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Odour-causing organic compounds in wastewater treatment plants: Evaluation of headspace solid-phase microextraction as a concentration technique

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ABSTRACT

Odorous emissions from wastewater collection systems and treatment facilities affecting quality of life have given local populations reasons to complain for decades. In order to characterise the composition of such malodorous emissions, a method based on headspace solid-phase microextraction (HS-SPME) and gas chromatography coupled to mass spectrometry (GC–MS) has been developed to determine a list of compounds belonging to different chemical families, which have been previously described as potentially responsible for odour complaints, in wastewater matrices. Some parameters affecting the chromatographic behaviour of the target compounds were studied (e.g. splitless time). Experimental conditions affecting the extraction process (temperature, time and salt content) were evaluated by applying a factorial design at two levels. Using a DVB/CAR/PDMS fibre and the optimised HS-SPME conditions, calibration curves were constructed with detection limits in the range of 0.003–0.6 $\mu g\,L^{-1}$. Recovery values higher than 70% and relative standard deviation values between 5 and 16% ($n=5$) were obtained for all compounds and found to be satisfactory. In wastewater samples, a decrease in the concentration of the analysed compounds through the different treatments was observed. Most of the target analytes were found in influent samples while only octanal and carvone were detected in samples from the plant effluent.

1. Introduction

Odorous emissions from wastewater collection systems and treatment facilities represent a problem that has affected citizens for decades [1,2]. Odour emissions affect quality of life, leading to psychological stress and symptoms such as insomnia, loss of appetite and irrational behaviour [3]. As a consequence of the poor public image of wastewater treatment plants (WWTPs), public concern and complaints have been increasing in recent years.

The composition of sewer gases is complex. Many of the emitted inorganic and organic gases and vapours come from anaerobic decomposition of organic matter containing sulphur and nitrogen. Thus, H_2S, NH_3, CO_2, and CH_4 are present at high concentrations, and the first two are powerfully malodorous [2]. Moreover, other highly malodorous compounds, such as mercaptans, organic sulphides, nitrogen-containing compounds (e.g. amines, indole and skatole), and oxygenated compounds (e.g. aldehydes, alcohols, organic acids and ketones) might also be present [1,2,4]. Concentrations of these key odorous compounds are often very low, reaching no more than a few $\mu g\,L^{-1}$ or $mg\,L^{-1}$.

Some of the compounds related with WWTP odours, in particular those present at higher concentrations can be determined directly without a concentration step. H_2S portable instruments have been designed for in situ determination [2,3,5]. Ammonia is often determined by specific methods, such as colorimetry and titrimetry [6]. Ion-selective electrodes have also been used for this purpose [6,7]. Primary and secondary amines are usually analysed by means of reversed-phase liquid chromatography with UV detection [6]. But due to the complex nature of most odours, it is difficult to identify the odorants present in air and wastewater without first using a separation technique. Gas chromatography with flame ionisation detection (GC-FID) and gas chromatography coupled to mass spectrometry (GC–MS) are frequently used to identify and quantify other components of gaseous mixtures [3]. Additionally, in order to ascertain the contribution of the detected compounds in the odour perception, a parallel olfactometry analysis is carried out [1–3,8]. However, in many cases these techniques are not sensitive enough and it is necessary to concentrate the sample prior to the analysis [3].

Solid sorbent capture followed by GC determination is commonly the technique of choice when volatile organic compounds (VOCs) are investigated in air samples [9–11]. Traps with more than one sorbent material are used to facilitate quantitative retention and desorption of VOCs over a wide range of compounds.

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Dincer et al. [2] collected samples from the headspace of tanks located in WWTP units and sludge management areas with a multi-bed trap packed with Tenax TA and Carboxen 1000. They identified 29 compounds belonging to four different types of chemicals (sulphur-containing compounds, aldehydes, monoaromatics and halogenated compounds). A method for the determination of volatile organic sulphur compounds (SVOCs) in air from sewage management plants in Tarragona and Reus (Spain) has also been developed [12]. A trap of Tenax TA and Unicarb was used and seven SVOCs (ethyl mercaptan, dimethyl sulphide, carbon disulphide, propyl mercaptan, butyl mercaptan, dimethyl disulphide and 1-pentanethiol) were detected and quantified.

The presence of odour compounds can be investigated directly in water and wastewater samples. In such cases, purge and trap and closed-loop stripping methods have been applied to concentrate VOCs [3,13,14]. Since the introduction by Pawliszyn and his research group of solid-phase microextraction (SPME) as a sample preparation technique, it has become an accepted method for the determination of volatile and semi-volatile substances. SPME offers some advantages compared to more traditional methods of extraction: it is a solvent-free, simple, inexpensive and efficient procedure [15]. Sampling, extraction and enrichment are accomplished in a single step, since the target analytes are transferred from the sample to the exposed fibre, and desorption is performed directly in the injector port of the GC instrument. As a result of these remarkable characteristics of SPME, most authors have chosen this technique for the analysis of odorous compounds in wastewater and air samples. Kleeberg et al. [8] analysed waste gas from a fat refinery using SPME. The fibre was exposed to the sample, collected in a sampling bag at ambient temperature and a total of 56 substances including aldehydes, terpenes and esters were identified. A procedure based on the application of Carboxen/polydimethylsiloxane (CAR/PDMS) fibre for the extraction and concentration of a group of seven SVOCs (ethyl mercaptans, dimethyl sulphide, carbon disulphide, propyl mercaptans, butyl mercaptans, dimethyl disulphide, and 1-pentanethiol) in air samples from a sewage treatment plant has also been developed [16]. In this case, target analytes were extracted in glass bulbs used for field sampling of air. Pan et al. [17] determined amines in air and water using derivatisation combined with SPME, and NPTFA (*p*-nitrophenyl trifluoroacetate) and PFBAY (2,3,4,5,6-pentafluorobenzylaldehyde) as derivatising reagents. As for aqueous samples, Tsai et al. [18] applied a method based on HS-SPME using on-fibre derivatisation with PFBHA (*O*-2,3,4,5,6-(pentafluorobenzyl)hydroxylamine hydrochloride) for the analysis of aldehydes in water. Ábalos et al. [19] developed a method based on HS-SPME for the determination of volatile sulphides and disulphides in wastewaters. Huang et al. [20] analysed amines in wastewater samples by means of HS-SPME technique using a PDMS fibre. Furthermore, an analytical procedure to determine free volatile fatty acids in wastewater samples has also been reported [21].

Most of the published works using HS-SPME as an extraction technique for VOCs in aqueous matrices determine groups of compounds belonging to the same chemical family (e.g. aldehydes, sulphides and mercaptans, amines, and volatile fatty acids). In this paper we describe a method we have developed based on HS-SPME and using GC–MS for the characterisation of a list of compounds belonging to different chemical families in wastewater matrices. We considered several variables affecting the chromatographic behaviour of the target compounds (e.g. splitless time) and investigated experimental conditions affecting their extraction using HS-SPME (e.g. type of sorbent, time and extraction temperature) according to the design of experiments (DoE) methodology. Finally, we applied the developed method in the analysis of aqueous samples from a wastewater treatment plant.

2. Experimental

2.1. Chemicals

Dimethyl disulphide (DMDS, 99%), octanal (99%), (R)-(+)-limonene (99%), *m*-cresol (99.7%), nonanal (95%), (−)-carvone (99%), butyric acid (99.5%), indole (99%), and skatole (98%) were obtained from Sigma–Aldrich (Steinheim, Germany). Phenol (99.5%) was obtained from Dr. Ehrenstorfer (Augsburg, Germany). Sodium chloride (99.9%) and HPLC-gradient grade methanol were from Carlo-Erba Reagents (Milan, Italy). Milli-Q water from a Milli-Q Plus water purification system (Millipore, Bedford, MA, USA) was used.

We prepared stock standard solutions by weight in methanol and stored them at 4 °C for up to a week. Working solutions were made daily by diluting the standard solutions to the required concentration with Milli-Q water.

We obtained influent, secondary treatment and effluent water samples from a WWTP located in Castell-Platja d'Aro (Girona, Spain), and stored them in glass bottles at −16 °C. Some of these samples were used for validation purposes as indicated in Section 3.3.

2.2. Headspace solid-phase microextraction (HS-SPME) procedure

SPME experiments were performed with a manual fibre holder. We tested two different commercially available fibre coatings: a 75 μm CAR/PDMS and a 50/30 μm divinylbenzene/Carboxen/polydimethylsiloxane (DVB/CAR/PDMS). The fibre holder and coatings were supplied by Supelco (Bellefonte, PA, USA). Before use, we conditioned each fibre according to the manufacturer's instructions to remove contaminants and stabilise the solid phase.

We introduced a sample solution (5 mL) into a 15 mL screw-cap glass vial, added NaCl, closed the vial and put it over a magnetic stirrer (Variomag®, Germany) in a water-thermostated bath. Magnetic stirring (medium speed) was applied during the extraction using a PTFE-coated stir bar and the fibre was exposed to the headspace above the aqueous solution. The final extraction conditions were: 1 g of NaCl added, extraction time 30 min, and extraction temperature 70 °C. After completion of sampling, we pulled the fibre into the needle and removed the SPME device from the vial and inserted it into the injection port of the GC for thermal desorption and analysis. After each chromatographic run we reinserted the fibre into the injection port of the GC during 15 min to ensure that no compounds remained in the coating.

2.3. Experimental design

A full factorial design was performed to evaluate the influence of the parameters on the extraction of odorous compounds from an aqueous solution. This allowed us to determine the influence of all the experimental variables studied and also to ascertain the interactions between them.

For each analyte, we considered three variable factors that can affect the extraction yield: ionic strength quantified as NaCl concentration (c), temperature (T) and extraction time (t). Then we selected a 2^3 full factorial design. Table 1 shows the experimental range for each factor. The central point (0.5 g, 50 °C, 20 min) was also measured and considered as an experiment.

Table 1
Factor levels considered in the experimental design optimisation.

Variable	Low level (−)	Medium level (0)	High level (+)
c (g)	0	0.5	1
T (°C)	30	50	70
t (min)	10	20	30

Table 2
Odour threshold concentrations (OTC), retention times and *m*/*z* ratios of the target compounds. Values in bold are the quantifier ions. n.a.: not available.

Compound	OTC[a] (μg L^{-1})	Retention time (min)	*m*/*z*
DMDS	0.3, 1.0	5.21	45, 79, **94**
Phenol	n.a.	18.81	66, **94**
Octanal	0.7, 1.4	19.44	69, 84, **95**
Limonene	200, 1000	20.33	68, **93**
m-Cresol	800	22.19	79, **107**, **108**
Nonanal	1, 2.5	23.09	81, 98, **143**
Carvone	10	27.42	82, 108, **151**
Indole	370	28.82	90, **117**
Skatole	1.2	31.34	130, **131**

[a] Compendium data from [6,20,23].

We carried out all the experiments in triplicate and in random order. The Minitab v14 computer program was used for data manipulation and calculations [22].

2.4. Equipment and chromatographic conditions

We performed gas chromatographic analysis with a Trace GC 2000 coupled to a PolarisQ ion trap mass spectrometer detector (Thermo Scientific, Waltham, MA, USA). Analytes were separated with a TRB-5 MS capillary column (Teknokroma, Spain) (30 m × 0.25 mm i.d.; 0.25 μm film thickness). The split/splitless injection port was equipped with a 0.75 mm ID SPME liner and operated at 250 °C. The carrier gas was helium at a constant inlet flow rate of 1 mL min^{-1}.

The oven temperature program was: initial temperature 35 °C, held for 10 min; then increasing by 5 °C/min up to 150 °C and by 15 °C/min up to 250 °C, and held for 2 min; total run 42 min. We conducted MS analyses in full-scan mode and monitored masses between 40 and 300 amu. Ionisation was carried out in the electron impact (EI) mode at 70 eV. We maintained the transfer line temperature at 280 °C and the ion source temperature at 225 °C. The acquisition of chromatographic data was performed using Xcalibur 1.4 software (Thermo Scientific). Table 2 shows the list of the target compounds, their respective odour threshold concentrations and details of the GC–MS analysis.

3. Results and discussion

In this study, we selected a list of odorous compounds belonging to different chemical families for determination in wastewaters by HS-SPME (Table 2); we included phenolic compounds, aldehydes, sulphur-containing compounds, nitrogen-containing compounds and terpenes. All of them had previously been reported as present in wastewaters and in the atmosphere [2,3,13,16,19,24]. Although H_2S, ammonia and amines are some of the most important contributors to the malodorous emissions from WWTPs, we discarded them after considering the specific chromatographic conditions required for their analysis.

We performed preliminary experiments to assay the possibility of adding volatile fatty acids to the list of target compounds. On-fibre silylation with *N*-(*tert*-butyldimethylsilyl)-*N*-methyl-trifluoroacetamide (MTBSTFA) was required to analyse these compounds [25]. We observed losses of other target analytes during the derivatisation step. For this reason, we did not include volatile fatty acids in the study.

3.1. Selecting fibre coatings and splitless time

Due to the different volatility of molecules studied, two fibre coatings – CAR/PDMS and DVB/CAR/PDMS – were selected for evaluation. CAR/PDMS fibre has previously been used to characterise odorous waste gas emissions [8] and to determine volatile alkyl sulphides [19] and BTX [26] in wastewaters. High efficiency is usually obtained with this fibre coating for small polar analytes that can be rapidly desorbed at temperatures around 270–280 °C. On the other hand, Larreta et al. have observed that DVB/CAR/PDMS fibre showed the best extraction/desorption yields for the determination of phenols and indoles in cow slurry [27]. DVB-based coatings have also been used for the analysis of a large variety of taste and odour compounds in water samples [28,29].

In this paper we have observed a clear difference between the two coatings in terms of peak shape. As can be seen in Fig. 1, for some selected analytes CAR/PDMS gave increased peak tailing especially in the case of limonene and *m*-cresol. This can be attributed to the presence of carbon in the coating composition causing a strong interaction with polar compounds that are not easily released from the fibre. Peak shape is improved when using DVB/CAR/PDMS coating and for this reason it was selected for further experiments.

In SPME, splitless injection using narrow-bore glass liners is required to produce a high linear flow rate of the carrier gas around the fibre and facilitate the rapid removal of desorbed analytes from the injector [15]. Selecting the most appropriate splitless conditions, good chromatographic peak shape and widths can be obtained as long as the GC oven temperature is held at a minimum of 50 °C below the boiling point of the most volatile compounds

Table 3
Statistical results for the experimental design. Significance *p*-values are given for main effects, double and triple interactions and for curvature evidence. Most relevant single and double variable terms effects are also shown in decreasing order of importance.

Analyte	Single variable effects		Double variable effects		Triple variable effects	*p*-Value for curvature evidence
	p-Value	Significant terms	*p*-Value	Significant terms	*p*-Value	
DMDS	0.000	−*T* +*c* +*t*	0.001	−*Tc*	0.043	0.496
Phenol	0.000	+*T* +*c* +*t*	0.000		0.009	0.226
Octanal	0.000	+*t* +*T* +*c*	0.265		0.008	0.019
Limonene	0.453		0.931		0.100	0.470
m-cresol	0.000	+*T* +*c* +*t*	0.000	+*c* −*tT*	0.000	0.005
Nonanal	0.000	+*t* +*T*	0.011		0.057	0.063
Carvone	0.000	+*T* +*c*	0.497		0.419	0.989
Indole	0.000	+*T* +*c* +*t*	0.000		0.000	0.083
Skatole	0.000	+*T* +*c* +*t*	0.000		0.015	0.070

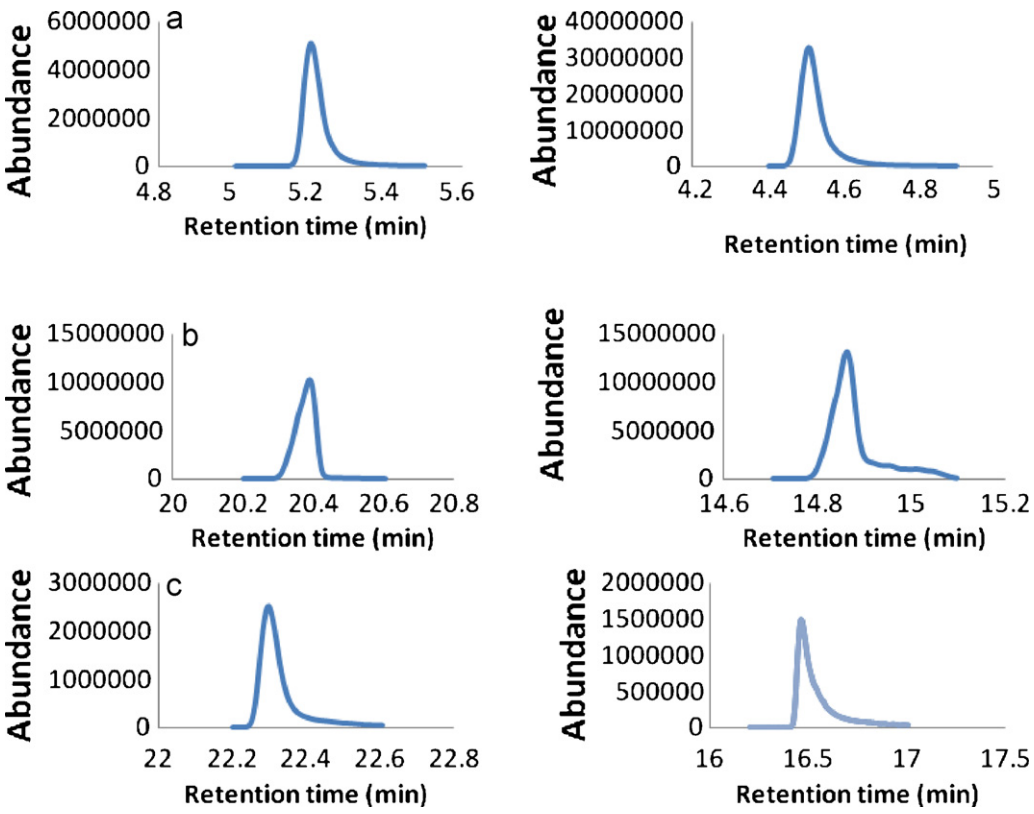

Fig. 1. Chromatographic peaks for some selected compounds (0.1 μg L^{-1} of each compound) obtained with the two fibre coatings: on the left, with DVB/CAR/PDMS fibre; on the right, with CAR/PDMS fibre. Extraction conditions: 30 min at 50 °C and 1.2 g of NaCl added to the sample: (a) DMDS (m/z = 94), (b) limonene (m/z = 93), (c) *m*-cresol (m/z = 107, 108).

Table 4
Quality parameters obtained in standard solutions analysis. Standard deviations are showed in parenthesis.

Compound	Working range (μg L^{-1})	a (S_a) (×10^5)	b (S_b) (×10^5)	r^2	LOD (μg L^{-1})	LOQ (μg L^{-1})
DMDS	0.1–100	4.7 (7.2)	1.8 (0.2)	0.9719	0.03	0.10
Phenol	1.4–250	2.4 (2.7)	0.5 (0.2)	0.9939	0.4	1.4
Octanal	1.9–15	0.2 (2.7)	0.61 (0.03)	0.9958	0.6	1.9
Limonene	1.1–10	3.7 (4.7)	8 (1)	0.9853	0.3	1.1
m-cresol	0.5–150	8.6 (7.2)	1.92 (0.09)	0.9940	0.2	0.5
Nonanal	1.9–10	3.4 (1.6)	5.0 (0.3)	0.9913	0.6	1.9
Carvone	0.1–10	2.9 (3.9)	6.3 (0.6)	0.9723	0.03	0.10
Indole	0.7–225	1.6 (3.9)	0.74 (0.04)	0.9926	0.2	0.7
Skatole	0.2–20	7.9 (9.9)	10 (1)	0.9780	0.06	0.20

a = intercept.
S_a = standard deviation of the intercept.
b = slope.
S_b = standard deviation of the slope.
r^2 = determination coefficient.
LOD = limit of detection.
LOQ = limit of quantitation.

Table 5
Concentrations, recoveries and intra-day precision values (n = 5) obtained in spiked Milli-Q water solution and real sample analysis. Standard deviations are shown in parenthesis.

Compound	Concentration (μg L^{-1})	Recovery (%)		Intra-day precision (% RSD)	
		Spiked Milli-Q water	Influent wastewater samples	Spiked Milli-Q water	Influent wastewater samples
DMDS	50	72 (4)	86 (3)	5	14
Phenol	150	79 (9)	96 (4)	12	9
Octanal	5	79 (6)	49 (7)	6	15
Limonene	7.5	75 (8)	82 (1)	10	20
m-Cresol	100	84 (9)	92 (15)	12	7
Nonanal	5	90 (10)	96 (2)	10	13
Carvone	7.5	90 (4)	94 (8)	5	11
Indole	90	90 (15)	73 (20)	16	18
Skatole	10	120 (20)	72 (30)	16	15

Table 6
Results obtained in WWTP samples analysis. Concentrations in $\mu g\,L^{-1}$. Standard deviations are showed in parenthesis. n.d.: not detected, n.q.: not quantified ($n = 3$).

Compound	Influent			Biologic treatment effluent			Plant effluent (after UV treatment)		
	Day 1	Day 2	Day 3	Day 1	Day 2	Day 3	Day 1	Day 2	Day 3
DMDS	5 (1)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Phenol	38 (5)	27 (2)	39.3 (0.8)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Octanal	n.d.	n.q	n.q.	n.q.	n.d.	n.q.	n.q.	n.d.	n.d.
Limonene	1.14 (0.09)	n.q.	1.28 (0.09)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
m-Cresol	80 (10)	100 (15)	151 (7)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Nonanal	n.d.	n.q.	n.q.	n.d.	n.d.	n.d.	n.q.	n.d.	n.d.
Carvone	0.70 (0.04)	1.00 (0.08)	1.26 (0.06)	n.d.	0.500 (0.007)	0.516 (0.002)	n.d.	0.520 (0.003)	0.50 (0.01)
Indole	90 (7)	47 (8)	66 (5)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
Skatole	10 (1)	10 (2)	13.5 (0.7)	n.d.	0.90 (0.06)	n.d.	n.d.	n.d.	n.d.

when 0.25 μm film thickness columns are used [30]. In the case of very volatile compounds, short desorption times (less than 1 min) are expected to be sufficient for the quantitative transfer of the extracted analytes [26]. On the contrary, splitless times from 1 to 5 min are usual for semi-volatiles. In this study splitless times of 30 s, 1 and 2 min were considered with DVB/CAR/PDMS fibre and for each analyte we evaluated several factors, for example peak shape, peak area and carryover. When desorption was performed for only 30 s, the peak areas values obtained were 50% lower than those obtained when desorption was performed during 1 min. Moreover, 1 and 2 min gave statistically comparable results without affecting the peak shape. The only exception were carvone and nonanal, which resulted in higher peak area values when 2 min of splitless time was considered. We evaluated the possible carryover for these two compounds at 1 min splitless time by acquiring a new chromatogram after the analysis of a sample. No peaks corresponding to these analytes were identified at the corresponding retention times. These findings let us select 1 min as the most appropriate desorption time for all the analytes.

3.2. Study of the sampling conditions

We defined an experimental domain to ascertain the influence of temperature, time of extraction and salt content on the extraction of odorous compounds from aqueous solutions (Table 1). We carried out a full two-level factorial design to check for the presence of double interactions and evidence of curvature effects that could not be detected using a classic procedure based on the evaluation of each variable individually. We analysed absolute peak areas and the results obtained are summarised in Table 3, where the significances (p-values) are given. The sign beside each variable name indicates the optimal level to maximise the response. Results showed that for all compounds no statistically relevant interactions occurred between the variables evaluated (the corresponding p-values for single interactions are much smaller than those for double and triple interactions). Additionally, there were no statistically relevant effects for limonene.

As can be seen in Table 3, temperature was a crucial variable as it had a noticeable influence on six analytes (DMDS, phenol, *m*-cresol, carvone, indole, and skatole) and the response was maximised when temperature was set at the highest level. Extraction yields can be enhanced when an optimum temperature is applied during sampling. In general, the amount of extracted analyte increased at higher temperatures that facilitate the transport of the analytes from the solution to the headspace phase. In the case of the most volatile target compound (DMDS), the extraction yield was not enhanced when the temperature was set at the highest level due to competition with the thermal desorption process. Thus, low temperatures might be used to avoid losses of this analyte. Taking into account the response for all compounds, we set the sampling temperature at 70 °C.

Extraction times with SPME usually vary from a few minutes to an hour or more, depending on the matrix, analytes, fibre phase and the desired sensitivity. In the case of sulphur-containing compounds, it has been found that small extraction times are required to reach equilibrium (less than 15 min) [31,32]. On the contrary, for semi-volatile compounds longer extraction times are necessary, even longer than 60 min [15,33]. Due to the range in volatility of the substances evaluated in this work, extraction times between 10 and 30 min were evaluated to find the best conditions for the majority of the target analytes. Extraction times longer than 30 min were not considered to avoid extending the total analysis time for each sample. As can be seen in Table 3, extraction time had a clear influence on octanal and nonanal extraction, and must be kept at the highest level. For this reason a extraction time of 30 min was selected.

When studying the NaCl content, it is expected as a general trend that increasing the ionic strength of the sample makes organic substances less soluble, increasing the partition coefficients [15]. This effect depends on the polarity of the analyte, the concentration of salt and the sample matrix. For the compounds evaluated in this study, the addition of salt enhanced the extraction. Therefore, sampling was carried out at the highest salt level (1 g NaCl). These main conclusions are better visualised in Pareto graphs (see supplementary materials).

3.3. Quality parameters

We tested the linearity of the HS-SPME method in the ranges shown in Table 4. Each concentration level was analysed in triplicate. For all compounds, residual plots confirmed linearity in the range evaluated, with a determination coefficient (r^2) greater than 0.97. We analysed samples ($n = 7$) at reduced concentrations to experimentally determine the limits of detection (LOD) and the limits of quantification (LOQ), and took the calculated standard deviation for each compound as the standard deviation of the blank. IUPAC 3σ and 10σ criteria were used to determine LODs and LOQs, respectively, which are summarised in Table 4. As can be observed, the developed method allows the quantification of odorous substances present in water samples well below their odour threshold concentration. Furthermore, LODs and LOQs were also evaluated using spiked samples prepared using water from the secondary treatment unit. No effect from the matrix was observed and equivalent limits were obtained.

Recoveries and intra-day precision ($n = 5$) of the method were evaluated at the concentration levels indicated in Table 5. We used spiked samples (Milli-Q water as well as water samples obtained at the influent of the WWTP) prepared just before analysis to evaluate these parameters. Concentrations of those compounds initially present were subtracted from the spiked values. We obtained recoveries ranging from 72 to 120% (Milli-Q water) and from 72 to 96% (WWTP water) for all compounds. Only recovery for octanal was lower which can be attributed to a rapid degradation of this

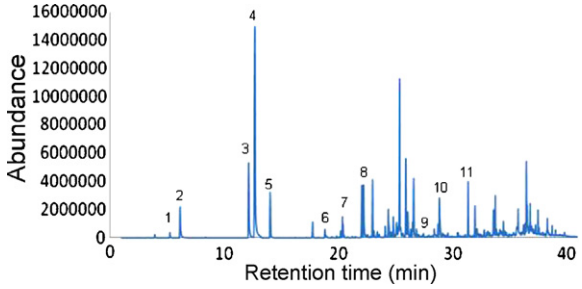

Fig. 2. Extracted chromatograms of a sample taken at the influent of the WWTP (day 1) using optimised experimental conditions: (1) DMDS, (2) toluene, (3) ethyl benzene, (4) *p*-xylene, (5) *o*-xylene, (6) phenol, (7) limonene, (8) *m*-cresol, (9) carvone, (10) indole, (11) skatole.

compound in the influent WWTP sample, probably due to microbial activity. The values in Table 5 are in agreement with the "single laboratory validation guidelines" of AOAC [34], which set an acceptable recovery range of between 70 and 120% at these concentration levels.

3.4. Analysis of wastewater samples

The proposed method was applied to the analysis of samples obtained from a WWTP in Castell-Platja d'Aro (Girona, Spain). We obtained samples from the influent, the biologic treatment effluent and the plant effluent (after UV treatment). Fig. 2 illustrates the extracted chromatograms of a sample taken at the influent of the WWTP (day 1). The method also allowed the semi-quantitative determination of benzene, toluene, ethylbenzene and xylenes which were also present in this sample.

The results, summarised in Table 6, show a decrease in the concentration of the target compounds along the different treatments. All compounds were usually detected in influent samples, and *m*-cresol, indole, phenol, and skatole were present at higher concentrations. Octanal was detected (but not quantified) in 55% of the wastewater samples analysed, which indicates that this compound was present at concentrations above its odour threshold value. Skatole and DMDS gave concentrations above their respective odour threshold values (Table 2) only in influent samples. Moreover, carvone was determined in samples from the plant effluent.

Our results are in agreement with those published in other papers. Islam et al. [6] detected DMDS in samples from the individual package treatment at concentrations between 0.08 and 7.49 $\mu g\,L^{-1}$. Additionally, they detected indole and skatole in samples from the sludge treatment process. Indole was found at concentrations between 6 and 61.8 $\mu g\,L^{-1}$ and skatole was found at 4.83 $\mu g\,L^{-1}$. Hwang et al. [1] detected DMDS in influent samples at concentrations between 3 and 27 $\mu g\,L^{-1}$ and indole at 570 $\mu g\,L^{-1}$. However, they also detected DMDS in samples from the plant effluent. Octanal was detected in snow samples by Sieg et al. [35] at concentrations between 0.324 and 0.594 $\mu g\,L^{-1}$.

4. Conclusions

We have developed and successfully applied an HS-SPME method followed by GC–MS to analyse odorous volatiles from aqueous samples from wastewater treatment plants. We have optimised the method for a list of compounds belonging to different chemical families, including volatile sulphides, aldehydes, phenols, indole, skatole and some terpenes. DVB/CAR/PDMS coating showed better performance in the microextraction process and experimental conditions were fixed as: 1 g of NaCl added, extraction time 30 min, and extraction temperature 70 °C. The optimised method was validated using spiked Milli-Q water and real water samples: good detection limits (between 0.03 and 0.6 $\mu g\,L^{-1}$) as well as good intra-day precision values (RSD ranging from 72 to 120%, $n = 5$) were found. From the analysis of water samples from WWTPs, the presence of almost all the target compounds was found. Some of these compounds appeared in concentrations above their odour threshold value.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.chroma.2011.02.017.

References

[1] Y. Hwang, T. Matsuo, K. Hanaki, N. Suzuki, Water Res. 29 (1995) 711.
[2] F. Dincer, A. Muezzinoglu, Environ. Sci. Health, Part A 43 (2008) 1569.
[3] P. Gostelow, S.A. Parsons, R.M. Stuetz, Water Res. 35 (2001) 579.
[4] C. Hort, S. Gracy, V. Platel, L. Moynault, Chem. Eng. J. 152 (2009) 44.
[5] P. Gostelow, S.A. Parsons, Wat. Sci. Technol. 41 (2000) 33.
[6] A.K.M.N. Islam, K. Hanaki, T. Matsuo, Wat. Sci. Technol. 38 (1998) 337.
[7] J. Kangas, A. Nevalainen, A. Manninen, H. Savolainen, Sci. Total Environ. 57 (1986) 49.
[8] K.K. Kleeberg, Y. Liu, M. Jans, M. Schlegelmilch, J. Streese, R. Stegmann, Waste Manage. 25 (2005) 872.
[9] J. Leach, A. Blanch, A.C. Bianchi, Atmos. Environ. 33 (1999) 4309.
[10] J. Volden, Y. Thomassen, T. Greibrokk, S. Thorud, P. Molander, Anal. Chim. Acta 530 (2005) 263.
[11] Ö.O. Kuntasal, D. Karman, D. Wang, S.G. Tuncel, G. Tuncel, J. Chromatogr. A 1099 (2005) 43.
[12] M.R. Ras, F. Borrull, R.M. Marcé, Talanta 74 (2008) 562.
[13] B. Ginzburg, I. Dor, O. Lev, Wat. Sci. Technol. 40 (1999) 65.
[14] K.J. James, M.A. Stack, J. Anal. Chem. 358 (1997) 833.
[15] J. Pawliszyn, Solid-Phase Microextraction: Theory and Practice, Wiley-VCH, New York, 1997.
[16] M.R. Ras, F. Borrull, R.M. Marcé, Talanta 77 (2008) 774.
[17] L. Pan, M. Chong, J. Pawliszyn, J. Chromatogr. A 773 (1997) 249.
[18] S.W. Tsai, C.M. Chang, J. Chromatogr. A 1015 (2003) 143.
[19] M. Ábalos, X. Prieto, J.M. Bayona, J. Chromatogr. A 963 (2002) 249.
[20] Y. Huang, L. Ortiz, J. García, P. Aguirre, R. Mujeriego, J.M. Bayona, Wat. Sci. Technol. 49 (2004) 89.
[21] M. Ábalos, J.M. Bayona, J. Pawliszyn, J. Chromatogr. A 873 (2000) 107.
[22] MINITAB version 14 for Windows, Minitab Inc., State College, PA, 2004.
[23] L.J. Van Gemert, ODOUR THRESHOLDS – Compilations of Odour Threshold Values in Air, Water and other Media, Oliemans Punter & Partners BV, The Netherlands, 2003.
[24] M.R. Ras, R.M. Marcé, F. Borrull, Talanta 72 (2007) 941.
[25] L. Pan, M. Adams, J. Pawliszyn, Anal. Chem. 67 (1995) 4396.
[26] J. De Crom, S. Claeys, A. Godayol, M. Alonso, E. Anticó, J.M. Sanchez, J. Sep. Sci. 33 (2010) 2833.
[27] J. Larreta, A. Vallejo, U. Bilbao, A. Usoblaga, G. Arana, O. Zuloaga, J. Sep. Sci. 35 (2007) 2293.
[28] M. Bao, O. Griffini, D. Burrini, D. Santianni, K. Barbieri, M. Mascini, Analyst 124 (1999) 459.
[29] S.B. Watson, B. Brownlee, T. Satchwill, E.E. Hargesheimer, Wat. Res. 34 (2000) 2818.
[30] J.J. Langenfeld, S.B. Hawthorne, D.J. Miller, J. Chromatogr. A 740 (1996) 139.
[31] A.T. Nielsen, S. Jonsson, J. Chromatogr. A 963 (2002) 57.
[32] K.C. Li, D. Shooter, Int. J. Environ. Anal. Chem. 84 (2004) 749.
[33] Y.H. Sung, T.Y. Li, S.D. Huang, Talanta 65 (2005) 518.
[34] AOAC International Guidelines for Laboratories Performing Microbiological and Chemical Analysis of Food and Pharmaceuticals, AOAC International, Gaithersburg, MD, 2006.
[35] K. Sieg, E. Fries, W. Püttmann, J. Chromatogr. A 1178 (2008) 178.