ^b
Dimethoate	10.2	10.7
Methyl Parathion	8.19	8.94
Ethion	7.39	8.42
Permethrin	14.35	18.80

^a CV (method) with pH = 5 (water pH) (n = 9).^b CV (robustness) with pH = 1 (n = 3).**Table 5**

Water samples collected in Perímetro Irrigado Propriá.

Sample	Concentration ($\mu\text{g L}^{-1}$)			
	Dimethoate	Methyl Parathion	Ethion	Permethrin
1	2.12	0.083	0.12	1.47
2	2.32	0.054	<LOD	0.28
3	2.52	0.060	0.05	0.16
4	2.79	0.050	0.05	NF
5	2.99	0.052	0.048	NF
6	2.44	0.070	0.04	0.28
7	2.43	0.064	0.045	NF
8	2.65	0.077	0.05	0.59
9	2.30	0.040	0.035	NF

NF: not found; <LOD: concentration below LOD.

Table 6

Water samples collected in San Francisco River (water samples 1–5) and tap water samples collected in cities of Propriá (State of Sergipe – Brazil) and Aracaju (State of Sergipe – Brazil) (water samples 6–7).

Sample	Concentration ($\mu\text{g L}^{-1}$)			
	Dimethoate	Methyl Parathion	Ethion	Permethrin
1	NF	0.15	0.063	1.65
2	2.30	0.07	0.065	NF
3	2.38	0.1	0.069	NF
4	NF	0.075	0.078	NF
5	NF	0.072	0.075	1.13
6	2.08	0.20	<LOD	<LOD
7	1.51	0.15	0.048	NF

NF: not found; <LOD: concentration below LOD.

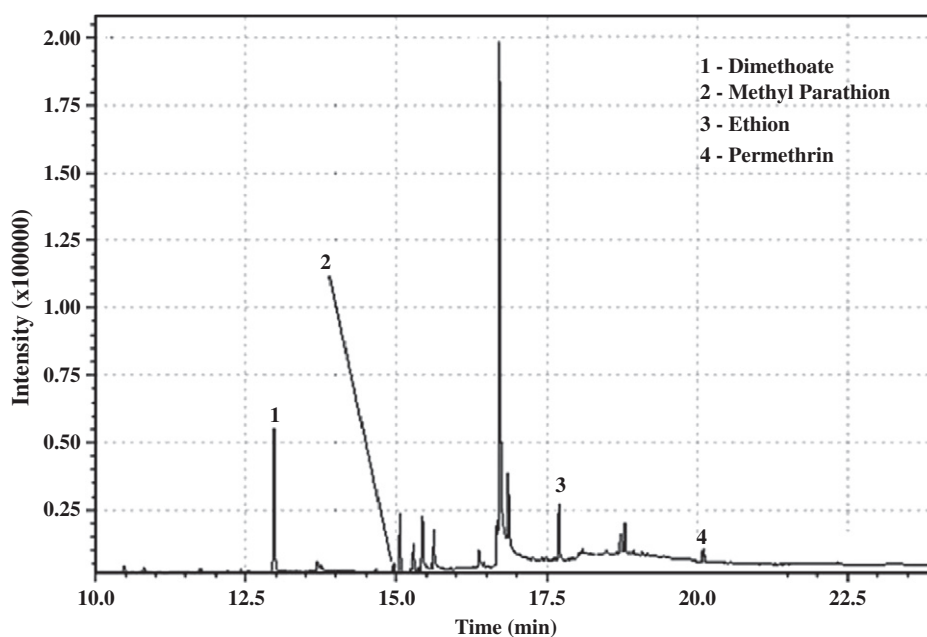
3.3. Application of SDME method

The developed SDME method was applied using field collected water samples and the data are presented in Tables 5 and 6. Fig. 7 shows the chromatogram obtained from the real samples in this study. The water sample used in testing this method were collected from an irrigation area in Propriá and river water from the banks of San Francisco River in Propriá city. The selected pesticides in this study are widely used in this rice cultivation field. Also tap water samples in two cities Propriá and Aracaju in Sergipe were collected.

The results for the analyzed pesticides in water samples collected from the irrigation field and from San Francisco River showed the presence of dimethoate, methyl parathion, permethrin and ethion. The data from this study were comparable to the pesticide legislations established by the European Union and the Brazilian laws. The pesticides concentrations in water samples from the two sites and also tap water from Propriá and Aracaju cities were below the maximum permissible levels by the Brazilian laws [45,46] for permethrin and methyl parathion. In contrast, the concentrations of all pesticides in water samples from the irrigation field and from San Francisco River exceeded the maximum permissible limit values set by European Union legislation for the individual pesticides except sample 4 (Table 6).

4. Conclusions

This study is a proposal of an alternative method for the determination of four pesticides (dimethoate, methyl parathion, ethion, and pyrethroid) in water samples by employing SDME extraction and GC–MS analysis. Our optimized method was validated by taking in consideration some analytical parameters, such as: linearity, linear range, precision, accuracy, limit of detection, limit of quantification, robustness. Also the validated methodology was utilized to determine pesticides in real samples (water samples from San Francisco River and from Perímetro Irrigado Propriá as well as tap water from Propriá and Aracaju). The present SDME/GC–MS method has demonstrated to be accurate, precise and reproducible with advantages of being rapid, simple, and to require smaller volumes of organic solvent.

**Fig. 7.** Chromatogram of a water sample (sample 1 – Perímetro Irrigado Propriá).

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References

- [1] A.A. D'Archivio, M. Fanelli, P. Mazzeo, F. Ruggieri, Comparison of different sorbents for multiresidue solid-phase extraction of 16 pesticides from groundwater coupled with high-performance liquid chromatography, *Talanta* 71 (2007) 25–30.
- [2] R. Romero-González, A.G. Frenich, J.L.M. Vidal, Multiresidue method for fast determination of pesticides in fruit juices by ultra performance liquid chromatography coupled to tandem mass spectrometry, *Talanta* 76 (2008) 211–225.
- [3] B. Gilbert-López, J.F. García-Reyes, A. Molina-Díaz, Sample treatment and determination of pesticide residues in fatty vegetable matrices: a review, *Talanta* 79 (2009) 109–128.
- [4] M.G.D. Silva, A. Aquino, H.S. Dórea, S. Navickiene, Simultaneous determination of eight pesticide residues in coconut using MSPD and GC/MS, *Talanta* 76 (2008) 680–684.
- [5] F.M. Rodrigues, P.R.R. Mesquita, L.S. Oliveira, F.S. Oliveira, A.M. Filho, P.A. de P. Pereira, J.B. de Andrade, Development of a headspace solid-phase microextraction/gas chromatography–mass spectrometry method for determination of organophosphorus pesticide residues in cow milk, *Microchem. J.* 98 (2011) 56–61.
- [6] E. Schreck, F. Geret, L. Gontier, M. Treilhou, Development and validation of a rapid multiresidue method for pesticide determination using gas chromatography–mass spectrometry: a realistic case in vineyard soils, *Talanta* 77 (2008) 298–303.
- [7] A.S. Pinheiro, J.B. de Andrade, Development, validation and application of a SDME/GC-FID methodology for the multiresidue determination of organophosphate and pyrethroid pesticides in water, *Talanta* 79 (2009) 1354–1359.
- [8] A. Menezes Filho, F.N. dos Santos, P.A.P. Pereira, Development, validation and application of a method based on DI-SPME and GC-MS for determination of pesticides of different chemical groups in surface and groundwater samples, *Microchem. J.* 96 (2010) 139–145.
- [9] M. Palit, D. Pardasani, A.K. Gupta, D.K. Dubey, Application of single drop microextraction for analysis of chemical warfare agents and related compounds in water by gas chromatography/mass spectrometry, *Anal. Chem.* 77 (2005) 711–717.
- [10] E. Psillakis, N. Kalogerakis, Developments in single-drop microextraction, *Trends Anal. Chem.* 21 (2002) 53–63.
- [11] D.A. Lambropoulou, T.A. Albanis, Application of solvent microextraction in a single drop for the determination of new antifouling agents in waters, *J. Chromatogr. A* 1049 (2004) 17–23.
- [12] L. Vidal, A. Chisvert, A. Canals, A. Salvador, Ionic liquid-based single-drop microextraction followed by liquid chromatography–ultraviolet spectrophotometry detection to determine typical UV filters in surface water samples, *Talanta* 81 (2010) 549–555.
- [13] J. López-Darias, M. Germán-Hernández, V. Pino, A.M. Afonso, Dispersive liquid-liquid microextraction versus single-drop microextraction for the determination of several endocrine-disrupting phenols from seawaters, *Talanta* 80 (2010) 1611–1618.
- [14] L. Zhao, H.K. Lee, Application of static liquid-phase microextraction to the analysis of organochlorine pesticides in water, *J. Chromatogr. A* 919 (2001) 381–388.
- [15] Y. He, H.K. Lee, Continuous flow microextraction combined with high-performance liquid chromatography for the analysis of pesticides in natural waters, *J. Chromatogr. A* 1122 (2006) 7–12.
- [16] L.S. Jager, A.R.J. Andrews, Solvent microextraction of chlorinated pesticides, *Chromatographia* 50 (1999) 733–738.
- [17] L.S. Jager, A.R.J. Andrews, Development of a rapid screening technique for organochlorine pesticides using solvent microextraction ((SME) and fast gas chromatography (GC)), *Analyst* 125 (2000) 1943–1948.
- [18] M.C. Lopez Blanco, S. Branco-Cid, B. Cancho-Grande, J. Simal-Gándara, Application of single-drop microextraction and comparison with solid-phase microextraction and solid-phase extraction for the determination of α - and β -endosulfan in water samples by gas chromatography–electron-capture detection, *J. Chromatogr. A* 984 (2003) 245–252.
- [19] C. López-Blanco, S. Gómez-Alvarez, M. Rey-Garrote, B. Cancho-Grande, J. Simal-Gándara, Determination of carbamates and organophosphorus pesticides by SDME-GC in natural water, *Anal. Bioanal. Chem.* 383 (2005) 557–561.
- [20] Q. Xiao, B. Hu, C. Yu, L. Xia, Z. Jiang, Optimization of a single-drop microextraction procedure for the determination of organophosphorus pesticides in water and fruit juice with gas chromatography–flame photometric detection, *Talanta* 69 (2006) 848–855.
- [21] P. Liang, L. Guo, Y. Liu, S. Liu, T. Zhang, Application of liquid-phase microextraction for the determination of phoxim in water samples by high performance liquid chromatography with diode array detector, *Microchem. J.* 80 (2005) 19–23.
- [22] H. Bagheri, F. Khalilian, Immersed solvent microextraction and gas chromatography–mass spectrometric detection of s-triazine herbicides in aquatic media, *Anal. Chim. Acta* 537 (2005) 81–87.
- [23] F. Ahmadi, Y. Assadi, S.M.R. Milani, M. Hosseini, M. Rezaee, Determination of organophosphorus pesticides in water samples by single drop microextraction and gas chromatography–flame photometric detector, *J. Chromatogr. A* 1101 (2006) 307–312.
- [24] L. Guo, P. Liang, T. Zhang, Y. Liu, S. Liu, Use of continuous-flow microextraction and liquid chromatography for determination of phoxim in water samples, *Chromatographia* 61 (2005) 523–526.
- [25] E. Zhao, L. Han, S. Jiang, Q. Wang, Z. Zhou, Application of a single-drop microextraction for the analysis of organophosphorus pesticides in juice, *J. Chromatogr. A* 1114 (2006) 269–273.
- [26] C. Ye, Q. Zhou, X. Wang, Improved single-drop microextraction for high sensitive analysis, *J. Chromatogr. A* 1139 (2007) 7–13.
- [27] M. Saraji, N. Esteki, Analysis of carbamate pesticides in water samples using single-drop microextraction and gas chromatography–mass spectrometry, *Anal. Bioanal. Chem.* 391 (2008) 1091–1100.
- [28] S. Pedersen-Bjergaard, K.E. Rasmussen, Bioanalysis of drugs by liquid-phase microextraction coupled to separation techniques, *J. Chromatogr. B: Anal. Technol. Biomed. Life Sci.* 817 (2005) 3–12.
- [29] J. Zhang, H.K. Lee, Application of liquid-phase microextraction and on-column derivatization combined with gas chromatography–mass spectrometry to the determination of carbamate pesticides, *J. Chromatogr. A* 1114 (2006) 269–273.
- [30] M.I. Pinto, G. Sontag, R.J. Bernardino, J.P. Noronha, Pesticides in water and the performance of the liquid-phase microextraction based techniques. A review, *Microchem. J.* 96 (2010) 225–237.
- [31] E. Psillakis, N. Kalogerakis, Solid-phase microextraction versus single-drop microextraction for the analysis of nitroaromatic explosives in water samples, *J. Chromatogr. A* 938 (2001) 113–120.
- [32] M. Zhang, J. Huang, C. Wei, B. Yu, X. Yang, X. Chen, Mixed liquids for single-drop microextraction of organochlorine pesticides in vegetables, *Talanta* 74 (2008) 599–604.
- [33] Y. Wu, L. Xia, R. Chen, B. Hu, Headspace single drop microextraction combined with HPLC for the determination of trace polycyclic aromatic hydrocarbons in environmental samples, *Talanta* 74 (2008) 470–477.
- [34] H.F. Maltes, D.L.G. Borges, E. Carasek, B. Welz, A.J. Curtius, Single drop microextraction with O, O-diethyl dithiophosphate for the determination of lead by electrothermal atomic absorption spectrometry, *Talanta* 74 (2008) 800–805.
- [35] J.L. Manzoori, M. Amjadi, J. Abulhassani, Ionic liquid-based single drop microextraction combined with electrothermal atomic absorption spectrometry for the determination of manganese in water samples, *Talanta* 77 (2009) 1539–1544.
- [36] D. Verma, S.K. Verma, M.K. Deb, Single-drop micro-extraction and diffuse reflectance Fourier transform infrared spectroscopic determination of chromium in biological fluids, *Talanta* 78 (2009) 270–277.
- [37] H. Kataoka, Recent developments and applications of microextraction techniques in drug analysis, *Anal. Bioanal. Chem.* 396 (2010) 339–364.
- [38] E.T. Sousa, F.M. Rodrigues, C.C. Martins, F.S. Oliveira, P.A. de P. Pereira, J.B. de Andrade, Multivariate optimization and HS-SPME/GC-MS analysis of VOCs in red, yellow and purple varieties of *Capsicum chinense* sp. Peppers, *Microchem. J.* 82 (2006) 142–149.
- [39] S.L.C. Ferreira, R.E. Bruns, E.G.P. Da Silva, W.N.L. Dos Santos, C.M. Quintella, J.M. David, J.B. de Andrade, M.C. Breitenkreitz, I.C.S.F. Jardim, B.B. Neto, Statistical designs and response surface techniques for the optimization of chromatographic systems, *J. Chromatogr. A* 1158 (2007) 2–14.
- [40] Y. Wang, Y.C. Kwok, Y. He, H.K. Lee, Application of dynamic liquid-phase microextraction to the analysis of chlorobenzenes in water by using a conventional microsyringe, *Anal. Chem.* 70 (1998) 4610.
- [41] F.G. Kitson, B.S. Larsen, C.N. McEwen, *Gas Chromatography and Mass Spectrometry: A Practical Guide*, Academic Press, San Diego, 1996.
- [42] L. Huber, Validation of analytical methods: review and strategy, *LC-GC Int.* (1998) 96–105.
- [43] Anvisa, Guia para Validação de Métodos Analíticos, Agência Nacional de Vigilância Sanitária, Resolução RE nº 475 de 19 de Março de 2002, Brazil Available on: http://www.anvisa.gov.br/hotsite/genericos/legis/resolucoes/2002/475_02re.htm 2002 [accessed: 15/02/2011].
- [44] M. Saraji, M. Bakhshi, Determination of phenols in water samples by single-drop microextraction followed by in-syringe derivatization and gas chromatography–mass spectrometric detection, *J. Chromatogr. A* 1098 (2005) 30–36.
- [45] Brasil, Controle e vigilância da qualidade da água para consumo humano, Portaria 518, de 25 de março de 2004, Brazil Available on: http://www.agrolab.com.br/portaria%20518_04.pdf 2004 [accessed: 15/08/2010].
- [46] Brasil, Classificação das águas doces, salobras e salinas do Território Nacional, Resolução CONAMA nº 357, de 18 de Junho de 2005, Brazil Available on: <http://www.mma.gov.br/port/conama/res/res86/res2086.html> 2005 [accessed: 15/08/2010].