Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Chromatography A, 1046 (2004) 203–210
JOURNAL OF CHROMATOGRAPHY A
www.elsevier.com/locate/chroma

Identification and quantification of *n*-octyl esters of alkanoic and hexanedioic acids and phthalates as urban wastewater markers in biota and sediments from estuarine areas☆

Roser Chaler[a], Lourdes Cantón[b], Menchu Vaquero[b], Joan O. Grimalt[a,*]

[a] *Department of Environmental Chemistry, Institute of Chemical and Environmental Research (IIQAB-CSIC), Jordi Girona 18, 08034 Barcelona, Catalonia, Spain*
[b] *Department of Applied Chemistry, Faculty of Chemistry, University of the Basque Country, Donostia, The Basque Country, Spain*

Received 8 March 2004; received in revised form 19 June 2004; accepted 29 June 2004

Abstract

A gas chromatographic method for the identification and quantification of *n*-octyl esters (from *n*-octyl tetradecanoate to *n*-octyl hexacosanoate including dioctyl hexanedioate) and phthalates [dibutyl phthalate, benzyl butyl phthalate and di(2-ethylhexyl) phthalate] in sediments and biota from estuarine environments is described. Standards used for identification and quantification of some *n*-octyl esters were synthesized. The method has allowed the analysis of these compounds in polychaeta (*Nereis diversicolor*), oysters (*Crassostea angulata*), crabs (*Carcinus maenas*) and fish (*Chelon labrosus*, *Platichtys flesus* and *Chondostroma polylepis*) that were collected at different locations of the Urdaibai estuary (Bizkaia, Basque Country, Spain). Total phthalates and *n*-octyl esters ranged between 0.01 and 12 $\mu g\,g^{-1}$ and 0.05 and 9.4 $\mu g\,g^{-1}$, respectively, and were predominantly found in polychaeta and fish. Sediments did not contain these compounds in significant amount, only benzyl butyl phthalate, dioctyl hexanedioate and di(2-ethylhexyl) phthalate were found above limit of detection (0.01–0.05 $\mu g\,g^{-1}$).

Keywords: Plasticizers; Sediments; Environmental analysis; Alkanoic acids; Hexanedioic acids; Phthalates; Organic acids; Octyl esters; Endocrine disruptors

1. Introduction

Estuarine areas receive large amounts of urban wastewater residues of complex composition. Monitoring of these wastes is often difficult due to transformations after spillage. Comprehensive understanding of the extend of this type of pollution in the different environmental compartments, e.g. sediments or biota, requires an increasing number of urban waste markers.

Plastic usually represents about 10% of wastes [1]. Landfill areas, urban and industrial wastewater treatment plants (WWTPs) and municipal incinerators are major sources of these contaminants. Phthalates, phthalic acid esters [$C_6H_4(COOR)_2$] and its derivatives, are commonly used to provide flexibility to rigid polymers. These compounds are also used in paper and paperboard manufacturing as defoaming agents, in capacitors as dielectrics, in food as preservatives, in construction adhesives, paper food wrapping, nail polish, dyes, plastic food wraps, insect repellents, plastic pipes, lubricants, detergents, shampoos and bath soap [2]. They are therefore ubiquitous in the environment as consequence of the production of thousands of tons every year. Among them, di(2-ethylhexyl)phthalate (DEHP) is predominant due to its high production (nearly 90% of European plasticizers in use). This compound tends to accumulate in biota due to its low

☆ Presented at the 3rd Meeting of the Spanish Association of Chromatography and Related Techniques and the European Workshop: 3rd Waste Water Cluster, Aguadulce (Almeria), 19–21 November 2003.

* Corresponding author. Tel.: +34 944006100; fax: +34 932045904.
E-mail address: jgoqam@cid.csic.es (J.O. Grimalt).

0021-9673/$ – see front matter © 2004 Published by Elsevier B.V.
doi:10.1016/j.chroma.2004.06.098

water solubility and relatively high octanol–water partition coefficient [3].

Phthalic acid esters are practically insoluble in pure water [4]. However, they may be solubilized by interaction with fulvic and humic acids or become adsorbed onto particulate matter [5]. Slow physico-chemical degradation of these compounds by photolysis or hydrolysis results in half-live values in the order of years [6,7]. However, several bacteria [8–10], freshwater invertebrates [11,12] and fish [13,14] may metabolize them, completely or in part, in aerobic conditions. These compounds are classified as priority pollutants and endocrine disrupting compounds by the US Environmental Protection Agency (EPA) and other governmental agencies [15,16]. High priority has been posed on understanding their fate in aquatic ecosystems such as estuaries [17].

n-Octyl esters of alkanoic and hexanedioic acids are currently found in urban wastes. They are commonly used in cosmetics as moisturizing agents, in pharmaceutical industry as topically adsorption agents, in food industry as defoaming agents and in textile industry as lubricants [18–29]. After release into the aquatic environment they have half lives in the order of weeks. Thus, they may be used as markers of relatively recent urban waste pollution inputs.

Several methods have been reported for the analysis of phthalates in environmental samples [3,30–33], including water [34–36], sediments [37–39], air [35,40] and biota [34,41,42]. However, to the best of our knowledge, the environmental analysis of *n*-octyl esters has not previously been addressed. In the present study a method for the simultaneous analysis of both groups of compounds is proposed for the first time. The analysis of polychaeta, oysters, crabs, fish and sediments collected at different locations of the Urdaibai estuary (Bizkaia, the Basque Country, Spain) is reported as example.

2. Study area

The Urdaibai estuary drains into the Bay of Biscay (Fig. 1). It has an extension of 220 km^2 along 12.5 km of the mouth of Oka River and other minor rivers, covering 1.9 km^2 with an average depth of 3 m and a maximum width of 1.2 km at the mouth. In 1984, it was declared Biosphere's Reserve by UNESCO. River discharges are dominant in the upper reaches and tidal inflow in the lower euhaline zone. The upper segment is partially mixed during low river regime and well mixed during enhanced river flows. The lower estuary is always well mixed by tidal flushing. A population of about 45,000 people living in several towns is situated at the source of Oka River. High nutrient loads from a WWTP are incorporated in the upper estuary.

The sampling sites selected for study are shown in Fig. 1. Samples were taken in February, June and October 1994. Sediments and organisms were analyzed after each sampling process, in April 1994, September 1994 and January 1995. Muxika and the sampling points before and after the WWTP are located in the upper estuary. Murueta, San Kristobal, Arteaga and Kanala are representative sites of the middle estuary. Txatxarramendi and Laida are characteristic of the lower estuary.

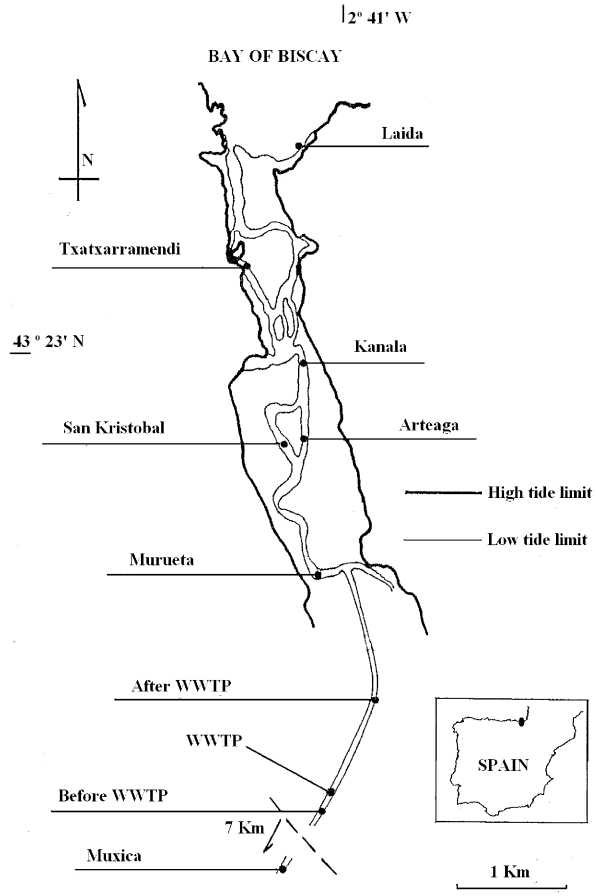


Fig. 1. Map of the Urdaibai estuary indicating the sampling sites.

3. Experimental

3.1. Chemicals

Solvents and reagents were from Merck, e.g. residue-analysis-grade *n*-hexane (reference 1.04371), methylene chloride (reference 1.06054), methanol (reference 1.06011), *iso*-octane (reference 1.15440), water for chromatography (reference 1.15333), bis(trimethylsilyl)trifluoroacetamide (reference 1.10255), silica (Kieselgel 40, 70–230-mesh), alumina (aluminium oxide, 70–230-mesh) and copper powder (<63 μm). Potassium hydroxide pellets and sodium hydroxide pellets were from Panreac.

3.2. Sampling

Polychaeta were collected with metallic tweezers. They were kept devoid of food in continuously aerated glass seawater tanks containing a layer of clean fine sand at the bottom (Probus 023700). Natural seawater was circulated

Table 1
Limits of detection ($\mu g\,g^{-1}$) in sediment and biota samples

	Compound[a]	Polychaeta	Crabs	Fish	Oysters	Sediments
1	Dibutyl phthalate	0.01	0.01	0.01	0.02	0.02
2	*n*-Octyl tetradecanoate	0.02	0.02	0.02	0.01	0.01
3	Benzyl butyl phthalate	0.02	0.05	0.02	0.02	0.01
4	Dioctyl hexanodioate	0.02	0.05	0.05	0.05	0.01
5	*n*-Octyl hexadecanoate	0.04	0.02	0.05	0.03	0.03
6	Di(2-ethylhexyl) phthalate	0.02	0.02	0.04	0.01	0.01
7	*n*-Octyl heptadecanoate	0.01	0.02	0.02	0.01	0.02
8	*n*-Octyl octadecanoate	0.01	0.03	0.03	0.02	0.02
9	*n*-Octyl eicosanoate	0.02	0.01	0.01	0.01	0.01
10	*n*-Octyl docosanoate	0.02	0.04	0.01	0.01	0.01
11	*n*-Octyl tricosanoate	0.02	0.03	0.02	0.01	0.02
12	*n*-Octyl tetracosanoate	0.02	0.03	0.02	0.01	0.02
13	*n*-Octyl hexacosanoate	0.02	0.03	0.01	0.01	0.01

[a] Linear calibration range: 0.01–30 $\mu g\,g^{-1}$.

semicontinuously through active charcoal and glass–wood filters in a thermally controlled system before introduction into the tanks. Polychaeta were maintained in this environment for 48 h in order to purge their gut contents and enhance elimination of adhering sediment prior to analysis.

Oysters and crabs were hand picked. Fish were collected using a web-like sampling metallic cage. Muscle was then dissected for analysis.

Sediments were collected with a Van Veen grab sampler. Sub-samples were taken with a metallic spatula from the central part of the sampler, avoiding the contact zones with the walls. They were then wrapped with aluminium paper and subsequently transferred to amber glass containers with the cap protected with aluminium foil. Sediment and biota subsamples were kept at −20 °C until analysis.

3.3. Blanks

All glassware was washed with detergent and successively rinsed with tap water, Milli-Q-grade water and reagent grade acetone. Then, it was introduced in an oven and heated to 220 °C overnight. Before use it was rinsed with hexane. Silica gel, alumina and the Soxhlet thimbles were extracted with dichloromethane–methanol (2:1, v/v) in a Soxhlet apparatus for 24 h. After evaporation, the silica gel and the alumina were heated for 12 h at 120 and 350 °C, respectively. These precautions led to blank values were below detection limits of all compounds under analysis (Table 1).

Procedure blanks were run between the samples (one every batch of five samples). Field blanks were taken in each sampling expedition.

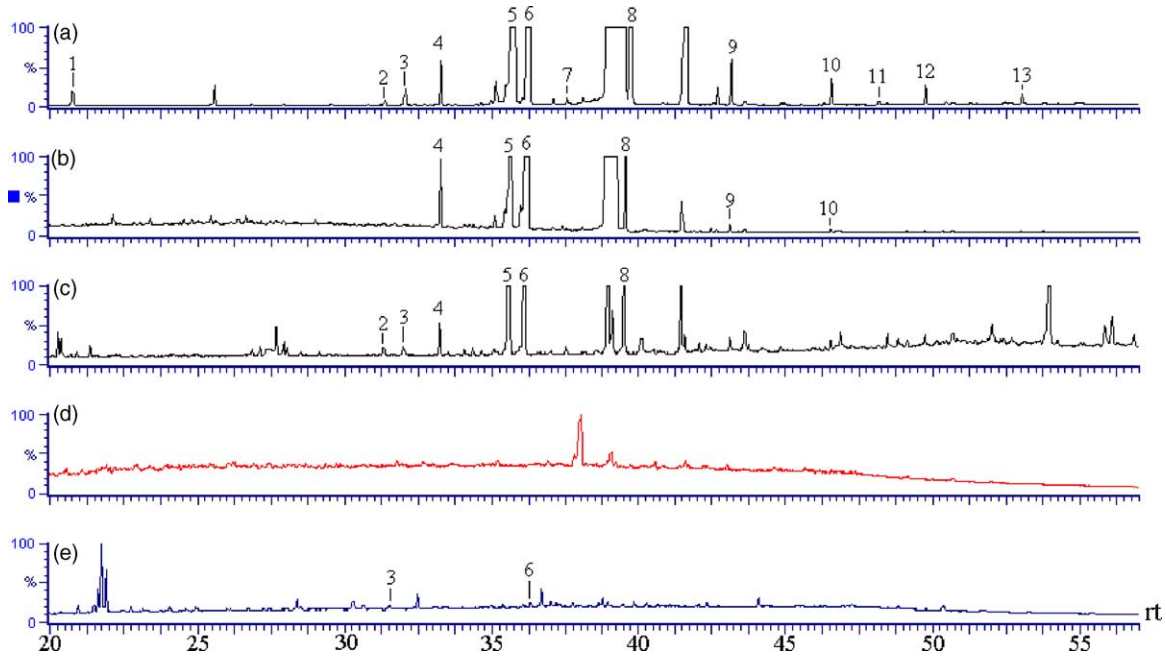


Fig. 2. Gas chromatograms (FID) showing the composition of phthalates and *n*-octyl esters in (a) fish, (b) oysters, (c) polychaeta and sediments [(d) Murueta; (e) after WWTP] from Urdaibai estuary. Peak labels as in Fig. 4.

Table 2
Mass spectrometric data of the compounds identified in biota and sediments

Peak no.	Compound	Retention time (min)	Molecular mass	Characteristic fragments (*m/z*; %)
1	Dibutyl phthalate	20.768	278	149 (100), 57 (20), 76 (10), 104 (9), 223 (9), 205 (7), 93 (3), 167 (2), 278 (1)
2	*n*-Octyl tetradecanoate	31.369	340	57 (100), 70 (95), 71 (78), 112 (72), 83 (38), 211 (23), 229 (12), 97 (11), 129 (8), 185 (5), 157 (4), 340 (1)
3	Benzyl butyl phthalate	32.070	312	149 (100), 91 (70), 206 (22), 65 (12), 104 (12), 123 (12), 238 (3), 178 (3), 312 (1)
4	Dioctyl hexanedioate	33.286	370	129 (100), 57 (62), 70 (51), 112 (45), 147 (38), 83 (30), 101 (18), 241 (6), 259 (3), 370 (1)
5	*n*-Octyl hexadecanoate	35.837	368	112 (100), 70 (95), 71 (92), 57 (91), 83 (73), 239 (42), 256 (32), 97 (22), 73 (20), 129 (15), 368 (3)
6	Di(2-ethylhexyl) phthalate	36.303	390	149 (100), 167 (82), 57 (70), 71 (68), 113 (47), 83 (32), 279 (31), 104 (20), 93 (8), 132 (8), 122 (8), 390 (1)
7	*n*-Octyl heptadecanoate	37.570	382	112 (100), 57 (95), 70 (88), 71 (85), 83 (47), 69 (27), 253 (18), 271 (15), 97 (15), 73 (12), 129 (9), 382 (1)
8	*n*-Octyl octadecanoate	39.737	396	112 (100), 57 (77), 71 (75), 70 (68), 83 (38), 285 (28), 267 (25), 69 (22), 97 (16), 129 (12), 185 (8), 396 (3)
9	*n*-Octyl eicosanoate	43.171	424	57 (100), 112 (98), 71 (83), 70 (68), 83 (39), 313 (20), 97 (18), 295 (13), 61 (9), 129 (8), 157 (6), 424 (3)
10	*n*-Octyl docosanoate	46.554	452	57 (100), 112 (92), 71 (80), 70 (58), 83 (33), 97 (17), 341 (15), 323 (8), 61 (8), 129 (7), 157 (5), 452 (2)
11	*n*-Octyl tricosanoate	48.155	466	57 (100), 71 (88), 112 (83), 70 (50), 83 (37), 97 (19), 355 (17), 129 (8), 61 (7), 337 (6), 157 (5), 466 (2)
12	*n*-Octyl tetracosanoate	49.755	480	57 (100), 112 (93), 71 (78), 70 (52), 83 (33), 97 (17), 369 (17), 61 (8), 129 (7), 351 (6), 157 (6), 480 (2)
13	*n*-Octyl hexacosanoate	53.038	508	112 (100), 57 (98), 71 (78), 70 (52), 83 (34), 97 (18), 397 (17), 61 (10), 129 (9), 379 (5), 157 (5), 508 (1)

3.4. Sample preparation and fractionation

The analytical procedure was developed from extraction and fractionation methods that were already used and tested in previous environmental studies involving other compounds [43–47]. Biota and sediments were defrosted the same day of analysis. Biota tissues (18 g) were cut in small pieces and introduced into centrifuge tubes where they were treated with 6 M NaOH (10 mL) at 30 °C for 18 h, in the dark, in a stirred bath. After this digestion period, the alkaline solution was extracted with *n*-hexane (3 × 15 ml). The extracts were concentrated to 0.5 mL by vacuum rotary evaporation and fractionated on a chromatographic column packed with 8 g each of 5% water-deactivated alumina (top) and 5% water-deactivated silica (bottom). Three fractions were collected: 20 mL of *n*-hexane, 20 mL of *n*-hexane–methylene chloride (90:10) and 40 mL of *n*-hexane–methylene chloride (80:20). The third fraction was the one of interest for the present study. Loss of analytes during the concentration was minimized by vacuum rotary evaporation to 0.5 ml without bath heating, transfer to vials and further evaporation under a gentle nitrogen stream, again without heating. Finally, the fractions were re-dissolved in *iso*-octane. Losses by evaporation were about 30% in the worst cases.

Wet sediments (20 g) were Soxhlet extracted with methylene chloride–methanol (2:1) for 36 h. The extract was hydrolysed with 6 M KOH in methanol for 12 h in the dark. The alkaline solution was extracted with *n*-hexane (3 × 30 mL) and treated with copper activated with HCl attack (1 g). The copper suspension was maintained for 2 h in the dark. The *n*-hexane solution was concentrated and fractionated by column chromatography as indicated for biota.

3.5. Instrumental analysis

Gas chromatography (GC) was performed on a Perkin-Elmer 8310 equipped with a flame ionization detection (FID) system and a 30 m × 0.32 mm i.d. SPB5 non-polar fused-silica capillary column (film thickness: 0.25 μm). Helium was used as carrier gas (1.5 ml min^{-1}). The oven temperature was programmed from 60 to 320 °C at 6 °C min^{-1}, with an isothermal hold of 10 min. Injection was in the splitless mode (hot-needle technique) keeping the split valve closed for 48 s. The injector and FID temperatures were 300 and 330 °C, respectively. Nelson software (Perkin-Elmer) was used for data acquisition and analysis. Typical injection volumes were 1 μl.

GC coupled to mass spectrometry (MS) was performed on a Thermo Quest MD800 equipped with a 30 m × 0.25 mm i.d. HP5-MS non-polar fused-silica capillary column (film thickness: 0.25 μm). Helium was used as carrier gas (1.0 ml min^{-1}). The oven temperature was programmed from 90 to 120 °C at 15 °C min^{-1} and then to 300 °C at 4 °C min^{-1}, with an isothermal hold of 15 min. Injection was in the splitless mode (hot-needle technique) keeping the split valve closed for 48 s. Injector temperature was 300 °C. The ion source and transfer-line temperatures were 200 and 280 °C, respectively. Mass spectra were acquired in the electron impact mode at 70 eV ionization potential. Data

were acquired in the full-scan mode between *m*/*z* 50–550 at 1 scan s^{-1}. Mass-Lab software (Thermo Quest) was used for data acquisition and analysis. Typical injection volumes were 1 μl.

3.6. Identification and quantification

n-Octyl tetradecanoate (**2**) and *n*-octyl docosanoate (**10**) were synthesized as reference standards for the identification of the distributions of *n*-octyl ester homologues (Fig. 4). The GC–FID system was calibrated using several mixtures of these standards and phthalates. They were also used as external standards for quantification being automatically injected within each sequence of samples. Recoveries of these standards were between 70 and 90%. The samples were currently diluted in order to fit within the linear range (between 0.01 and 30 μg g^{-1}). Detection limits are shown in Table 1. The limits for phthalates in biota and sediments are coincident with those reported in other studies [48,49].

GC–MS was used in order to confirm compound identity based on mass spectral interpretation and comparison of retention indices.

4. Results and discussion

4.1. Compound identification

Representative samples of polychaeta, oysters, crabs and fish were randomly collected in February, June and October 1994 at the locations described above (Fig. 1). Representative chromatograms of these samples are shown in Fig. 2. *n*-Octyl hexadecanoate, DEHP and *n*-octyl octadecanoate were currently major compounds. Mass spectra of some of the compounds found in the samples are shown in Fig. 3. The diagnostic ions, molecular weight and retention time data are given in Table 2.

The mass spectrum of *n*-octyl hexadecanoate (**5**; Fig. 4) exhibits the characteristic *m*/*z* 112 ion of the *n*-octyloxy alkyl esters, corresponding to the octyl group after hydrogen transfer to the alkylcarboxylate (Fig. 3). It also shows two prominent ions at *m*/*z* 256 and 239 that correspond to the hexadecanyl carboxylate after incorporation of one hydrogen and the hexadecane acyl moiety, respectively. These mass fragments are easily observed in polar fractions isolated from real samples and can be used as diagnostic ions. The molecular ions in the mass spectra of these *n*-octyl esters are weak. However, as shown in Fig. 3 for real sample spectra, magnification of the higher mass fragment region provides well defined molecular ions above baseline noise, e.g. *m*/*z* 368 for *n*-octyl hexadecanoate.

Phthalates can be easily recognized by their characteristic *m*/*z* 149 fragment, e.g. the spectrum of DEHP (**6**; Fig. 4) has the characteristic *m*/*z* 149 and 167 ions of the di(2-alkyl) phthalates. Magnification of the high mass spectral region also allows the recognition of the molecular ion (Fig. 3).

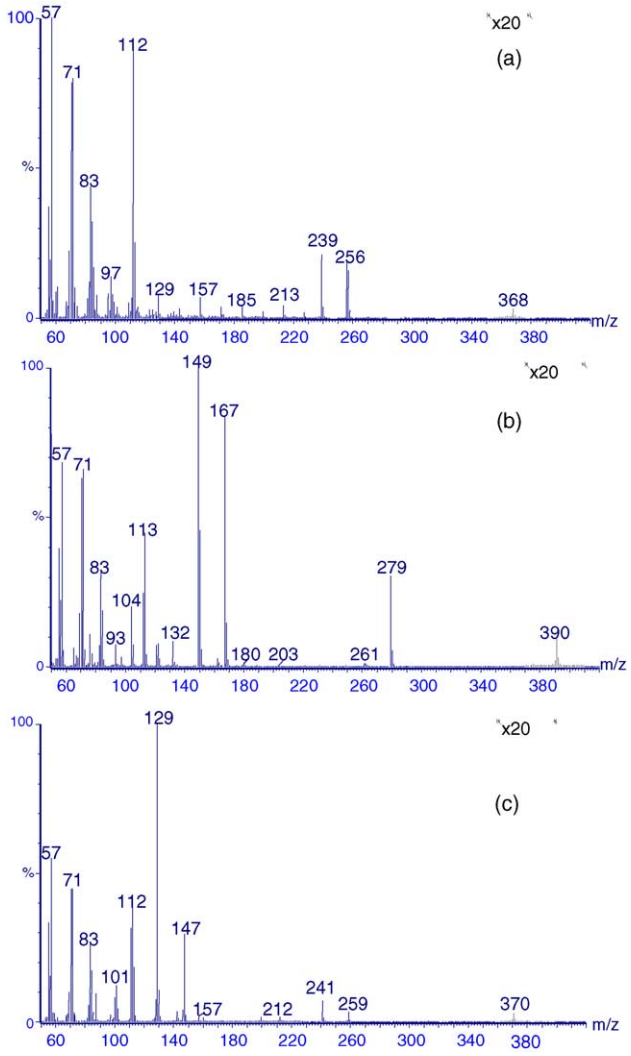
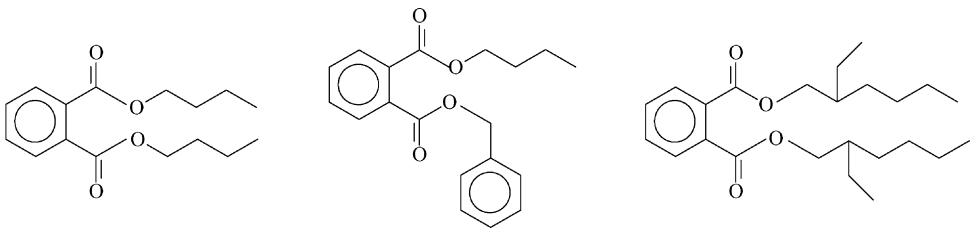


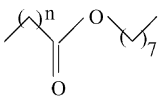
Fig. 3. Mass spectra of (a) *n*-octyl hexadecanoate, (b) di(2-ethylhexyl) phthalate and (c) dioctyl hexanedioate.

Dioctyl hexanedioate (**4**; Fig. 4) exhibits a spectrum dominated by *m*/*z* 129 corresponding to the octyloxy moieties. Again, the octyl group mass fragment, *m*/*z* 112, is found. Magnification of the high mass fragment zone allows again the observation of the molecular ion (*m*/*z* 370) without ambiguity (Fig. 3).

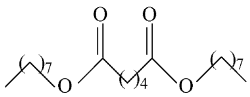
Mass fragmentograms for the combined *m*/*z* 112, 129 and 149 ions allows the enhancement of the response of these plasticizers within the GC–MS traces (Fig. 5). In agreement with their mass spectral abundance, use of mass fragmentograms based on these ions allow increased specificity and sensitivity for detection of *n*-octyl esters and phthalates in real samples. In this respect, mass spectral interpretation and examination of the GC–FID and *m*/*z* 112 + 129 + 149 fragmentograms have allowed the identification of 10 *n*-octyl ester derivatives (from *n*-octyl tetradecanoate (**2**; Fig. 4) to *n*-octyl hexacosanoate (**13**) including dioctyl hexanedioate (**4**)) and three different phthalates [dibutyl phthalate (**1**), benzyl butyl phthalate (**3**) and DEHP (**6**)].



Dibutil phthalate (**1**) Benzyl butyl phthalate (**3**) Di-(2-ethylhexyl) phthalate (**6**)



n= 12, *n*-octyl tetradecanoate (**2**);
n= 14, *n*-octyl hexadecanoate (**5**);
n= 15, *n*-octyl heptadecanoate (**7**);
n= 16, *n*-octyl octadecanoate (**8**);
n= 18, *n*-octyl eicosanoate (**9**);
n= 20, *n*-octyl docosanoate (**10**);
n= 21, *n*-octyl tricosanoate (**11**);
n= 22, *n*-octyl tetracosanoate (**12**);
n= 24, *n*-octyl hexacosanoate (**13**)



Dioctyl hexanedioate (**4**)

Fig. 4. Phthalates and *n*-octyl esters identified in the biota and sediment samples collected in Urdaibai estuary.

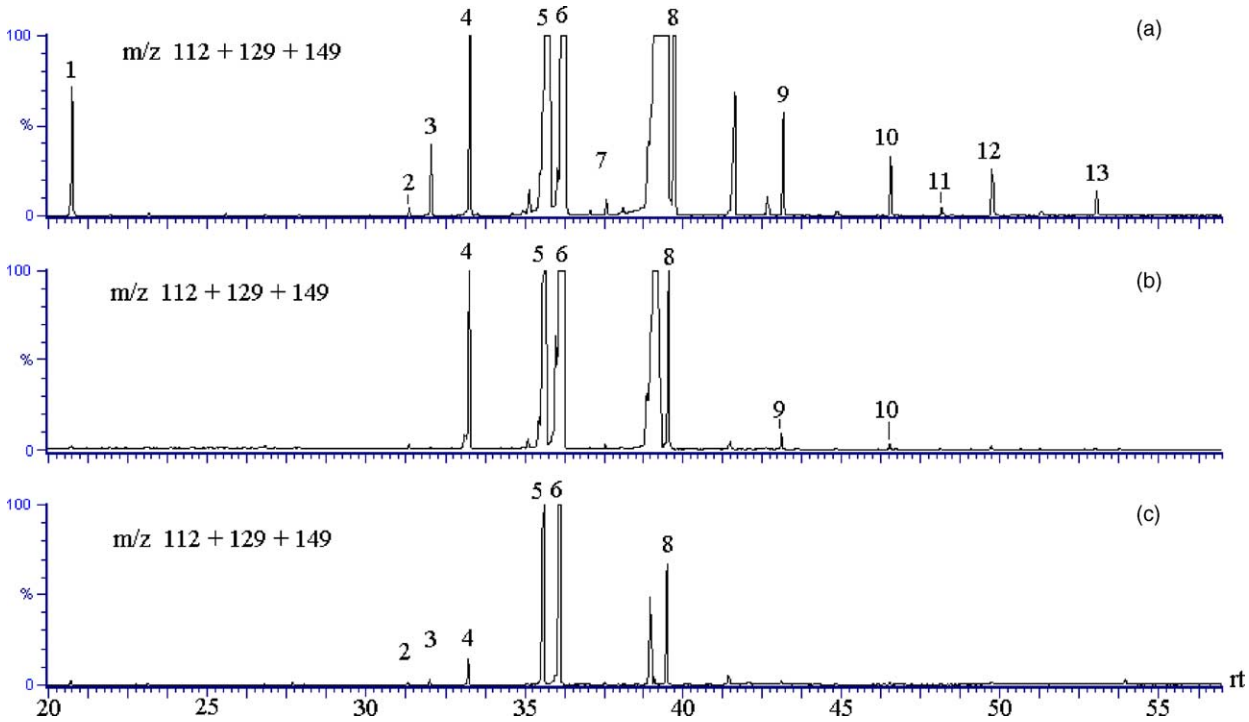


Fig. 5. Mass fragmentograms showing the composition of phthalates and *n*-octyl esters in (a) fish, (b) oysters and (c) polychaeta from Urdaibai estuary. Peak labels as in Fig. 4.

4.2. Quantification in biota and sediments

The concentrations of the compounds identified in polychaeta (*Nereis diversicolor*), oysters (*Crassostea angulata*), crabs (*Carcinus maenas*), fish (*Chelon labrosus*, *Platichtys flesus* and *Chondostroma polylepis*) and sediments are shown in Fig. 6. The results represented in this figure correspond to the samples in which highest concentrations were found in each sampling period (February, June and October 1994).

Fish were the only type of organisms in which all above mentioned compounds were found. These organisms where those having highest concentrations of *n*-octyl esters

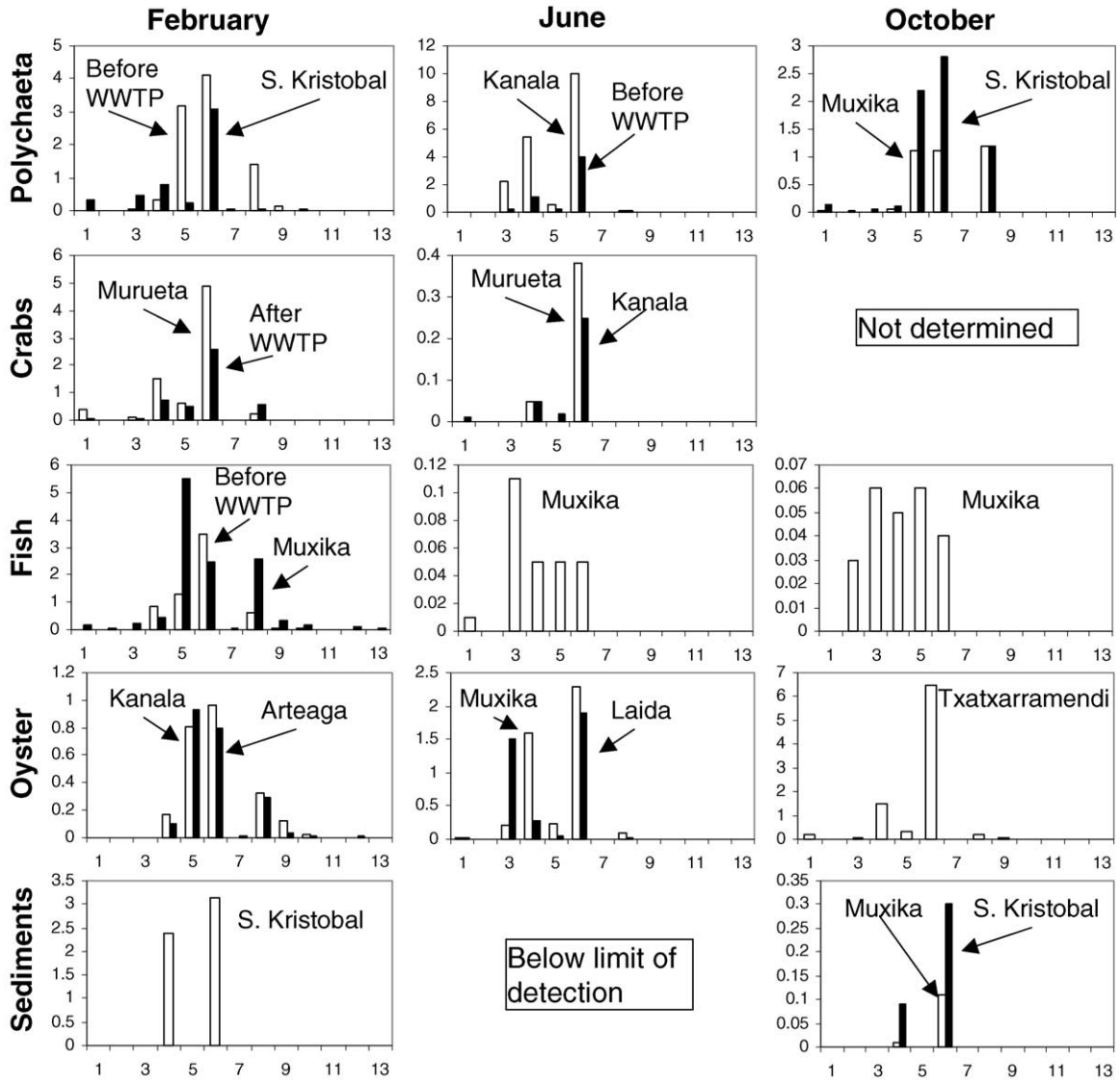


Fig. 6. Summary of the major concentrations of *n*-octyl esters and phthalates in the sample sites from Urdaibai estuary. Numbers in abscissas refer to the compounds listed in Table 1.

(<0.01–9.4 $\mu g\,g^{-1}$). In fact, the highest-molecular-mass *n*-octyl esters identified, e.g. between *n*-octyl tricosanoate (**11**; Fig. 4) and *n*-octyl hexacosanoate (**13**; Fig. 4), were hardly found in other biota than fish (Fig. 6).

Polychaeta showed the major concentrations of phthalates (<0.01–12 $\mu g\,g^{-1}$) and also high abundance of the *n*-octyl esters (<0.01–6.15 $\mu g\,g^{-1}$). Phthalates were also abundant in crabs (<0.01–4.9 $\mu g\,g^{-1}$) but these organisms contained *n*-octyl esters in low concentration (up to 0.64 $\mu g\,g^{-1}$). Among biota, the lowest values of both phthalates and *n*-octyl esters were found in oysters (<0.01–2.3 and <0.01–1.6 $\mu g\,g^{-1}$).

In contrast, sediments did not show detectable amounts for most of these compounds, only benzyl butyl phthalate (**3**; Fig. 4), dioctyl hexanedioate (**4**; Fig. 4) and DEHP (**6**) were found in few samples at low concentrations (Fig. 6). The DEHP levels were similar to those found in industrial areas along the Missouri River, 3.2 $\mu g\,g^{-1}$ [42], and the Mersey River estuary, 1.2 $\mu g\,g^{-1}$ [37], but lower than those found in Taihu Lake, 13 $\mu g\,g^{-1}$ [50], Mexico Bay, 20 $\mu g\,g^{-1}$ [41], or Klang River, 0.49–15 $\mu g\,g^{-1}$ [39], the later receiving untreated waste from plastic factories. Studies in agricultural zones such as Texas Rio Grande valley also reported higher levels of phthalates, 12–80 $\mu g\,g^{-1}$ [51], than Urdaibai estuary.

The lack of *n*-octyl alkanoates in sediments suggests that these compounds have low accumulation potential in this compartment or that significant degradation processes occur soon after sedimentation. Phthalates and the dicarboxylic dioctyl ester, the compounds identified in the Urdaibai estuary, are likely more resistant to microbial action since their structures are more branched or cyclic than *n*-octyl alkanoates.

5. Conclusions

The gas chromatographic method described in the present study is useful for the identification and quantification of phthalates and *n*-octyl esters in biota and sediments. Ten *n*-octyl ester derivatives (from *n*-octyl tetradecanoate to *n*-octyl hexacosanoate including dioctyl hexanedioate) and three different phthalates (dibutyl phthalate, benzyl butyl phthalate and DEHP) have been analyzed in polychaetes, oysters, crabs, fish and sediments from the Urdaibai estuary.

DEHP was the only compound found in all types of biota examined. Dioctyl hexanedioate and *n*-octyl hexadecanoate were found in all polychaetes and fish. These two types of organisms were those showing the major concentrations of these compounds, 0.01–12.23 and 0.05–9.37 $\mu g\, g^{-1}$ for total phthalates and *n*-octyl esters, respectively.

In contrast, sediments did not show the presence of most of these compounds, only benzyl butyl phthalate, dioctyl hexanedioate and DEHP were found in a few of them. These low concentrations suggest that, although both *n*-octyl esters and phthalates may be good markers of urban waste in biota, they may not be useful in sediments.

Acknowledgement

Financial support from the EU Project MATBIOPOL EVK3-CT-1999-00010 is acknowledged.

References

[1] G. Nebbia, A. Brera, Inquinamiento 1 (1994) 8.
[2] J.A. Brydson, Plastic Materials, Butterworth, 1985.
[3] M. Vitali, M. Guidotti, G. Macilenti, C. Cremisini, Environ. Int. 23 (1997) 337.
[4] D.L. DeFoe, G.W. Holcombe, D.E. Hammermeister, K.E. Biesinger, Environ. Toxicol. Chem. 9 (1990) 623.
[5] L.A. Al-Omran, M.R. Preston, Environ. Pollut. 46 (1987) 177.
[6] N.L. Wolfe, W.C. Steen, L.A. Burns, Chemosphere 9 (1980) 403.
[7] W.E. Gledhill, R.G. Kaley, W.J. Adams, O. Hicks, P.R. Michael, V.W. Saeger, G.A. LeBlanc, Environ. Sci. Technol. 14 (1980) 301.
[8] W.C. Steen, D.F. Paris, G.L. Baughman, in: R.A. Baker (Ed.), Contaminants and Sediments: Fate and Transport, Case Studies, Modelling, Toxicity, vol. 1, Ann Arbor Science Pub., Ann Arbor, MI, 1980.
[9] D.W. Ribbons, P. Keyser, D.A. Kunz, B.F. Taylor, R.W. Eaton, B.N. Anderson, in: D.T. Gibson (Ed.), Microbial Degradation of Organic Compounds, Marcel Dekker, New York, 1984.
[10] R. Kurane, Microbiol. Sci. 3 (1986) 92.
[11] H.W. Wofford, C.D. Wilsey, G.S. Neff, C.S. Giam, J.M. Neff, Ecotoxicol. Environ. Saf. 5 (1981) 202.
[12] W.W. Walker, C.R. Cripe, P.H. Pritchard, A.W. Bourquin, Chemosphere 13 (1984) 1283.
[13] A.H. Karara, W.L. Hayton, Drug. Metab. Dispos. 16 (1988) 146.
[14] M.G. Barron, B.D. Tarr, W.L. Hayton, Toxicol. Appl. Pharmacol. 88 (1987) 305.
[15] L.H. Keith, W.A. Telliard, Environ. Sci. Technol. 13 (1979) 416.
[16] Endocrine-Disrupting Substances in the Environment: What Should Be Done? Environment Agency, UK, 1998.
[17] J.B. Pearce, L. Despres-Patanjo, Aquatic Toxicol. 11 (1988) 323.
[18] J.B.M. Ratray, J. Am. Oil Chem. Soc. 61 (1984) 1701.
[19] X.-L. He, B.-Q. Chen, T.-W. Tan, J. Mol. Catal. B: Enzym. 18 (2002) 333.
[20] M. Ito, A. Ishida, JP Patent 20427374 A2 (2004).
[21] K. Suda, T. Sugimoto, S. Okutani, JP Patent 2003278048 A2 (2003).
[22] K. Taniguchi, K. Ishiyama, JP Patent 02242971 A2 (1990).
[23] W. Catlin, M.F. Fuller, US Patent 2904522 (1959).
[24] S. Kawai, M. Ichinohe, JP Patent 2003300816 (2003).
[25] M. Singh, US Patent 2002098158 A1 (2002).
[26] B. Mompon, WO Patent 2002051378 A1 (2002).
[27] P. Noah, US Patent 2003215397 A1 (2003).
[28] K.-U. Furstenau, CA Patent 2202707 AA (1997).
[29] M. Fujii, K. Shiozawa, T. Henmi, S. Yamanouchi, H. Suzuki, N. Yamashita, M. Matsumoto, Int. J. Pharm. 137 (1996) 117.
[30] S.Y. Yuan, C. Liyçu, C.S. Liao, B.V. Chang, Chemosphere 49 (2002) 1295.
[31] H. Fromme, T. Küchler, T. Otto, K. Pilz, J. Muller, A. Wenzel, Water Res. 36 (2002) 1429.
[32] M. Petrovic, S. Lacorte, P. Viana, D. Barceló, J. Chromatogr. A 959 (2002) 15.
[33] R. Céspedes, M. Petrovic, D. Raldúa, U. Saura, B. Piña, S. Lacorte, P. Viana, D. Barceló, Anal. Bioanal. Chem. 378 (2004) 697.
[34] A. Thuren, Bull. Environ. Contam. Toxicol. 36 (1986) 33.
[35] C.S. Giam, H.S. Chan, G.S. Neff, E.L. Atlas, Science 199 (1978) 419.
[36] S. Jobling, T. Reynolds, R. White, M.G. Parker, J.P. Sumpter, Environ. Health. Persp. 103 (1995) 582.
[37] M.R. Preston, A. Al-Omran, Environ. Pollut. 62 (1989) 183.
[38] H.E. Schwartz, C.J.M. Anzion, H.P.M. Van Vliet, J.W. Copius Peerebooms, U.A.Th. Brinkman, Int. J. Environ. Anal. Chem. 6 (1979) 133.
[39] G.H. Tan, Bull. Environ. Contam. Toxicol. 54 (1995) 171.
[40] A. Thuren, P. Larsson, Environ. Sci. Technol. 24 (1990) 554.
[41] C.S. Giam, H.S. Chan, G.S. Neff, Anal. Chem. 47 (1975) 2225.
[42] F.L. Mayer, D.L. Stalling, J.L. Jonhson, Nature 238 (1972) 411.
[43] J.O. Grimalt, C. Marfil, J. Albaigés, Int. J. Environ. Anal. Chem. 18 (1984) 183.
[44] M. Aceves, J.O. Grimalt, J. Albaiges, F. Broto, L. Comellas, J. Chromatogr. 436 (1988) 503.
[45] L. Cantón, J.O. Grimalt, Chemosphere 23 (1991) 327.
[46] L. Cantón, J.O. Grimalt, J. Chromatogr. 609 (1992) 279.
[47] L. Berdie, J.O. Grimalt, J. Chromatogr. A 823 (1998) 373.
[48] M.J. Waldock, Chem. Ecol. 1 (1983) 261.
[49] J.C. Peterson, D.H. Freeman, Int. J. Environ. Anal. Chem. 12 (1982) 277.
[50] H. Wang, C. Wang, W. Wu, Z. Mo, Z. Wang, Chemosphere 50 (2003) 557.
[51] S.S. García, C. Ake, B. Clement, H.J. Huebner, K.C. Donnelly, S.L. Shalat, Environ. Int. 26 (2001) 465.