NEW POLYMERIC AND OTHER TYPES OF SORBENTS FOR SOLID-PHASE EXTRACTION OF POLAR ORGANIC MICROPOLLUTANTS FROM ENVIRONMENTAL WATER

ABSTRACT

This article describes the different types of sorbents for the solid-phase extraction of polar organic pollutants from environmental water and emphasizes the use of the new synthesized sorbents. The most important new sorbents are the chemically modified polymeric resins with functional groups such as acetyl, hydroxymethyl, benzoyl and *o*-carboxybenzoyl, and the highly crosslinked polymers. These sorbents are compared and their properties described. Recoveries for the most polar compounds are better when these new sorbents are used. Immunosorbents and molecularly imprinted polymers are new types of extraction sorbents which are specific for a group of compounds and they enable chromatograms with no matrix interference to be obtained when analyzing some polar compounds. The advantages and drawbacks of off-line and on-line procedures are also discussed, and solutions to their limitations are proposed.

Keywords: Solid-phase extraction; Polar pollutants; Water analysis; Polymeric sorbents; Immunosorbents; Molecularly imprinted polymers.

INTRODUCTION

Solid-phase extraction (SPE) overcomes many drawbacks of the liquid-liquid extraction (LLE) and also makes it possible to tune the selectivity, affinity and/or capacity via the development of new materials. SPE has been included in some US EPA Methods which analyze pollutants in drinking water (e.g. Methods 549.1 and 525.1 which use C_8 and C_{18} bonded silica sorbents, respectively, to analyze pesticides).

Solid-phase extraction and high performance liquid chromatography (HPLC) are both based on differential migration processes in which compounds are adsorbed and eluted as they are swept through a porous medium by a mobile-phase flow, which is dependent on the differential affinities between the sorbent material and the mobile phase. Retention is achieved with strong but reversible interactions between the analyte and the surface of the sorbent. Typical interactions are hydrophobic (van der Waals forces), polar (hydrogen bonding and dipole-dipole forces) or ion exchange interactions. Before selecting a sorbent for SPE, it is necessary to take into account some physico-chemical considerations such as the functional groups of the analytes, the nature of the bonded phase, the energetics of the interactions, the secondary interactions between the analytes and the sorbent, the interactions between the sorbent and the components of the sample matrix, and the interactions between the analytes and the sample matrix [1].

Apart from the main goal of extracting traces of the compounds of interest, SPE is also used to remove the interfering components of the matrix, change the solvent (e.g. aqueous to organic) and store and transport the analytes [2].
To date, typical SPE materials are modified silicas with C_8, C_{18}, CN and other groups, carbon blacks and styrene-divinylbenzene copolymers (PS-DVB). The most problematic compounds are the polar ones, whose breakthrough volume is low in most of the sorbents mentioned. Recently, new functionalized polymeric

sorbents and highly crosslinked polymers have appeared as alternatives to the typical SPE sorbents for the trace enrichment of these polar analytes.

In this article, we review the new sorbents for the SPE of polar organic pollutants from environmental waters and compare them with the classical ones. We concentrate on chemically modified polymeric resins and highly crosslinked sorbents because they have a greater capacity than the typical SPE materials for polar compounds. Also, we propose that immunosorbents and molecularly imprinted polymers, which are new tools for the SPE of environmental pollutants from water, be used to selectively retain specific groups of analytes.

SPE PROCEDURES

The most frequently used design in SPE is the cartridge. Many different types and amounts of sorbent are contained between two polyethylene or stainless steel frits in glass or polypropylene cartridges which have different column volumes. Another design which has become available in the last few years is the disk. Two different kinds of disk are now available: particle-loaded membranes (PLMs) and particle-embedded glass fiber disks (PEGFDs). The former is the most common. PLMs consist of a web of polytetrafluoroethylene (PTFE) microfibrils in which sorbent particles with a diameter of about 8 μm are suspended, and the membranes have a homogeneous flexible structure. The PEGFDs contain particles embedded in a glass fiber supporting matrix.

Cartridge design has certain disadvantages for water analysis. For example, the cross-sectional area is small, therefore sample processing rates are slow and the tolerance to blockage by particles and adsorbed matrix components is low, and channelling reduces the capacity to retain analytes. Some of these problems may be solved by using disks [3,4]. But, at present, cartridges are still the most frequently used, mainly because of the limited number of commercially

available sorbents on disks. Normally cartridges use less solvent volume than disks [5], but some authors used more solvent with cartridges [3].

Empore disks containing C_{18} or PS-DVB are the two main types of disk format in use. One of the drawbacks of using disks instead of cartridges is the decrease in the breakthrough volume, mainly for more polar compounds. For this reason, disks are used when there is a strong interaction between the analyte and the sorbent. These types of disk have been used to determine phenol and chlorophenols in sea water, and in this respect they have been compared with C_{18} cartridges. The recovery of these analytes was increased by using two disks in the same device [5].

Recently, SPE disks with new sorbents have become available, such as PS-DVB containing functional groups (sulfonic and acetyl groups) with the resin beads permanently encased in a PTFE membrane [4]. Membranes impregnated with acetyl-PS-DVB have been applied to phenols and higher recoveries were obtained compared with PS-DVB disks [6]. Membranes embedded with sulfonated PS-DVB have been used to extract polar analytes, such as phenolic compounds, and non-polar analytes, such as halogenated alkanes, which were all efficiently recovered in the preconcentration of 15 ml of sample, with recoveries over 90% for all compounds [7]. Extraction disks with a strong anion exchanger (SAX) have recently been developed and applied to determine haloacetic acids using the EPA Method 552.1.

SPE can be carried out in the off-line and on-line modes. On-line procedures mainly use an extraction sorbent in a precolumn but disks can also be used in an special device which allow the use of different numbers and types of disks [8].The methods which hyphenate SPE with HPLC are the most frequently used for determining polar pollutants in environmental water. Capillary gas chromatography (CGC) requires a derivatization step to enhance the volatility of these analytes and supercritical fluid chromatography (SFC) requires more

expensive equipment and an additional step to dry the sorbent before the desorption.

In the on-line procedures there is no sample manipulation between preconcentration and analysis, so loss and contamination risk are avoided and detection limits and reproducibility values can be better. Likewise, all the especies are analyzed, so the volume of sample can be smaller than in off-line procedures, the consumption of organic solvents is lower and the potential for automation is higher [9]. But the off-line SPE approach will remain useful for analyzing complex samples due to its greater flexibility and the fact that it can analyze the same extract using various techniques.

In on-line SPE procedures, sorbents are chosen not only for their efficiency at trapping analytes but also for their compatibility with the sorbent in the analytical column which should have the same (or higher) retention capabilities as the precolumn material. The precolumn should be as small as possible in order to prevent band broadening [1,10]. This effect can be minimized by using a suitable gradient which causes peak compression at the top of the analytical column and a good compromise between breakthrough volume and peak efficiency [11]. The equipment design has also been modified to reduce this problem. In this new design, excessive peak broadening was effectively supressed by eluting the analytes, in the backflush mode, by the organic solvent of the mobile phase alone and then both solvents of the mobile phase were mixed before reaching the analytical column. In this case the mixer chamber was replaced with a T junction without dead volume [12]. This design was used to quantitatively desorb phenolic compounds and pesticides from polymeric sorbents and graphitized carbon blacks on-line coupled to a C_{18} analytical column and narrow peaks were obtained [10,12-17]. However, when using this design in the SPE of environmental samples, a slight increase in the matrix effect was observed. This was caused by the greater strength of the solvent used to desorb the analytes from the precolumn [12].

When a powerful sorbent is used in the on-line SPE of real water samples a broad initial band appears due to the presence of humic and fulvic acids. This band can make the analysis of most polar compounds difficult. Methods for reducing this band by chemical treatment with $KMnO_4$ [18] and Na_2SO_3 [13,16,17] prior to extraction have been applied. Fig. 1 shows the chromatograms of 50 ml of Ebro river water with and without the addition of Na_2SO_3. They were obtained by on-line SPE with a chemically modified polymeric sorbent with an *o*-carboxybenzoyl moiety. To solve matrix effects in humic-containing surface water samples, some authors have used restricted access which consists of a precolumn material that uses non-adsorptive size exclusion of macromolecules and simultaneous dynamic partitioning of the analytes [19].
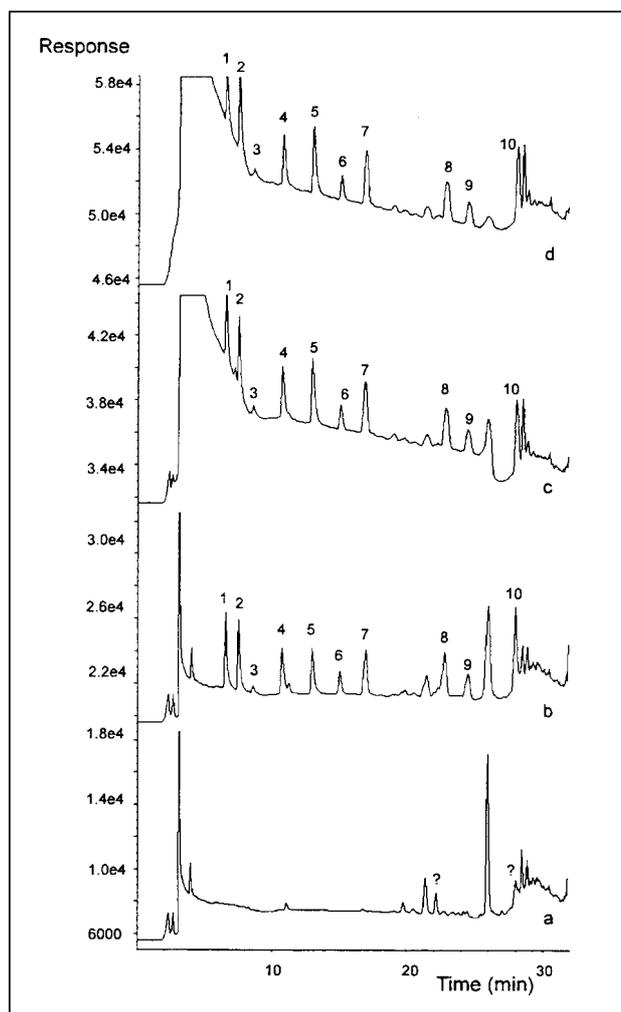
Fig. 1. Chromatograms obtained by on-line trace enrichment of 50 ml of Ebro river water with and without the addition of different volumes of 10% Na_2SO_3 solution for every 100 ml of sample in a chemically modified polymeric sorbent. (a) Blank with the addition of 1000 µl of sulfite. (b) Spiked with 4 µg l^{-1} of the analytes and 1000 µl of sulfite. (c) Spiked with 4 µg l^{-1} of the analytes and 500 µl of sulfite. (d) Spiked with 4 µg l^{-1} of the analytes without sulfite. Peak designation: 1, oxamyl; 2, methomyl; 3, Ph; 4, 4-NP; 5, 2,4-DNP; 6, 2-CP; 7, bentazone; 8, simazine; 9, MCPA; 10, atrazine.

SORBENTS FOR SOLID-PHASE EXTRACTION

The fact that the most commonly used materials for SPE are inadequate for solving the problems involved in isolating polar contaminants from large water volumes has encouraged investigators to evaluate alternative sorbent materials, such as chemically modified polymeric resins, highly crosslinked polymers, immunosorbents and molecularly imprinted polymers. All these new sorbents types will be discussed here.

Modified Silicas

The most widely-used bonded silica sorbents are octyl (C_8) [20-22], octadecyl (C_{18}) [18,21-23], ethyl (C_2) [20], butyl (C_4) [20], phenyl [22], cyclohexyl (CH) [23] and cyanopropyl (CN) [22]. They are reversed-phase sorbents, and so are adequate for aqueous samples. The interactions between the bonded functional group and the analyte carbonated chain are due to van der Waals forces. The most widely used modified silica in the trace enrichment of pollutants in environmental waters is the C_{18} sorbent because its capacity is greater than other bonded silicas [20] in the analysis of polar compounds. It has been shown that C_{18} is more efficient at extracting the most polar phenolic compounds than CH [23]. However, another study [22] has shown that phenyl bonded silica was more efficient than C_{18}, C_8 and cyanopropyl sorbents at preconcentrating phenolic compounds in water, even though the unsubstituted phenol was not completely recovered.

However, apart from the limited breakthrough volumes for polar analytes, these sorbents suffer from chemical limitations, such as residual surface silanol groups, a narrow pH stability range and poor selectivity.

Ion Exchangers

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The interactions between the analytes and the ion exchanger are electrostatic attractions. They are formed by a silica or polymeric support with a quaternary, secondary or primary amine (anion exchanger) or with sulfonic or carboxylic acid (cation exchanger) bonded functional groups.

Aminex A-28 (SAX), a quaternary ammonium anion exchanger, was used for the trace enrichment of phenol in tap and river water [24]. SCX, Aminex A-5 and Aminex A-7 have been used as cation exchangers for the preconcentration of polar pollutants in real water and industrial wastewater samples [25,26].

A new carboxylic acid functionalized crosslinked polymer was used for the SPE of fifteen different substituted anilines and lutidines from water which were retained on the resin by a cation exchange mechanism. But a second type of interaction, π-π, between the analytes and the resin was also present due to the unsaturated moieties of the resin. The more polar and less basic anilines were hardly retained on this resin [27].

Graphitized Carbon Black

Graphitized carbon blacks (GCBs) are obtained from heating carbon blacks at 2700-3000 ºC in an inert atmosphere. They are essentially non-specific and non-porous sorbents with a surface area of about 100 m^2 g^{-1} [28]. Because of the presence of positively charged chemical heterogeneities on their surface, they can be considered to be both reversed-phase sorbents and anion exchangers [29]. It has been shown that organic compounds are adsorbed on a GCB surface

more strongly than on a C_{18} surface when analyzing many acidic organic compounds in water [29].

Some commercial GCBs, such as Carbopack, Carbograph 1 and Envi-Carb, have been used in the analysis of several micropollutants such as phenolic compounds [11,13,28,29], pesticides [10,13,28,30] and surfactants [28] in drinking and river water. Two new GCB sorbents, Carbograph 4 and Carbograph 5, with surface areas greater than 100 m^2 g^{-1} were used to determine these analytes in water and they yielded better recoveries for the most polar compounds than Carbograph 1 [28].

Some drawbacks of the GCB sorbents are that they have excessive retention (some analytes can even be irreversibly adsorbed) but this can be overcome by performing the elution in the backflush mode [28]. Their mechanical stability is also poor. Porous immobilized graphitic carbon (PGC) is a new carbon-based sorbent in which the graphite is immobilized on a silica structure, and this is why PGC is more stable than GCB. The use of PGC was investigated for the trace enrichment of polar phenolic compounds and compared with polymeric sorbents; except for 4-chloro-2-aminophenol, the breakthrough volumes and detection limits were worse with the PGC than those obtained using the polymeric sorbents [11].

Polymeric sorbents

The most widely used polymeric sorbents are the styrene-divinylbenzene copolymers (PS-DVB) which have a hydrophobic surface. PS-DVB resins overcome many of the limitations of bonded silicas. The broader range of pH stability of these resins increases the flexibility of the method, and they have no silanol groups [31]. In addition, PS-DVB resins have greater analyte retention, mainly for polar compounds, than bonded silicas because their hydrophobic

surface contains a relatively large number of active aromatic sites which allow π-π interactions with unsaturated analytes.

XAD resins have long been used to extract a variety of organic pollutants from water. XAD-1, XAD-2 and XAD-4 are PS-DVB with a highly hydrophobic character. XAD-7 and XAD-8 are acrylic ester resins with a higher affinity for polar solutes [32]. XADs based on PS-DVB have some drawbacks including lack of selectivity, low breakthrough volumes for very polar compounds, low sampling rate, and they require extensive cleaning before use. XAD-2 has given better results than C_{18} and an activated carbon in the preconcentration of *N*-methylcarbamates and their degradation products in aqueous samples [33].

Another widely used PS-DVB copolymer is the PLRP-S resin. It has been used in the extraction of organic pollutants such as phenolic compounds [23] and pesticides [9] from natural waters. It has been shown to be more efficient than C_{18} bonded silica for the preconcentration of these analytes [9,23]. PLRP-S has the same drawbacks as XADs: lack of selectivity and low breakthrough volumes for very polar compounds.

In recent years, chemically modified resins have been developed and used in the SPE of polar compounds from environmental waters. Different polar functional groups such as acetyl [6,14,34,35], hydroxymethyl [34,35], benzoyl [15] and *o*-carboxybenzoyl [16,17] have been introduced into polymeric resins. These modified resins have excellent hydrophilicity, and they also give higher recoveries than their unmodified analogues [6,14,34,35]. This has been attributed to an increase in surface polarity which enables the aqueous sample to make better contact with the resin surface. The chemical structures of these sorbents are shown in Fig. 2.
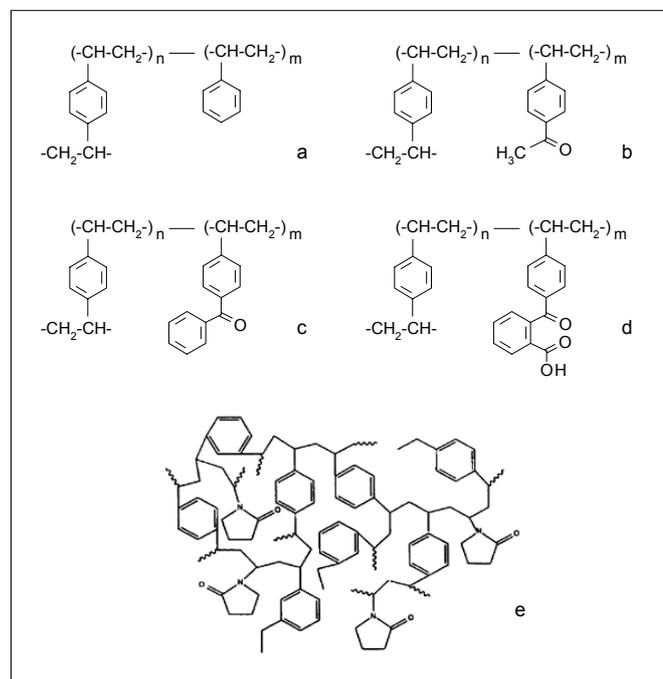
Fig. 2. Structures of different solid-phase extraction sorbents. Assignation: (a) styrene-divinylbenzene copolymer (PS-DVB), (b) functionalized PS-DVB with an acetyl moiety, (c) functionalized PS-DVB with a benzoyl moiety, (d) functionalized PS-DVB with an *o*-carboxybenzoyl moiety, (e) Oasis HLB.

They have also been compared with other SPE materials such as PLRP-S and GCB (Envi-Carb and Carbopack B) in the preconcentration of pesticides and phenolic compounds, and they yielded higher breakthrough volumes for the most polar compounds than the latter sorbents [14,15,17]. Table 1 compares the recoveries of several analytes on modified and unmodified resins and C_{18} bonded silica. The recoveries for many of the organic analytes tested were significantly higher with the modified sorbent [35]. Another functionalized PS-DVB with different functional groups were tested and compared with XAD-4 for the preconcentration of nitrobenzenes and their reduction products from water [36]. When XAD-4 was used, the recoveries of phenylenediamines were the poorest.

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Bond Elut PPL is a commercially available functionalized PS-DVB which has been developed to extract highly polar species such as phenol from large water volumes. Its extreme hydrophilicity ensures high recoveries. It has been used and compared with GCBs in the SPE of phenolic compounds and pesticides from natural waters, and recoveries were higher with the functionalized polymeric sorbent than with the GCBs, mainly for the most polar compounds [13,17]. Table 2 shows the recovery values obtained in the on-line SPE of different sample volumes of a mixture of pesticides and phenolic compounds with a GCB and Bond Elut PPL.

Slightly sulphonated resins have also been used [7,35]. They have excellent hydrophilicity and extract polar organic compounds more efficiently than underivatized resins. If a polymeric resin is sulfonated, it changes into a cation exchanger. A hydrogen bonding between the amine groups of some analytes and the sulfonic acid group on the resin is also possible. Some authors [37] have studied SPE for organic pollutants in water with a binary cartridge, made up of a chemically modified resin (acetyl-derivatized) and an anion exchanger phase, for use in a broadly-based screening method as an alternative to LLE.

Table 1
Recovery of phenolic and aromatic compounds with different sorbents in the preconcentration of 20 ml solution containing 5 mg l^{-1} of each analyte.

Compound	Recovery (%)			
	C_{18}	PS-DVB[a]	PS-DVB-CH_2OH	PS-DVB-$COCH_3$
Phenol	6	91	94	100
p-Cresol	16	91	98	101
p-Ethylphenol	66	96	99	101
2-NP	45	93	95	96

3-NP	<5	81	85	93
4-NP	<5	87	86	87
2,4-DMP	71	95	97	100
4-*tert*-Buthylphenol	83	88	96	100
Anisol	78	91	94	98
Aniline	9	94	96	100
Benzylalcohol	10	92	98	99
Nitrobenzene	54	92	96	100
2,4-Dinitrofluorobenzene	44	83	96	98
o-Hydroxyacetophenone	88	85	95	96
Isopentyl benzoate	84	72	89	95
Diethyl phthalate	90	87	96	100

[a]The PS-DVB is Amberchrom CG-161.

Table 2

Recovery (%R) values obtained on-line preconcentrating different sample volumes with different sorbents (*n*=6).

Compound	Carbopack B		Bond Elut PPL			HYSphere-1		
	50 ml	100 ml	50 ml	100 ml	200ml	50 ml	100 ml	200 ml
Oxamyl	68	35	82	82	81	82	82	82
Methomyl	80	68	79	77	74	78	78	80
Ph	7	3	68	43	23	77	73	50
4-NP	57	38	86	86	86	84	83	83
2,4-DNP	24	27	85	87	89	84	86	86
Bentazone	64	54	82	82	83	86	82	83
Simazine	86	88	85	88	89	86	87	89
MCPA	82	102	83	86	86	79	87	86
Atrazine	82	83	81	82	82	83	83	84

In the last few years, several new highly crosslinked polymers, such as Envi-Chrom P [14,15,21,28,38], LiChrolut EN [11,21,28,30,39], Styrosorb [40], Isolute ENV [11,39] and HYSphere-1 [13], have become available. These sorbents have a higher degree of crosslinking, and so have an open structure (high-porosity material), which increases their specific surface area and allows greater π-π interactions between analytes and sorbent [41]. The breakthrough volumes will therefore be higher than those obtained when less crosslinked sorbents are used. This has been demonstrated by comparing these sorbents with sorbents such as GCB [13], Bond Elut PPL [13], PS-DVB (PLRP-S and Amberchrom) [11,14,15] and cyclohexyl bonded silica [38]. The recoveries obtained in the on-line SPE of different sample volumes of a mixture of pesticides and phenolic compounds with HYSphere-1 are included in Table 2, where they are compared with those obtained with a GCB (Carbopack B) and a functionalized PS-DVB (Bond Elut PPL). We can see that recoveries for phenol were higher with the

highly crosslinked sorbent. LiChrolut EN has also been compared with chemically modified polymeric resins with acetyl [14] and benzoyl [15] groups in the SPE of pesticides and phenolic compounds in tap and river water. Slightly higher recoveries were obtained when LiChrolut EN was used, mainly for the most polar analytes such as oxamyl, methomyl and phenol whereas for the rest of compounds the recoveries were similar. Some authors obtained better recoveries with Carbograph 4 and Carbograph 5 than with LiChrolut EN and Envi-Chrom P in the analysis of phenolic compounds and pesticides [28].

Using LiChrolut EN and Isolute ENV recoveries were better than with other sorbents, although these sorbents, in on-line SPE procedures, have a broader front because of their large specific surface which affords a high sorption power over a wide range of hydrophilic substances (humic and fulvic acids) present in real water samples and makes analyte elution difficult [11,30]. This band can make it difficult to determine the most polar compounds, but some methods to decrease it have been mentioned in SPE procedures section. When elution is carried out with the whole mobile phase, the band broadening is greater with LiChrolut EN than with PLRP-S, because of its higher sorptive power.

Other recently developed porous polymeric sorbents are Porapak RDX and Oasis HLB which are divinylbenzene-vinylpyrrolidone copolymers. Porapak RDX was used in the SPE of nitroaromatic and nitramine explosives and their degradation products in water, and recoveries were greater than 89% in the preconcentration of 500 ml of water spiked with 1 and 10 µg l^{-1} of each analyte [42]. This sorbent has also been used for the analysis of herbicides and their metabolites, pesticides, fungicides, phenolic compounds and carbonyl compounds, and recoveries were high even for the most polar compounds [43]. Porapak RDX was compared with LiChrolut EN and Isolute ENV for the preconcentration of a variety of phenolic compounds from water samples and breakthrough volumes were lower when Porapak RDX was used [39].

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Oasis HLB is a macroporous poly(divinylbenzene-co-*N*-vinylpyrrolidone) copolymer which is water wettable because the hydrophilic *N*-vinylpyrrolidone and the lipophilic divinylbenzene provide the reversed-phase retention necessary to retain analytes. Therefore, it is more flexible at processing samples since it can dry out during the extraction procedure without diminishing its ability to retain analytes. This is an important advantage that the previously mentioned sorbents do not have. Moreover, its pH stability and retention are greater than C_{18} bonded silica sorbents [31]. The structure of Oasis HLB is shown in Fig. 2.

The efficiency of the polymeric sorbents depends on various physico-chemical parameters such as particle size, surface area, pore diameter, pore volume, degree of crosslinking and particle size distribution [11]. Table 3 shows the physical characteristics of some polymeric sorbents. The highly porous materials have a higher real active surface available which allows higher π-π interactions and so higher adsorption capacity. The band broadening with highly porous sorbents is only slightly greater than with other sorbents because porosity minimizes band dispersion. Band broadening is also minimized when working with sorbents with a narrower particle size distribution [11]. Several papers have shown that the capacity of LiChrolut EN is higher than that of Envi-Chrom P in the analysis of polar compounds [14,15,28]. This is due to the fact that LiChrolut EN has a higher specific surface and a lower particle size (1200 m^2 g^{-1}, 40-120 µm) than Envi-Chrom

Table 3
Physical characteristics of some commercial polymeric sorbents

Sorbent	Supplier	Base material	Particle size (µm)	Surface area (m²/g)	Pore size (Å)	Pore volume (ml/g)
Amberlite XAD-2	Supelco	PS-DVB	20-60	300	90	0.65
PLRP-S 100A	PolymerLabs.	PS-DVB	20	500	100	a
Amberchrom GC-161m	TosoHaas	PS-DVB	50-100	900	150	a
Bond Elut PPL	Varian	F-PS-DVB	125	700	150	1.0
Envi-Chrom P	Supelco	HC-PS-DVB	80-160	800-950	110-175	a
LiChrolut EN	Merck	HC-PS-DVB	40-120	1200	a	0.75
Isolute ENV+	IST	HC-PS-DVB	40-140	1000-1100	900	1.0
HYSphere-1	Spark Holland	HC-PS-DVB	5	> 1000	a	a
Styrosorb MN-150	Purolite Int.	HC-PS-DVB	300-800	1070	a	0.74
Styrosorb 2m	Purolite Int.	HC-PS-DVB	70-80	910	10	0.2-0.3
Oasis HLB	Waters	DVB-VP	31.4	831	82	1.4
Porapak RDX	Waters	DVB-VP	125-150	a	a	a

PS-DVB: polystyrene-divinylbenzene; F-PS-DVB: functionalized polystyrene-divinylbenzene; HC-PS-DVB: highly crosslinked polystyrene-divinylbenzene; DVB-VP: divinylbenzene-vinylpyrrolidone.
[a] Value not available.

P (800-950 m^2 g^{-1}, 80-160 μm) which allows better contact between the analytes and the sorbent. This in turn increases analyte retention.

Immunosorbents

The main problem encountered with SPE is the lack of selective sorbents when analyzing surface water. In this sense, the matrix of these samples is difficult to eliminate and it produces a noisy baseline and a large peak at the beginning of the chromatogram, thus making the determination of the most polar analytes laborious.

Recently, new extraction sorbents involving reversible and selective antigen-antibody interactions, called immunosorbents (ISs), have been synthesized in order to trap structurally related pollutants. They give extracts which are free of interfering substances. Using ISs, extraction, trace enrichment and cleanup are accomplished in one step when surface waters are analyzed. This selective preconcentration should not require an additional cleanup step [44,45].

Immunosorbents are obtained by covalently binding antibodies onto appropriate sorbents. The ideal support for an immunoaffinity sorbent is rigid and porous so that the flow rates of environmental samples can be high. Likewise, it should provide functional groups to enable appropriate coupling with a sufficient number of antibodies and be hydrophilic to prevent nonspecific interactions with the analytes and the sample matrix [46,47]. At present, diol or aldehyde activated silica supports are the best sorbents, meeting all these requirements. In both cases, the antibody is coupled via amino groups using Schiff base methods. Ion attraction, hydrogen bonds, hydrophobic attraction and van der Waals forces are involved in the antigen-antibody interactions. In addition, when the shapes of the antibody receptors and the antigen are complementary, forces of steric repulsion contribute to the overall strength of the interaction. So, the formation of the

antibody-antigen complex can be affected by the chemical composition of the sample [47]. The capacity of immunosorbents can be increased by a better selection of the silica used for the bonding procedure [46], and also depends on the orientation and the purity of the immobilized antibodies [44].

Nevertheless, the problem of the crossreactivity of the antibody is still a difficult one when quantitative results are needed. Taking advantage of the crossreactivity of a specific antibody it is possible to extract many compounds of the same family [44,48].

These sorbents have been applied in the analysis of triazines and phenylurea herbicides in different waters [44-50], using a single antibody (anti-isoproturon, anti-chlortoluron, anti-atrazine and anti-simazine) or a multi-antibody (mixed antibodies). They have been compared with C_{18} [49], PRP-1 [50] and PLRP-S [46] in the SPE of these analytes from enviromental samples. The high selectivity of the immunosorbents gave chromatograms that were cleaner than those obtained with the other sorbents.

Immunosorbents have been applied to the determination of several pesticides in the off-line [44,48,49] and on-line [45,46,50] SPE. Ferrer et al. [45] demonstrated that the coupling of ISs with LC-APCI-MS detection is a powerful technique for the determination and quantitation of polar pesticides in environmental matrices at the low ppt level without the need of additional cleanup steps. By using a small sample volume, very low detection limits can be reached due to the enhanced selectivity and high sensitivity.

Few immunosorbents are commercially available thus far, so the number of analytes which can be analyzed are limited. Moreover, it is well known that polyclonal antibodies can be produced and prepared in SPE cartridges and this takes approximately one year in research laboratories. The costs at this stage are high as compared to conventional systems. Only when industry is involved will the cost be reduced due to the production of larger amounts. Also, the half-

life of the columns depends on many parameters and can vary from 3-6 months to up to 2 years. On the other hand, industry has demonstrated interest in such columns and most probably they will be available at reasonable prices in the near future [45].

Molecularly Imprinted Polymers

An emerging technology, called molecular imprinting, leads to highly stable synthetic polymers that possess selective molecular recognition properties. Recognition sites within the polymer matrix are complementary to the analyte in the shape and positioning of functional groups. Molecularly imprinted polymers (MIPs) are made by synthesizing highly crosslinked polymers in the presence of a template molecule. After removal of this molecule, the polymer can be used as a selective binding medium for the template (analyte) or structurally related compounds. The mechanisms by which these polymers specifically bind the template and related ligands are attributed to the formation of functional groups in a specific arrangement within the polymer that corresponds to the template and to the presence of shape-selective cavities.

Some of these polymers have high selectivities and affinity constants, comparable with naturally occurring recognition systems such as monoclonal antibodies, which make them especially suitable as constituents in chemical (biomimetic) sensors for analytical chemistry or simply for enhancing the selectivity in SPE.

The imprinted materials compare favourably to immunsorbents mainly with respect to their ease of preparation, cost and time of preparation, reproducibility of preparation, sample load capacity, material durability and material composition. However, for their more general use in separations, the range of

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imprintable compounds needs to be extended and the problems related to site heterogeneity and mass transfer solved.

There are already a few applications of these polymers. Sellergren [51] has reviewed some of these applications and the properties of these sorbents.

CONCLUSIONS

In the last few years, SPE has replaced LLE because of the drawbacks of the latter. Sorbents in disks overcome some of the drawbacks of cartridges, but the limited number of commercially available sorbents in this format means that there are fewer applications than with cartridges. Nowadays, on-line trace enrichment is increasingly being used, but it has some drawbacks such as band broadening which can be avoided by eluting the analytes trapped in the precolumn only with the organic solvent of the mobile phase.

The classical bonded silicas are being displaced by polymeric sorbents because they cannot effectively trap the most polar compounds. Currently, chemically modified polymeric resins with different functional groups (e.g. acetyl, hydroxymethyl, benzoyl and *o*-carboxybenzoyl) and highly crosslinked polymeric sorbents are being developed. They have a higher sorption capacity for the most polar compounds. These sorbents are the most suitable materials for the SPE of polar analytes from environmental waters. The development of other functionalized polymers and highly crosslinked sorbents would appear to be the next objective in the synthesis of new materials for SPE procedures. New immunosorbents and molecularly imprinted polymers need to be developed in order to monitor a wider range of micropollutants in water samples by the selective SPE of specific groups of analytes.

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