



Cyclodextrin-based nanosponges for the dispersive-solid phase extraction of pesticides from environmental waters

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ABSTRACT

In this work, harmless, biodegradable, and low-cost cyclodextrin-based nanosponges (CDNS) were synthesized with an optimized green approach based on the use of a natural cyclodextrin (α -CD, β -CD) as the monomer, citric acid as the cross-linker, and NaH_2PO_4 as the catalyst. The resulting materials were studied using thermal analysis (TGA, DSC), IR spectroscopy, and SEM imaging. For the first time, α -CDNS and β -CDNS were successfully applied for the dispersive-solid phase extraction (d-SPE) from surface waters of eleven pesticides, including achiral compounds (ametrocradin, methoxyfenozide, tebufenozide, pyraclostrobin, spiromesifen), optical stereoisomers (mandipropamid, penconazole, difenoconazole, pyriproxyfen, hexythiazox) and geometric stereoisomers (dimetomorph). The extracts were analysed by high performance liquid chromatography-tandem mass spectrometry (HPLC-MS/MS). A cholesteryl-bonded silica column allowed the baseline separation of the geometric stereoisomers of dimetomorph, while a polysaccharide-based column was effective in the separation of the optical stereoisomers. Although no nanosponge displayed stereo-discrimination, all of them provided absolute recoveries depending on the analyte logP, steric hindrance, and spike level. In particular, β -CDNS gave yields between 26 and 93 %, and intra-day and inter-day precision of 2–9 % and 4–18 %. Limits of detection and limits of quantitation were 0.05–40 ng/L and 0.2–120 ng/L, respectively. After validation, the d-SPE-HPLC-MS/MS method was applied to the analysis of water samples from the Tiber River: ametrocradin, mandipropamid, penconazole, pyraclostrobin and pyriproxyfen were detected at concentrations between 16 and 43 ng/L. Last but not least, recycling tests have proved that CDNSs can be reused three times with unvaried extraction yields.

1. Introduction

Analytical methodology development often implies the use of harmful chemicals and the production of toxic wastes [1]. Therefore, since the introduction of the twelve principles of Green Analytical Chemistry (GAC) in 2013 [2] and more recently of ten rules of Green Sample Preparation (GSP) rules [3], efforts of researchers are increasingly directed towards the development of overall greener analytical methodologies. In particular, novel green analytical strategies focus on 2 main goals [4]: i) the miniaturization of extraction procedures to reduce wastes, time and energy consumption [5]; ii) the development of highly efficient extraction materials synthesized by using safe and sustainable precursors and methods, as recommended by the second and third GSP rules. The first goal is limited by the complexity of the matrices and the low concentration of target analytes, so that solid-phase extraction (SPE)

still plays a fundamental role in the clean-up of liquid samples such as environmental waters [6] and biological fluids (milk, blood, saliva, urine, etc.) [7,8]. The second goal is pursued through the development of natural-based sorbents, alternative to petroleum-derived polymeric materials (polystyrene-divinylbenzene, polydivinylbenzene-N-vinylpyrrolidone, etc.), to minimize the dependence on fossil reserves and to increase their potential for biodegradability [9]. Besides sustainable characteristics, including also multiple reuses, such state of the art sorbents should exhibit analogous analytical figures of merit as those achievable with conventional materials [5]. From this perspective, cyclodextrins (CDs) represent an excellent starting point to synthesize natural-based sorbents. In fact, these cyclic oligosaccharides are obtained from starch, one of the cheapest (<1 € per kg) and most available biomasses after cellulose and chitin, whose world production is around 80 million tons per year [10]. A mixture of homologues, which include

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up to 13 glucose units, is the natural product of starch degradation catalysed by cyclodextrin-glucanotransferase, an enzyme produced by a large number of microorganisms (e.g. *Bacillus macerans*). CDs are composed of optically active D-glucopyranose units bound via α -(1,4)-glycosidic linkages: six units in α -CD, seven units in β -CD, eight units in γ -CD, and so forth. In addition, many other CDs, differing in both ring size and functional groups, have also been obtained synthetically [11]. What makes CDs interesting is their capability to form “host-guest” complexes due to their toroidal shape, characterized by an external hydrophilic surface and a hydrophobic cavity reacting as a Lewis base. Such “microheterogeneous” environment allows the formation of inclusion complexes with organic molecules with stoichiometry 1:1, 1:2 and 2:1 [12]. CDs are soluble in water and this feature determines a technological problem, which is their recovery downstream of a SPE procedure. In order to overcome this issue, CDs can be polymerized through a cross-linking reaction producing a material which is insoluble in water [13]. Currently, only few examples of SPE applications with β -CDNS have been reported [14], all synthesized using Brown Chemistry processes. For instance, Appell et al. [15] extracted ochratoxin A from grape juice and wine using β -CDNS, cross-linked with methylene bis-diphenyl diisocyanate. Bhaskar et al. [16] prepared a β -CD-polyurethane polymer using hexamethylene diisocyanate in dry dimethylformamide and applied it for the SPE of carcinogenic aromatic amines from water samples. Zhang et al. [17] employed tetrafluoroterephthalonitrile as the cross-linker and K_2CO_3 as the catalyst to prepare a mesoporous material to recover quinolones from wastewater samples. Thus far, CDNSs obtained by using citric acid as the crosslinker [18,19] have been designed for water remediation [18,20] and for *in vitro-in vivo* studies to remove organic toxic molecules from gastrointestinal fluid [19].

The objective of the present work was to develop sorbents responding to both the requirements of sustainability and high analytical standards, especially in terms of recovery and precision. To this end, α -CD and β -CD were employed as monomers, citric acid as the crosslinker, and NaH_2PO_4 as the catalyst. All CDNSs were characterized by Attenuated Total Reflection Fourier-Transform Infrared spectroscopy (ATR-FTIR) spectroscopy, Thermogravimetric Analysis (TGA), Differential Scanning Calorimetry (DSC), and Scanning Electron Microscopy (SEM). For the first time, their feasibility was assessed in real applications for the dispersive-SPE (d-SPE) of pesticides from environmental waters, comparing the recovery performance of α -CDNS and β -CDNS. Since it is known that CDs are used as chiral selectors in chromatography, tests geared to recovering pesticides possessing optical and geometric isomerism were performed. Finally, both CDNSs were submitted to a set of experiments with the aim of assessing the recyclability of the materials.

2. Materials and methods

2.1. Chemicals and materials

α -Cyclodextrin (α -CD), β -cyclodextrin (β -CD), monohydrate citric acid, monosodium phosphate (NaH_2PO_4), ametocradin, methoxyfenozide, tebufenozide, pyraclostrobin, spiromesifen, mandipropamid (racemic mixture), penconazole (racemic mixture), difenoconazole (racemic mixture), pyriproxyfen (racemic mixture), hexythiazox (racemic mixture), dimetomorph (mixture of geometric isomers), all with a purity greater than 95 %, were purchased from Merck Life Science S.r.l. (Milan, Italy).

RS grade (elevated purity grade) solvents (methanol, acetonitrile, ethyl acetate, formic acid) were obtained from the same supplier. Ultrapure water was produced with a Milli-Q Plus apparatus (Millipore, Bedford, MA, U.S.A.).

2.2. Synthesis of cyclodextrin-based nanosponges

For the synthesis of the different nanosponges, 0.95 g (4.94 mmol) of

monohydrate citric acid, 0.95 g of β -CD or 0.82 g of α -CD (0.84 mmol), and 0.6 g of catalyst (corresponding to 5.0 mmol of NaH_2PO_4) were introduced in a flat bottom glass container. Bidistilled water (3 mL) was added to cover the powder, shaking for some minutes; a lactescent suspension was obtained for β -CD due to its limited solubility in water (18.8 g/L), while a clear solution was attained for α -CD due to its greater solubility in water (145.0 g/L). After that, the samples were frozen and then lyophilized overnight (Lyovapor™ L-200, BUCHI). The fully dried samples were transferred to an electric muffle, kept at 160 °C, where the polymerization occurred for 2 h. After the cross-linking reaction, the resulting samples took an expanded globular form (like a meringue) and a brownish colour (Fig. 1).

The sample was finely ground, weighted (mass m_0), and then transferred into a centrifuge tube and washed with four 10-mL fractions of water to remove catalyst residues and excess reactants. Other washings were carried out with methanol (10 mL \times 2) and ethyl acetate (10 mL \times 2) to remove thermal degradation products of citric acid. The cleaned powder was transferred into a Petri dish, heated for 2 h in an electric oven at 50 °C to complete dryness, and finally weighed (mass m_1).

2.3. Instrumental apparatus

2.3.1. Scanning electron microscopy

The morphology and structure of CDNSs were evaluated by scanning electron microscopy (SEM). The acquired images were obtained thanks to an HR-FESEM (High Resolution Field Emission Scanning Electronic Microscopy) platform, precisely the AURIGA ZEISS model, having a resolution of 1 nm, complete with EDS (Energy Dispersive X-Ray Spectroscopy) Bruker, EBL (Electron-Beam Lithography) Reith, FIB (Focus Ion Beam). In accordance with the functioning of the FESEM, the prepared samples, fixed on a stub by means of a graphite-based double-sided conductive tape, were metallized using chromium as the metal. Since the CDNSs are insulating, this treatment allows the increase of conductivity and mechanical stability of CDNS samples.

2.3.2. Fourier-transform infrared spectroscopy

Vibrational spectra of CDs, citric acid and CDNSs were recorded by using a Nicolet 6700 spectrometer (Thermo Scientific, Waltham, USA). The spectra were obtained in attenuated total reflection, through the use of a single reflection ATR accessory, i.e. the Golden Gate Single Reflection ATR System model, characterized by a synthetic diamond having an angle of incidence of 45°. The measurements were carried out on a spectral region between 4000 cm^{-1} and 650 cm^{-1} , with a resolution of 4 cm^{-1} and with a number of accumulations equal to 200 for each spectrum.

2.3.3. Thermogravimetric analysis

Thermogravimetric measurements were carried out to confirm cross-linking and to obtain information regarding the thermal stability of the material under investigation. The instrument was a Mettler TG 50 thermobalance equipped with a TC 10 A processor. About 7 mg of a CDNS sample was weighed and placed in a special ceramic pot. The ceramic pot was subsequently placed in the furnace, under a constant nitrogen flow of about 20 mL/min, setting a heating ramp from 25 °C to 500 °C, with a heating rate of 10 °C/min.

2.3.4. Differential scanning calorimetry

The DSC analyses were performed by means of a Mettler TA 3000 calorimeter (Mettler Toledo, Greifensee, Switzerland), equipped with a TC 10 A processor and a DSC 30 measuring cell. For each measurement, about 7–8 mg of sample was weighted in a 40 μ L aluminium pan and sealed. The analyses were recorded in two different ways, i.e. under a constant flow of nitrogen at 20 mL/min or under a constant flow of compressed air 20 mL/min. The temperature protocol applied comprises a first heating from 30 °C to 200 °C, with a heating rate of 30 °C/min, a cooling step up to 25 °C at 30 °C/min, and, lastly, a second heating from

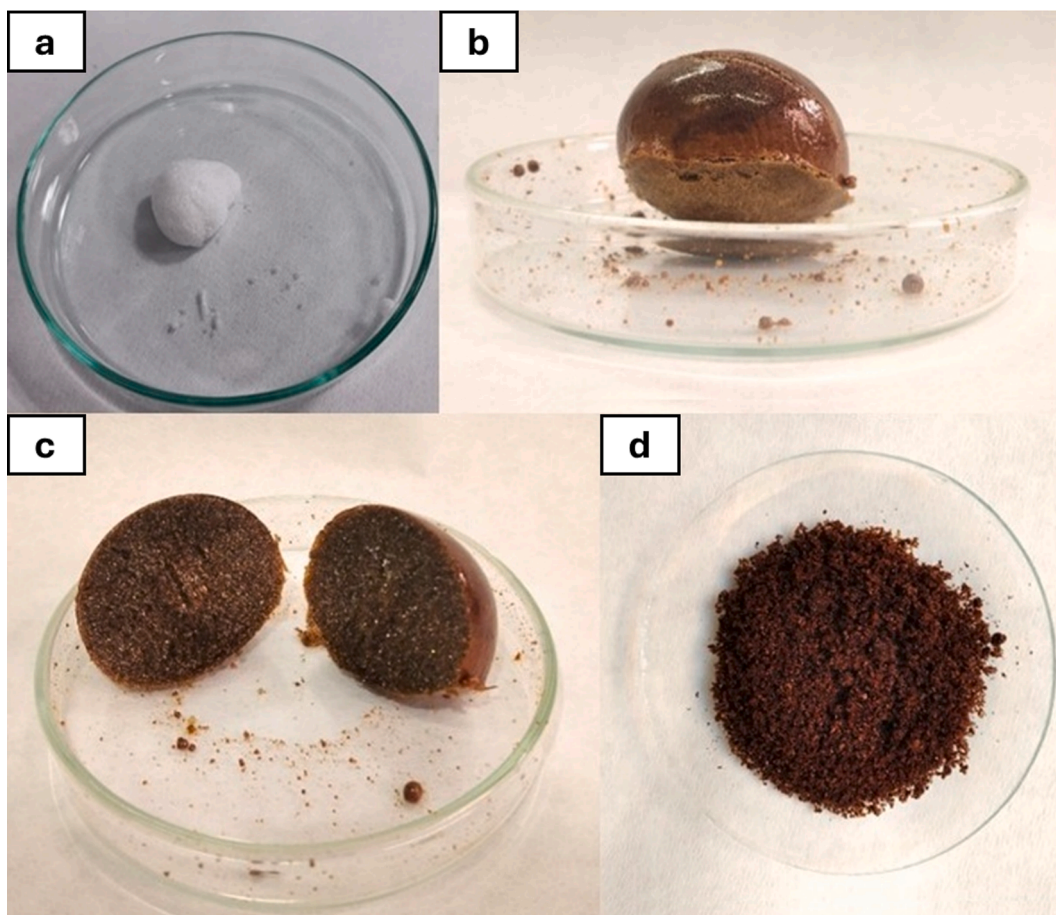


Fig. 1. A sample of α -cyclodextrin-based nanosponge: (a) pre-crosslinked polymer; (b) crosslinked polymer with the typical globular shape; (c) the section of the crosslinked polymer shows the characteristic sponge morphology; (d) the nanosponge in powder form.

25 °C to 350 °C at a rate of 10 °C/min.

2.4. Standard solutions

Individual stock solutions of the pesticides were prepared at 1 mg/mL by dissolving weighted amounts of the analytical standards (Ohaus DV215CD Discovery semi-micro and analytical balance, 81/210 g capacity, 0.01/0.1 mg readability) in appropriate solvents: ethyl acetate was used for mandipropamid, pyriproxyfen and spiromesifen, and methanol for the other pesticides.

Working composite standard solutions of pesticides were obtained by diluting aliquots of the individual stock solutions with methanol at concentrations depending on the purpose. All standards and solutions were stored at -18 °C.

2.5. River water samples

Water samples were grabbed from Tiber River at three different points: Oasis of Farfa and Fiumicino. The samples were collected in glass bottles and kept at 4 °C in the dark. Before processing samples, suspended particulate matter was removed by filtration with a 1.5- μ m pore size Whatman GF/C glass fibre pad (Maidstone, U.K.).

2.6. Extraction of pesticides from river water samples

A 500 mg aliquot of CDNS was dispersed into a 50-mL aqueous sample; then, the suspension was left under magnetic stirring for 15 min to favour the analyte adsorption. After centrifugation at 6000 rpm per 5 min, the supernatant was discharged, while the sorbent was treated with

9 mL of ethyl acetate, by vortexing for 5 min at 1600 rpm to promote the analyte desorption. After centrifugation, the extract was evaporated to dryness at 45 °C; the residue was then dissolved in 200 μ L of a water: acetonitrile (50:50, v/v) solution. The extract was transferred into a 1.5-mL Eppendorf and centrifuged for 5 min at 13000 rpm to remove any trace of CDNS. Finally, 5 μ L of the extract was injected into the UPLC-MS/MS system.

2.7. High-performance liquid chromatography-tandem mass spectrometry

The chromatographic system consisted of ACQUITY UPLC H-Class PLUS instrumentation (Waters, Milford, MA, USA). The detection was performed by a triple quadrupole mass spectrometer, API 4000 model by AB SCIEX (Foster City, CA, USA) equipped with an ESI source, operating in dual polarity at 350 °C (capillary voltage was -4500 V and $+5000$ V). High purity nitrogen was used as curtain and collision gas, while air as nebulizer and drying gas. The full width at half maximum (FWHM) was set at 0.7 ± 0.1 m/z in each mass-resolving quadrupole to operate with a unit resolution. All chromatograms were acquired in Multiple Reaction Monitoring (MRM), selecting two MRM transitions for each analyte.

The eleven pesticides were all separated under reversed phase conditions on a Cosmosil Cholesterol column (4.6×250 mm, 5 μ m) (Nacalai Tesque Inc, Japan), by using water and acetonitrile, both 5 mM in formic acid, as the mobile phase. The flow rate was 1 mL/min and $\frac{1}{4}$ was diverted into the ESI source. The elution gradient was as follows: after an equilibration time of 5 min at 50:50 (v/v), phase B was linearly increased to 70 % in 10 min and then, to 85 % in 0.1 min; finally, it was raised to 90 % until the end of the race (20 min total).

The separation of the five chiral pesticides (mandipropamid,

penconazole, pyriproxyfen, hexythiazox, difenoconazole) was achieved on the column Lux Cellulose-2 (4.6 × 250 mm; 5 μm), kept at 20 °C, under isocratic conditions (65/35 ACN/H₂O (%v/v) 0.1 % formic acid. The flow rate was 0.8 mL/min and ¼ was diverted into the ESI source [21].

Data processing was managed by Analyst®1.6 Software (AB Sciex, Foster City, CA, USA). All the HPLC–MS/MS parameters useful for the analysis of the target pesticides are reported in Table S1. Fig. S1 and Fig. S2 are representative chromatograms obtained on the Cosmosil Cholesterol column and on the chiral cellulose-based column respectively.

2.8. Method validation

The d-SPE-HPLC-MS/MS was validated in terms of recovery, enrichment factor (EF), precision (intra-day and inter-day), sensitivity, linearity, limit of detection (LOD), and limit of quantitation (LOQ). All validation experiments were performed in matrix, by using 50-mL aliquots of river water taken from Oasis of Farfa, an unspoiled naturalistic oasis in the north of Rome.

Five replicates, prepared at three different concentration levels (0.05, 0.5 and 5 μg/L), were analysed to evaluate recovery and precision for each analyte, both within an analytical session (intra-day precision) and two different analytical sessions performed over a period of fifteen days (inter-day precision). Recoveries were calculated applying the area method (see section 3.6 for the details), while precision was expressed in terms of relative standard deviation (RSD%). For each analyte, the EF was calculated as follows:

$$EF = \frac{C_{final\ extract}}{C_{sample}} = R \frac{V_{sample}}{V_{final\ extract}}$$

with R = recovered moles.

Matrix-matched calibration curves were built by spiking eight 50-mL aliquots of blank river water pre-extraction with increasing concentrations of the analytes (see section 3.9 for the details). Sensitivity was deduced by the curve slope.

For each pesticide, LOD and LOQ were evaluated as the analyte concentration in a river water sample able to provide a signal-to-noise of 3 and 10, respectively. Upper limit of quantitation (UPLQ) was set as the spike level of a river water sample for which the percentage difference between $y_{measured}$ and $y_{linear\ regression}$ was 3 %.

Carry-over was evaluated by injecting methanol after having injected the calibrator at ULOQ.

3. Results and discussion

3.1. Optimization of the CDNS synthesis

In this work, starting from the synthesis conditions proposed by Bednarz et al. [22], some modifications were introduced following a systematic investigation to optimize key parameters, including reaction time, temperature, type of catalyst, and the catalyst-to-CD molar ratio. Bednarz and his co-workers managed to synthesize poly(β-cyclodextrin-co-citric acid)s, by heating a mixture of citric acid, β-CD, and Na₂HPO₄ (in a 6:1:2 M ratio) at 160, 170, and 180 °C for 10 and 20 min. The main purpose of their investigation was to unravel the mechanism of polymerization and to understand the adsorption capability of the synthesized material. The amount of the insoluble polymer fraction they obtained ranged between 13 % (10 min at 160 °C) and 69 % (20 min at 170 °C). Testing the adsorption of methyl orange from aqueous solutions, they found that only the 40 % of CDs were able to form inclusion complexes, explaining the result as a consequence of a limited accessibility of the CD cavity in the polyester.

Under Bednarz's conditions, citric acid has the double role of reaction solvent and crosslinker (direct melt copolycondensation). In this work, considering that the melting point of citric acid is 153 °C and that

it starts decomposing at 177 °C [23], the reaction temperature was set at 160 °C to keep it liquid while minimizing its decomposition. As far as the catalyst is concerned, since previous studies on cellulose and 1,2,3,4-butanetetracarboxylic acid (BTCA) have shown that the acid anion of the catalyst plays a role in the esterification by assisting the removal of protons on intermediates [24], we preferred using NaH₂PO₄ instead of Na₂HPO₄ due to the greater acidity of the acid anion. Moreover, we increased the amount of catalyst by mixing citric acid, CD and NaH₂PO₄ in a molar ratio 6:1:6 instead of 6:1:2. Finally, we extended the reaction time to 2 h to obtain a better crosslinked product. In the several steps of the optimization, the insoluble fraction was evaluated by using the following formula:

$$\% \text{ of insoluble fraction} = \frac{m_1}{m_0} \times 100$$

with m_1 = weight (g) of the washed and dried CDNS and m_0 = weight (g) of the CDNS obtained after the crosslinking in the oven (see section 2.2).

Under the optimized conditions, the average insoluble fraction was 69 % for α-CDNS, and 71 % for β-CDNS.

When the lyophilization step was omitted to try to obtain a more sustainable synthesis procedure (see section 2.2), the insoluble fraction for β-CDNS dropped to 56 %. Moreover, recovery tests of pesticides from Milli-Q water provided recoveries similar to those of the corresponding lyophilized CDNS, but with a greater irreproducibility (RSD values up to 17 %).

3.2. FE-SEM analysis

The SEM images of the prepared CDNSs (Fig. 2a-f) were obtained at different magnifications. The morphological analysis of the nanosponges (Fig. 2a-d) confirms the presence of a heterogeneous macrostructure, resembling a sponge due to the presence of several macropores. These voids in the material are generated by the the CO₂ released during the CDNS heat treatment in the muffle (see section 2.2), which triggers the decarboxylation of citric acid to form itaconic acid (see Fig. S3) [22]. Such macropores disappear when the material is grounded. SEM images did not detect mesopores and micropores but lamellar structures that are clearly observable in Fig. 2f.

Through SEM analysis, we were also able to confirm the effectiveness of the washing treatments: in fact, residues of the catalyst and reaction byproducts are evident on the unwashed CDNS in Fig. 2e, while they disappear after the washing step (Fig. 2f).

3.3. ATR-FTIR characterization

The ATR-FTIR analyses were performed to verify the occurred cross-linking of the synthesized CDNSs, collecting and comparing the spectra of: i) blanks (α-CD, β-CD, and citric acid); ii) crosslinked and washed nanosponges.

As regards the spectra of the blanks (Fig. 3a), it is possible to observe the O–C–O stretching around 1160 cm⁻¹ and the asymmetric –CH₂ stretching at 2900 cm⁻¹ for both types of CDs, while the C–OH stretching, and the O–H stretching were at 1030 cm⁻¹ and 3450 cm⁻¹ for both CDs and citric acid. These absorptions are also characteristic of the respective CDNS spectra (Fig. 3b). As far as the citric acid is concerned, two intense peaks are detected at 1743 cm⁻¹ and 1689 cm⁻¹, ascribable to the stretching of the C=O groups, free (monomer) or involved in intermolecular H bonds (dimer).

To obtain information regarding the occurred cross-linking, the spectra of pre- and post-crosslinked CDNSs were compared; Fig. 3b shows this comparison for α-CDNS. The formation of the ester function, between the carboxylic group of citric acid and the hydroxyl group of CD, is confirmed by the shift of the C=O peak from 1700 cm⁻¹ for pre-crosslinked CDNS (due to the C=O of citric acid), to 1721 cm⁻¹ for cross-linked CDNS (due to the C=O of the ester group). Moreover, the

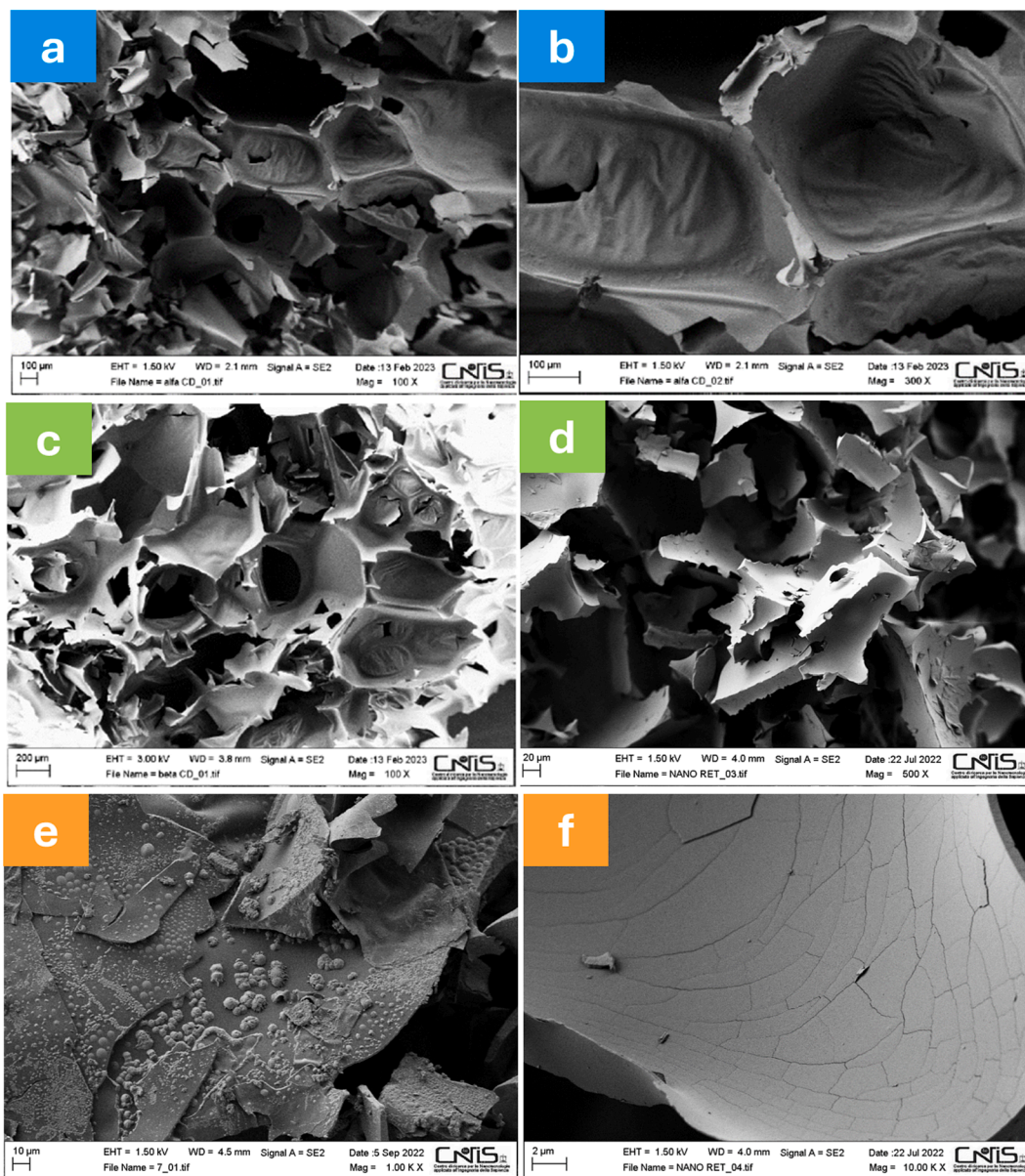


Fig. 2. SEM images of the synthesized CDNSs. (a) α -CDNS at 100 \times magnification; (b) α -CDNS at 300 \times magnification; (c) β -CDNS at 100 \times magnification; (d) β -CDNS at 500 \times magnification, (e) β -CDNS before washing and (f) β -CDNS after washing to remove the residues of catalyst and decomposition products of citric acid.

shoulder around 1630 cm^{-1} can be assigned to the C=C stretching of an unsaturated carboxylic acid, namely *cis*-aconitic acid, which is a by-product of the thermal degradation of citric acid and which, unlike its *trans* isomer, has the geometry suitable to form two cyclic anhydrides and to act as a crosslinking agent together with citric acid [22].

3.4. Thermal analysis

The thermal stability of the CDNSs was studied by TGA and DSC. Moreover, the thermograms of pre-crosslinked and crosslinked CDNS (α -CDNS, β -CDNS) were compared to obtain further evidence of the occurred crosslinking. For both crosslinked CDNSs, two significant weight losses were noted. The first one occurs at 100 $^{\circ}\text{C}$, and concerns their dehydration, while their degradation is recorded for temperatures slightly below 300 $^{\circ}\text{C}$. As a representative example, Fig. S4 shows the TGA curves (a,b) and their first derivative signals (DTGA) (c,d) of α -CDNS and β -CDNS.

As regards the analyses conducted on the pre-crosslinked CDNSs, two important weight losses can be observed at 170 $^{\circ}\text{C}$ and around 300 $^{\circ}\text{C}$,

corresponding to the degradation of citric acid and CDs respectively. These samples do not show water loss at 100 $^{\circ}\text{C}$ because all water was efficiently removed during the process of freeze-drying. It is also evident that such drying process does not activate the crosslinking. On the other hand, the crosslinked CDNSs exhibit a weight loss at 100 $^{\circ}\text{C}$ due to their dehydration, which proves their tendency to absorb water, and a significant weight loss around 300 $^{\circ}\text{C}$ (temperature of degradation of the material). The loss at 170 $^{\circ}\text{C}$, detected in the thermograms of the pre-crosslinked CDNSs, is no longer observable, indicating that citric acid has reacted with the CDs to produce crosslinked polymers.

DSC analyses performed on the CDNSs evidenced in the first heating cycle an endothermic band at ca. 100 $^{\circ}\text{C}$ related to water evaporation and no significant transitions in the second heating cycle (data not shown).

3.5. Optimization of chromatographic conditions

The chromatographic separation of the eleven pesticides was studied under reversed phase conditions, comparing the performance of a

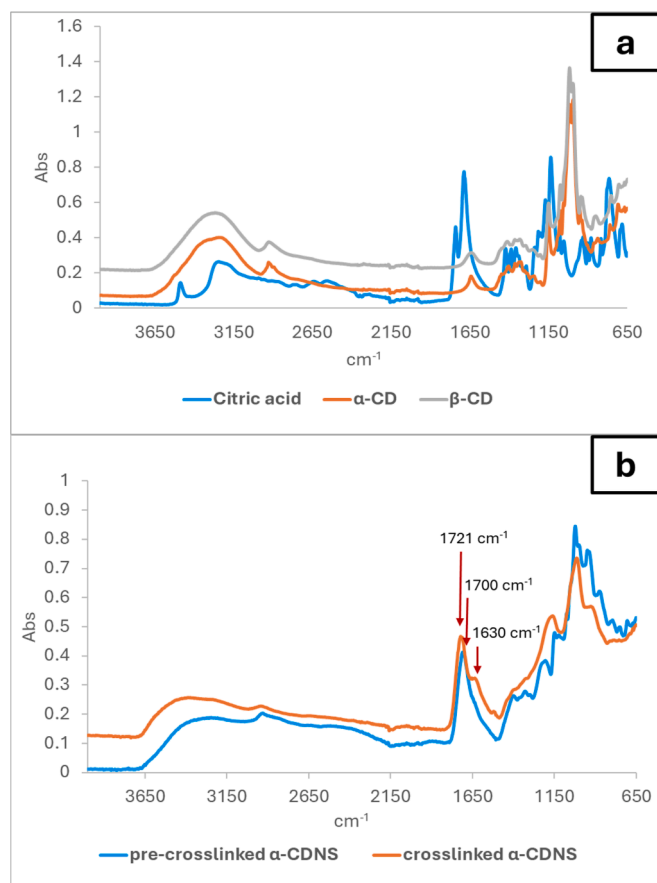


Fig. 3. ATR-FTIR spectra of (a) blanks and (b) pre-crosslinked α -CDNS and crosslinked α -CDNS.

conventional C18 column (X-Terra C18 column (2.1 \times 150 mm; 3.5 μ m), Waters, Milford, Massachusetts, USA) and a cholesteryl-bonded silica-based column (Cosmosil Cholesterol column (4.6 \times 250 mm, 5 μ m)).

By using water (phase A) and acetonitrile (phase B), both 5 mM in formic acid, the eleven analytes were baseline separated on the C18 column by applying the following gradient elution (flow rate = 0.2 mL/min): phase B was increased from 20 % to 60 % in 5 min and to 100 % in 15 min, then held at 100 % for 1 min [25]. The analytes were also completely separated on the Cholesterol column with a different gradient elution starting from a greater percentage of phase B due to its slightly higher hydrophobicity. Even if the chromatographic run was about 5 min longer using the Cholesterol column, its separation capability towards the geometric isomers of dimethomorph was greater despite the larger size of the packing microparticles. In fact, while the calculated resolution for the two stereoisomers was $R_{C18} = 1.0$, it was improved by using the Cholesterol column ($R_{Cholesterol} = 1.5$) allowing their more accurate quantitation. Fig. S5 shows the different performance of the two columns in separating the dimethomorph stereoisomers. Owing to its stronger selectivity, the Cholesterol column was chosen to carry out the separation of the eleven pesticides involved in this study.

To verify the potential of nanosponges in the chiral discrimination of the stereoisomers of the five chiral pesticides (mandipropamid, penconazole, pyriproxyfen, hexythiazox, difenoconazole), their baseline separation was achieved on a Lux Cellulose-2 column (cellulose tris(3-chloro-4-methylphenylcarbamate as the chiral selector) by applying the chromatographic conditions carefully optimized in a previous work of ours (Fig. S2) [21].

3.6. Efficiency of the SPE modes and type of CDNS

With the aim of evaluating the extraction capability of the synthesized sorbent materials, SPE on cartridge and d-SPE tests were conducted with the eleven pesticides including achiral molecules, optical stereoisomers and geometric stereoisomers. Blank water samples were grabbed from Tiber River at the Oasis of Farfa. At the analysis moment, 50-mL aliquots were spiked at 5 μ g/L with a composite working solution of the selected pesticides and allowed to equilibrate under magnetic stirring for 15 min.

The first trials were performed by packing 500 mg of β -CDNS into a glass cartridge (ID 1 cm, 6 cc, Supelco Inc., Bellefonte, PA, USA) between two PTFE frits. The cartridge was fitted into a sidearm filtering flask and sequentially washed with methanol (5 mL) and milli-Q water (5 mL). Liquids were forced to pass through the cartridge by the aid of vacuum from a water pump. After loading the aqueous sample, the analytes were re-extracted from the SPE cartridge by passing through it 12 mL of ethyl acetate, at a flow rate of about 4 mL/min. The eluate, collected in a 1.4 cm i.d. glass vial with a conical bottom, was completely dried at 45 $^{\circ}$ C under a gentle stream of nitrogen. Then, the residue was reconstituted with 200 μ L of a water:acetonitrile 50:50, v/v, and 5 μ L of the final extract was injected into the HPLC-MS system. The analysis was performed in triplicate together with a control sample, i.e. a blank sample spiked post-extraction with the same nominal concentration of the analytes. For each analyte, recovery values were calculated through the comparison of chromatographic areas as follows:

$$R(\%) = \frac{\text{Area}_{\text{sample spiked pre-extraction}}}{\text{Area}_{\text{sample spiked post-extraction}}} \times 100$$

The same conditions were applied to perform the d-SPE experiments (in triplicate). To this end, 500 mg of β -CDNS were dispersed in a spiked aqueous sample and left under magnetic stirring for 15 min to permit the analyte adsorption. After centrifugation, the supernatant was removed, and the analytes were re-extracted with ethyl acetate (12 mL). Evaporation, reconstitution and analysis of each extract was carried out as above-described.

The comparison between the two SPE extraction modes is shown in Fig. 4a and clearly underlines the better performance of d-SPE both in terms of recoveries and RSD%: in fact, the SPE on cartridge exhibits lower recovery yields (36 % on average) and slightly higher RSD% values (up to 11 %), if compared with d-SPE performance (75 %; RSD% up to 6 %). The greater efficiency of the d-SPE mode can be explained through the better contact that both sample and solvent establish with the sorbent, favouring the adsorption and desorption of the analytes. Afterwards, the same extraction conditions were applied to evaluate the performance of α -CDNS. The results are shown in Fig. 4b. As can be seen, β -CDNS provides higher or similar yields as α -CDNS, which only in one case (spiromesifen) shows a better performance. Therefore, β -CDNS was used for further experiments.

3.7. Optimization of the d-SPE conditions using β -CDNS

The optimization of the d-SPE conditions to extract the pesticides from river water samples was performed by means of one-variable-at-a-time method (OVAT), also known as monothetic analysis. Also, for these experiments, 50-mL aliquots of blank water river samples were spiked at 5 μ g/L with the analytes. For each condition evaluated (amount of sorbent, adsorption time, type and volume of desorption solvent, desorption time), three replicates were performed.

The first series of experiments were carried out to establish the amount of sorbent (600 mg, 500 mg, 250 mg, 150 mg). The analyte adsorption was allowed for 15 min under magnetic stirring. After centrifugation at 6000 rpm per 5 min, the supernatant was discharged while the sorbent was washed with 12 mL of ethyl acetate for 10 min; then, the extract was evaporated to dryness, reconstituted with 200 μ L of

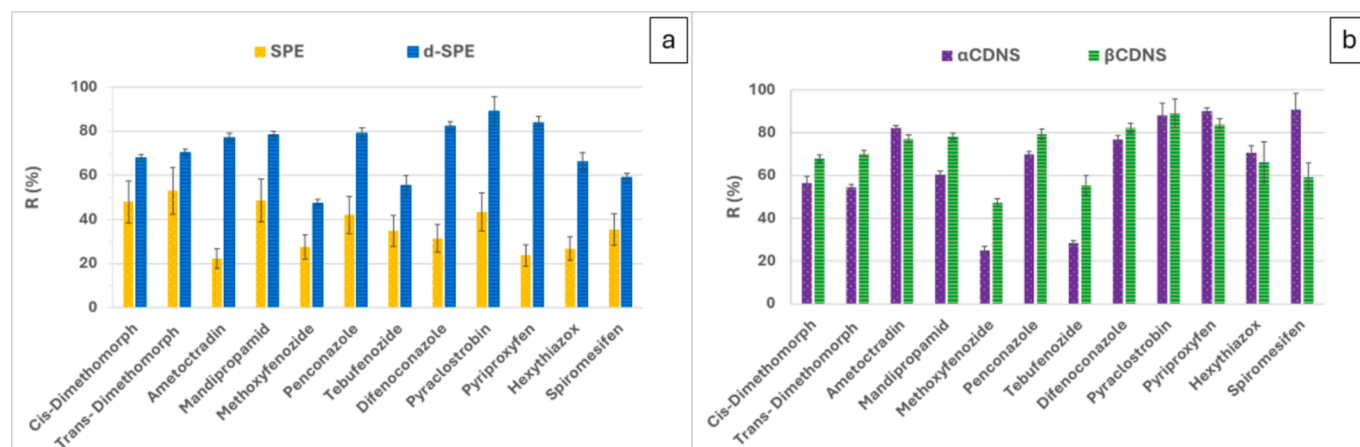


Fig. 4. (a) Comparison of the recovery yields obtained using β -CDNS as sorbent in the two different SPE extraction modes: SPE on cartridge (■) and d-SPE (■). (b) Comparison of the recovery yields obtained using α -CDNS (■) and β -CDNS (■) as sorbents working in d-SPE mode.

a water:acetonitrile (50:50, v/v) solution and 5 μ L was injected into the HPLC-MS system. The results, reported as absolute recovery averaged on all the analytes, are shown in Fig. S6. A gradual increase in recoveries from 48 % to 75 % was observed by passing from 150 to 500 mg of sorbent, while the yields obtained with 600 mg of β -CDNS was substantially unvaried (77 %); for this reason, 500 mg was the amount selected for the following experiments.

As far as the adsorption time is concerned, the analyte/sorbent contact was allowed to take place for 15, 30, 45 and 60 min under continuous magnetic stirring. The recovery values (average recovery of 75 %) indicated that 15 min was a contact time enough for the analyte adsorption since longer contact times were not able to increase the yields.

Regarding the optimization of type and volume of the desorption solvent, the chromatographic areas obtained by using four 3-mL fractions of methanol and ethyl acetate, i.e. organic solvents in which pesticides have good solubility, were compared (10 min of contact time per fraction): basically, no distinct difference was found between the two solvents. Moreover, it was observed that three fractions (9 mL in total) allowed one to reach the maximum extraction capability for all the analytes. Based on CHEM21 selection guide of “classical” and “less classical” solvents [26], both ethyl acetate and methanol are ranked as recommended solvents, but ethyl acetate has lower scores that make it greener (safety score: yellow for both solvents; health score: green_{ethyl acetate} vs yellow_{methanol}; environmental score: green_{ethyl acetate} vs yellow_{methanol}).

The last parameter to be optimised was the desorption time. To this end, 5, 10, 15, and 20 min were compared to verify their effectiveness on the analyte recoveries, by using a single 9-mL fraction of ethyl acetate. A contact time of 5 min was enough since longer desorption times do not provide greater yields.

Finally, a recovery study was performed with the aim of studying the CDNS performance in the presence of interfering compounds. To this end, five common environmental contaminants were selected: sulfamerazine, sulfamethoxazole, diclofenac, propoxur and 4-chloro 2-methylphenol. They all contain aromatic groups having good affinity with the hydrophobic cavity of the CDs. Among the eleven pesticides, pyriproxyfen was chosen to verify if its extraction yield from Milli-Q water was changed in presence of such interfering compounds. To this end, three 50-mL aliquots of Milli-Q water were spiked pre-extraction at 5 μ g/L with pyriproxyfen only, while other three aliquots were spiked pre-extraction at 5 μ g/L with both pyriproxyfen and the interferents. Within the experimental error, the average recovery of pyriproxyfen did not show differences (89 % vs 86 %), as expected given that the entire procedure was optimized in a real matrix, already containing many different interferents.

3.8. A study on stability and reuse of CDNS

Information on the shelf life of a product are important for its potential industrial applications. Although CDNSs are widely applied in fields such as drug delivery, catalysis, and pollutant removal, their stability under various environmental conditions remains a critical factor to ensure their effectiveness. In the present study, the stability of nanosponges in both anhydrous and controlled humidity environment was investigated.

The freshly synthesized material was divided in three 0.1-g aliquots: an aliquot was used for the immediate characterization via FTIR and TGA; an aliquot was stored in a desiccator at room temperature (anhydrous conditions), while the other aliquot was stored in a desiccator under controlled humidity, provided by a saturated NaNO₃ solution (water activity, $a_w = 0.74$). After one week of storage, the three aliquots revealed no significant changes in the IR spectra (Fig. S7a), with a lack of new absorbance peaks, and an overlap in the thermogravimetric curves (Fig. S7b).

During a long period of work (more than six months), we also had the opportunity to verify that a batch of synthesized nanosponges could be simply stored in a falcon at room temperature without variations in terms of extraction efficiency (for the figures of merit see section 3.9).

Finally, with the aim of studying the reuse of CDNS, a dedicated study was planned. To this end, after the first use of some aliquots of β -CDNS (500 mg) to extract the pesticides from water samples, their recyclability was assessed by applying two following washing cycles, with 3 \times 3 mL of methanol and 3 \times 3 mL of ethyl acetate, to regenerate the sorbent and allow its reuse. After three cycles of reuse, it was verified an unvaried mechanical stability of the sorbent, a similar recovery efficiency and the absence of memory effects. When the same tests were performed with the α -CDNS, the recoveries obtained on their third use indicated the occurrence of memory effect (not observed with the β -CDNS), probably due to the smaller size of the α -CD cavity (4.7–5.3 Å) [14]; for this reason, with this type of nanosponge, it is important to wash the sorbent carefully among the reuse cycles.

3.9. Figures of merit of the validated method

Once optimized, the extraction method using β -CDNS was validated and the corresponding figures of merit listed in Table 1 (recovery, precision, EF, LOD, LOQ) and Table 2 (linear dynamic range, linear regression parameters).

Recovery and precision were evaluated by spiking blank river samples at 50 ng/L, 500 ng/L and 5 μ g/L, concentrations that might be close to real levels of surface water contamination with pesticides. The results (five replicates per spike level) are reported in Table 1. Good recoveries

Table 1

Validation parameters of the d-SPE-UPLC-MS/MS method, developed to analyse the selected pesticides in river water samples.

Analyte	Recovery ^a (%)			Precision ^b						EF	LOD ^c	LOQ ^c
	Spike levels			Intra-day Spike levels			Inter-day Spike levels					
	50 ng/L	500 ng/L	5 µg/L	50 ng/L	500 ng/L	5 µg/L	50 ng/L	500 ng/L	5 µg/L			
<i>cis</i> -dimethomorph	n.e. ^d	50	72	n.e. ^d	7	2	n.e. ^d	10	7	125–180	30 ± 4	100 ± 20
<i>trans</i> -dimethomorph	40	49	74	8	6	2	12	9	9	100–185	3.0 ± 0.1	10 ± 1
ametoctradin	58	63	81	7	5	2	12	9	4	145–203	3.0 ± 0.2	10 ± 3
mandipropamid	65	70	82	5	2	2	7	7	5	162.5–205	0.20 ± 0.04	10 ± 1
methoxyfenozide	n.e. ^d	26	51	n.e. ^d	6	4	n.e. ^d	8	13	65–128	20 ± 1	70 ± 2
penconazole	56	68	83	7	5	3	10	10	7	140–208	1.0 ± 0.2	3.0 ± 0.3
tebufenozide	31	38	59	8	6	6	18	9	9	77.5–148	10 ± 1	20 ± 3
difenoconazole	68	73	86	10	7	2	13	11	3	170–215	3.0 ± 0.2	10 ± 2
pyraclostrobin	80	92	93	8	5	5	12	12	3	200–233	0.050 ± 0.001	0.20 ± 0.03
pyriproxyfen	79	86	88	5	2	3	10	7	3	197.5–220	1.0 ± 0.1	3.0 ± 0.1
hexythiazox	60	67	70	5	4	4	10	9	12	150–175	10 ± 1	40 ± 2
spiromesifen	n.e. ^d	60	63	n.e. ^d	9	4	n.e. ^d	12	16	150–158	40 ± 2	120 ± 30

^a Mean of five independent analyses.^b Relative standard deviation (RSD%) of five independent analyses performed within the same day or within two weeks (intra-day precision; inter-day precision).^c Mean and standard deviation of five independent analyses.^d n.e. = not evaluated because the spike level is less than LOQ.**Table 2**

Linear regression parameters of the d-SPE-UPLC-MS/MS method.

	Linear dynamic range (ng/L)	Equation		Determination coefficient (R ²)
		$y = ax + b'$		
		$a' \pm s_{a'} t_{(0.05;6)} (10^4)$	$b' \pm s_{b'} t_{(0.05;6)} (10^4)$	
<i>cis</i> -dimethomorph	100–20·10 ³	50.20 ± 0.25	−15.10 ± 0.89	0.998
<i>trans</i> -dimethomorph	10–20·10 ³	100.00 ± 0.19	19.50 ± 0.10	0.996
ametoctradin	10–20·10 ³	100.00 ± 0.48	51.90 ± 0.44	0.994
mandipropamid	10–12.5·10 ³	200.00 ± 0.21	400.00 ± 0.49	0.992
methoxyfenozide	100–20·10 ³	66.50 ± 0.10	8.302 ± 0.012	0.999
penconazole	3–20·10 ³	92.70 ± 0.10	100.00 ± 0.29	0.999
tebufenozide	10–20·10 ³	10.40 ± 0.10	−1.900 ± 0.010	0.997
difenoconazole	10–20·10 ³	200.00 ± 0.96	72.7 ± 1.8	0.996
pyraclostrobin	0.2–12.5·10 ³	200.00 ± 0.09	600.00 ± 0.17	0.996
pyriproxyfen	3–12.5·10 ³	700.00 ± 0.40	200.00 ± 0.23	0.998
hexythiazox	10–20·10 ³	100.00 ± 0.11	−18.8 ± 1.1	0.991
spiromesifen	100–20·10 ³	21.90 ± 0.22	6.80 ± 0.16	0.993

were achieved for all the analytes which, having logP values in the range 2.67–5.37, showed close affinity for the hydrophobic cavity of β-CD. Obviously, the molecule size and the presence of functional groups to establish hydrogen bonds with the OH groups of β-CD units are other crucial parameters. For example, pyriproxyfen has a diameter of 5.36 Å (data calculated by Biomodel software, free online <https://biomodel.uah.es/en/DIY/JSME/draw.en.htm>) that fits perfectly into the β-CD cavity, whose diameter is 6.0–6.5 Å [14]. However, the extended length of pyriproxyfen is 15.78 Å; this means that part of its molecule comes out of the β-CD cavity, whose height is 7.9 Å. Besides a simple stoichiometry 1:1, an inclusion complex 2:1 might be supposed even if its formation is difficult to obtain in a 3D network such as β-CDNS. Compared to the other analytes, the greater volume of methoxyfenozide and tebufenozide can explain their lower recovery (see Fig. S8). Regarding the discrimination capability of the sorbent towards geometric stereoisomers, the *cis*- and *trans*- forms of dimethomorph showed the same absolute recovery within the experimental error, proving that in this case β-CDNS did not favour a particular stereoisomer. Investigating the selectivity towards the optical stereoisomers of mandipropamid, penconazole, pyriproxyfen, hexythiazox, difenoconazole, no chiral discrimination was revealed since both enantiomers of each pesticide were recovered with the same yields.

For each analyte, EF was as a function of recovery at the different spike levels. The EF values ranged between 65 and 233, being

methoxyfenozide the least recovered pesticide (R = 26 % at 500 ng/L spike level), and pyraclostrobin the analyte which gave the highest recovery (R = 93 % at 5 µg/L spike level).

As reported in Table 1, the LOD values were in the pg/L-ng/L range, while the LOQ values were in the ng/L range, providing to be effective in the detection and quantification of environmental contamination levels. These values are comparable or lower than those reported in the literature (see section 3.9).

Eleven 50-mL aliquots of blank river water were spiked with increasing concentrations of the analytes (0.2, 3.0, 10, 50, 100, 750, 2·10³, 4·10³, 7.5·10³, 12.5·10³, 20·10³ ng/L), processed and analysed according to the protocols described in sections 2.6 and 2.7. For each analyte, the extension of the linear dynamic range was established depending on its LOQ and ULQ, and nine to ten of these calibrators were selected to build matrix-matched calibration curves (see Table 2). The linear regression analysis provided regression coefficients (R²) between 0.991 and 0.999, which was an excellent result considering the use of pre-extraction spiked calibrators.

3.10. Comparison with other methods from the literature

The main figures of merit of our d-SPE-LC-MS/MS method (EF, recovery, precision, LOQ and analysis time) were compared with those of other methods addressed to the analysis of some common pesticides

from surface water samples [27–34] (see Table 3). Our procedure exhibits comparable or significantly higher EFs, with LOQ values significantly lower compared with those of two mass-spectrometric methods [32,34]. Recoveries and precisions are difficult to compare since, in most methods, they were calculated at different spike levels; however, for some analytes such as pyraclostrobin and hexythiazox, the recoveries in this work are higher than those obtained with other methods at the same or even higher spike levels [27,28]. Moreover, recovery values above 100 % reported in some applications may suggest some matrix effect affecting the signal and, therefore, a not efficient sample clean-up. Considering common analytes (methoxyfenozide, penconazole, pyriproxyfen, hexythiazox), β -CDNS gave extraction recoveries similar to those obtained with a GO composite material [33], with the advantage of using a greener and safer sorbent, as well as a quicker procedure. Regarding extraction time, our procedure takes 10–20 min less than a typical SPE procedure on cartridge [27,28], while the dispersive

liquid–liquid microextraction (DLLME)-LC-MS/MS procedure [29] is undoubtedly the quickest, being rapidly an intrinsic characteristic of the DLLME technique. Regarding the sustainability of the procedure, CDNSs were prepared using reagents from renewable sources: as a matter of fact, CDs are obtained from the enzymatic biodegradation of starch, while citric acid, although is naturally occurring in citrus fruits, is produced at industrial scale via a sustainable process based on fermentation with *Aspergillus niger* [35]. However, also the sorbent used for the SPE-GC-triple quadrupole procedure [28] proposes a valid green alternative to conventional SPE sorbents, being the cork powder a by-product of the cork stopper production. Nevertheless, the study of the sustainable characteristics of our procedure also covers the reuse of the sorbent for more cycles of extraction (see section 3.8).

Table 3

Comparison of the main merit figures of some recent methods.

Method	Common analytes	EF	Recovery % (spike level, ng/L)	Intra-day precision % (spike level, ng/L)	Time per sample (min)	LOQ (ng/L)	Reference
SPE-HPLC-Q/Orbitrap (sorbent: OASIS-HLB)	difeconazole, dimethomorph, hexythiazox, mandipropamid, methoxyfenozide, penconazole, pyraclostrobin, spiromesifen, tebufenozide	62–172	31–105 (10–200)	7–14 (not specified; ultra-pure water)	> 60	0.5–25	[27]
SPE-GC-MS/MS (sorbent: cork powder)	dimethomorph, pyraclostrobin	16–17	80.1–87.2 (100)	7.8–9.1 (100)	<60	8.1–27	[28]
LTTM-DLLME-HPLC-triple quadrupole (extraction solvent: the LTTM choline chloride: acetylsalicylic acid 1:2 M ratio)	dimethomorph, hexythiazox, methoxyfenozide, penconazole	37–46	74–92 (60–100)	3–14 (0.006–0.1)	~15	10–100	[29]
SPE-UPLC-MS/MS (sorbent: silica-based mesoporous material)	dimethomorph, ametoctradin, mandipropamid, methoxyfenozide, penconazole, tebufenozide, difenoconazole, pyraclostrobin, pyriproxyfen, hexythiazox, spiromesifen	80–142	80–142 (0.1·10 ⁶ –0.5·10 ⁶)	6–28 (0.1·10 ⁶ –0.5·10 ⁶)	~15 (semi-automatic SPE)	–	[30]
Disk-SPE-LC-DAD (sorbent: MWCNTs)	tebufenozide	4000	117.3–108.5 (40–50)	7.0–8.6 (500)	–	20.7	[31]
QuEChERS-LC-MS/MS	methoxyfenozide, pyraclostrobin	–	72–98 (1·10 ³ –8·10 ³)	1–6 (1·10 ³ –8·10 ³)	~ 20	1·10 ³	[32]
rotating-disk SPE-HPLC-MS/MS (sorbent: chitosan–graphene oxide composite)	methoxyfenozide, penconazole, pyriproxyfen, hexythiazox	~ 130–220	~ 53–88 (5·10 ³)	–	~ 16.5 h including the analyte adsorption overnight	–	[33]
SPE-UPLC-MS/MS (sorbent: C18)	difenconazole, dimethomorph, pyraclostrobin, penconazole, tebufenozide, hexythiazox	7–9 (the eluate was diluted 10 times)	72.0–94.7 (10·10 ³)	1.7–6.8 (10·10 ³)	–	1.4·10 ³ –4.7·10 ³	[34]
d-SPE-HPLC-MS/MS (sorbent: β -CDNS)	dimethomorph, ametoctradin, mandipropamid, methoxyfenozide, penconazole, tebufenozide, difenoconazole, pyraclostrobin, pyriproxyfen, hexythiazox, spiromesifen	65–233	26–93 (50–5·10 ³)	0–19 (50–5·10 ³)	~ 45	0.2–120	[this work]

^aLTTM-DLLME: Low transition temperature mixture-dispersive liquid–liquid microextraction.

3.11. Analysis of real samples

The validated method was applied to the analysis of samples grabbed from the Tiber River in Fiumicino. The detected fungicides were mandipropamid, penconazole and pyraclostrobin, with comparable concentrations; all the others were undetected or detected under LOQ (see Fig. 5 and Table 4).

Cis- and *trans*- dimethomorph were found at different levels in two real samples which does not respect the area ratio in which the two geometric isomers occur in the pesticide formulation. From this analysis, it seems that biodegradation of *cis*-dimethomorph could be quicker than that of the *trans* isomer.

4. Conclusions

When compared with other chemical processes, Analytical Chemistry involves operations on “small scale”. However, the millions of analyses made each day all around the world in industry and research centres make it clear the impossibility to overlook the impact of analytical methods. In particular, the amount of chemicals, materials

Table 4

Pesticides levels in real samples from Fiumicino.

Analyte	Concentration ^a (ng/L)
<i>cis</i> -dimethomorph	n.d.
<i>trans</i> -dimethomorph	<LOQ
ametoctradin	16 ± 1
mandipropamid	43 ± 1
methoxyfenozide	n.d.
penconazole	43 ± 3
tebufenozide	n.d.
difenoconazole	<LOQ
pyraclostrobin	41 ± 2
pyriproxyfen	35 ± 1
hexythiazox	n.d.
spiromesifen	n.d.

^a Mean of three real samples.

and energy required for extraction, separation, detection and determination is both an environmental and economic problem. Therefore, the possibility of resorting to more sustainable analytical solutions is becoming the driving force of the research in this sector. In this work, we have proposed the optimization of a more sustainable and effective procedure for the preparation of CDNSs. The reagents (CDs and citric acid) are obtained from renewable sources. The polymerization reaction is realized “in mass”, i.e. without using solvents due to the double role of citric acid that acts both as a crosslinker and as a solvent. Compared to Bednarz’s protocol [20] that involves the use of Na₂HPO₄ as a catalyst and a maximum polymerization time of 20 min, our investigation has proved that NaH₂PO₄ is more effective and that, combined with 2-h polymerization time, allows one to obtain an insoluble fraction around 70 %, reducing wastes.

All prepared nanosponges were effective in recovering the selected pesticides from environmental waters, with a little better performance of β-CDNS (3–5 % more on average). Nevertheless, such natural CD-based polymers were not able to discriminate the stereoisomers of the chiral pesticides.

Since a range of synthetically derivatized CDs are commercially available, future perspectives will be aimed at obtaining more efficient and selective nanosponges to be used for advanced SPE applications on a wider range of target compounds contained in matrices of different complexity. In particular, our research group is currently studying the application of CDNSs for the extraction of emerging contaminants belonging to different chemical classes (such as UV filters, polychlorinated biphenyls, polycyclic aromatic hydrocarbons, drugs, antibacterial agents) from surface waters with very promising results.

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CRedit authorship contribution statement

Nina Felli: Validation, Methodology, Investigation. **Alberto Lorenzet:** Methodology, Investigation. **Valerio Di Lisio:** Investigation. **Valerio Bussetti:** Investigation. **Chiara Dal Bosco:** Methodology, Data curation. **Luisa Maria Migneco:** Writing – original draft, Supervision, Conceptualization. **Alessandra Gentili:** Writing – review & editing, Supervision, Project administration, Funding acquisition. **Iolanda Francolini:** Writing – review & editing, Supervision, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial

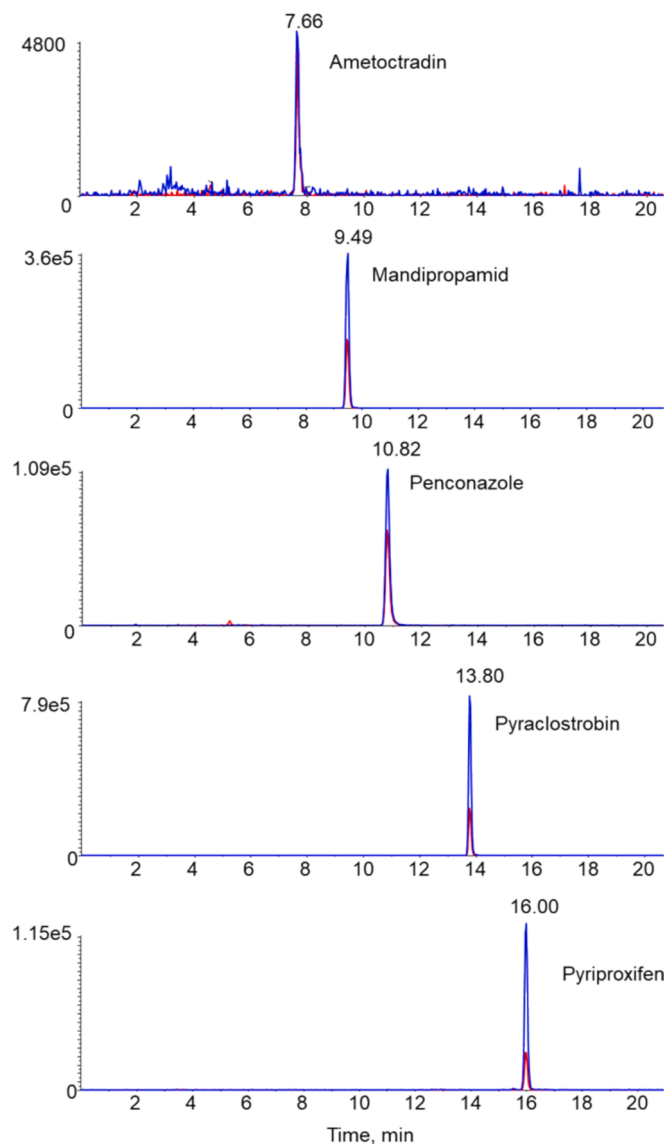


Fig. 5. Extracted Ion Currents (XICs) of a real sample from the Tiber River (Fiumicino). The analyses, performed in triplicate, provided the mean concentrations reported in Table 4.

interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.microc.2024.112135>.

Data availability

Data will be made available on request.

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