Simultaneous Determination of Phenolic Endocrine Disruptors Using 1-Butyl-3-Aminopropyl Imidazolium Tetrachloroferrate Functionalized Graphene Oxide GO-(Bapim)FeCl$_4$ as a Nanoadsorbent

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Abstract

This paper evaluated the removal efficiency of eight selected phenolic endocrine disrupting chemicals (bisphenol A (BPA), octylphenol (OP), nonylphenol (NP) and diethylstilbestrol (DES) Chlorophenol Red-β-D galactopyranoside (CPRG), 17β-estradiol (E2), estriol (E3), and 17 α-Ethinylestradiol (EE2)). from water samples with the use of magnetic ionic liquids (MILs) functionalized graphene oxide GO-MILs. The 8 phenolic EDCs were subjected to hydrolysis and then to solid phase microextraction with 1-butyl-3-aminopropyl imidazolium tetrachloroferrate functionalized graphene oxide GO-(Bapim)FeCl$_4$. The GO-MILs nanocomposite was packed into a standard filter to make homemade SPME column. Alkylphenols, chlorophenols, and bisphenol in GO-(Bapim)FeCl$_4$ nanocomposite were eluted with acetonitrile, and the eluate was concentrated under a nitrogen stream. The phenolic endocrine disrupting chemicals were then determined by high-resolution high performance liquid chromatography (HPLC). The recoveries for E3, E2, EE2 and CPRG were above 95% even to 100%, adsorption abilities for BPA, DES, OP, and NP are significantly weaker, with the removal efficiencies were between 80% to 87. The limits of quantitation (LOQ) for the 8 phenolic EDCs in the nanogram-per-milliliter range 0.025~1.000 ng/mL (S/N = 3) were thus achieved by using 1 mL of water, and the SIM responses were linear with the correlation coefficient varying by 0.9300~0.9943 and the highest enrichment factor was up to 250-fold. The results obtained are accurate and highly reproducible, making it a good alternative approach for routine analysis of phenolic endocrine disrupting chemicals in water samples.

Keywords: Ionic Liquid; Graphene Oxide; Endocrine Disruptor; Phenol; Microextraction

Introduction

One of the major class of pollutants that present in our environment is the endocrine-disrupting chemicals (EDCs), which normally damage normal physiological and hormone synthesis functions of male and female reproductive organs. Once in our body, EDCs usually bind to the receptors of steroid hormone such as the progesterone receptor (PR), oestrogen receptor (ER), and androgen receptor (AR). EDCs normally interfere with the actions of hormones, stimulation of cancer cell growth, induce abnormal reproduction, dysfunction of immune and neuronal system [1-3]. Most EDCs are able to produce abnormalities in mammalian, avian, fish, reptilian, and invertebrate species [4-10]. Recently, there is an increase concern about the impacts of human exposure to chemical compounds with endocrine-disrupting activity [11-18]. There is speculation that EDs can disrupt the endocrine system in different ways including: block the action

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of hormones, can act as "imposters" of naturally occurring hormones, can alter the chemical message sent by hormones, and can disrupt the production of hormones or hormone receptors. In fact, most EDs normally act as naturally occurring estrogens in the body. Moreover, many EDs block androgens, the male sex hormones such as testosterone. The idea of endocrine disruptors received credibility from a number of studies showing reproductive problems in wildlife exposed to toxic environmental chemicals, such as EDCs. The endocrine hormonal system is the main systems for controlling, communicating, and coordinating the mammals body activities [19]. The endocrine hormone works coordinate with the nervous system to regulate essential body activities. These activities include reproduction, growth, development, energy metabolism, homeostasis and osmoregulation. Moreover, it also regulates skeletal development and reproductive processes [20-24]. Indeed, it has been reported incidences of abnormalities in human sexual and cognitive development in some communities which normally being caused by EDC exposure [25-30]. The abnormalities in human sexual include decrease in the sperm quality, low sperm count, and many neurological and immunological effects [31].

Many of the carbon-based nanomaterials, such as: carbon nanotubes, fullerenes, nanofibers, nanohorns, graphene, and their chemically-modified analogues, has been investigated as an adsorption material. Due to their properties, these carbon-based nanomaterials have received huge applications in different areas of analytical chemistry and in many other techniques of medicine [32-35]. The unique structures of carbon-based nanomaterials allow them to interact with molecules via different non-covalent and covalent forces. These interactions includes hydrogen bonding, electrostatic forces, π-π stacking, van der Waals forces and hydrophobic interactions. Recently, graphene which is a novel and indeed fascinating carbon material, has sparked a tremendous research work from both the theoretical and experimental scientific societies [36]. Due to its extraordinary chemical properties and very high specific surface area, carbon-based nanomaterials have been extensively applied as adsorbents in solid-phase microextraction techniques and applications.

The ionic liquids has received a wide attention as an alternative to environmentally toxic organic solvents, it is increasingly applied in many techniques in analytical chemistry. These include nanoadsorbents techniques, different methods of chromatography and electro-chemistry science. Ionic liquids have many unique properties that can be changed by the proper choice of the building cations and anions species, these properties including its polarity, hydrophobicity and viscosity. Their chemical nature allows the synthesis of many different ionic liquid solvents with different properties for different applications and areas [37-40]. The impact of ILs in analytical chemistry resulted from their unique properties as negligible vapor pressure associated with high thermal stability, non-molecular solvents, tunable viscosity, good extractability in various inorganic and organic compounds, and miscibility with water and organic solvents. Modified materials, which consist of ionic liquid and magnetic composites, has been applied in many analytical chemistry extraction techniques [41]. One of the problem in the use of ionic liquids in chemistry applications is that at high temperatures the viscosity of the IL is reduced, therefore resulting in a flowing state in which the IL can be lost. Scientists overcomes this problem by the modification of the ILs to polymeric ILs (PILs). The PILs have unique properties such as higher thermal stability compared to monomeric ILs and indeed more resistance to flow. Moreover, PILs are tunable by proper functionalization and modification of the IL monomers, thus changing extractive capabilities along with their physicochemical properties [42-44].

Additionally, the combining the two above-mentioned fields, i.e. carbon-based nanomaterials and MILs, it is possible to design and develop new microextracting phases with outstanding properties for extraction of selected phenolic endocrine disrupting chemicals (8 phenolic EDCs) from water samples. The Functionalized carbon-based nanomaterials with MILs are expected to possess unique adsorbents advantages with tunable microextraction capabilities. This paper provides snapshot of our applications of magnetic ionic liquids (MILs) functionalized graphene oxide GO-MILs in microextraction, as an integral step of sample preparation for the removal of eight selected phenolic endocrine disrupting chemicals (8 phenolic EDCs) from water samples. The results obtained are accurate and highly reproducible, making it a good alternative approach for routine analysis of were applied to assess and detect the phenolic EDCs in water samples. The low-cost approach is straightforward, environmentally safe and exhibits high enrichment factors and absolute extraction percentages and satisfactory recoveries. To the best of our knowledge, this is the first time that a MIL-GO is used for analytical purposes in a practical, efficient and environmentally friendly MIL-GO microextraction approach for phenolic endocrine disrupting chemicals.

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Materials and Methods
Reagents and chemicals

Chemicals used such as graphite powder, KMnO$_4$, NaNO$_3$, H$_2$SO$_4$, HCl, DMF, standard substances in the chemical analysis and ionic liquids 1-butyl-3-aminopropyl imidazolium tetrachloroferrate were purchased from Sigma-Aldrich Chemical Co. bisphenol A (BPA), octylphenol (OP), nonylphenol (NP) and diethylstilbestrol (DES)) Chlorophenol Red-β-D galactopyranoside (CPRG), 17β-estradiol (E2), bisphenol A (BPA), and bisphenol A-d16 (BPA-d16) were purchased from ANPEL Laboratory Technologies Incorporation (Shanghai, China). Chromatographic grade methanol, acetonitrile, and isopropanol were purchased from Sigma-Aldrich (St. Louis, MO). The aqueous solutions were prepared using deionized water (a Milli-Q water purification system from Millipore), methanol, dichloromethane, n-hexane and acetone used of HPLC grade. The glass fiber filters (GF/F, pore size 0.7 μm) were prebaked at 250°C for 2h prior to use. The solid-phase microextraction sorbent GO-(Bapim)FeCl$_4$ was prepared in our lab. Stock solutions of the phenols and standard were prepared in methanol at 1 mg/mL and stored in amber glass bottles at 4°C.

Synthesis of ILs-modified GO composites

GO was prepared using the well known modified Hummers method, and MILs was prepared GO through amidation reaction between the carboxyl groups of GO and the amino groups of the MILs. 50 mg of GO was dispersed in 100 mL of deionized water by ultrasonication for 1h. After 1hr 200 mg of EDC and 160 mg of NHS were added to ensure the homogeneity of the solution. Then, solution was magnetically stirred for 2h to activate the carboxyl groups of GO, after that 200 mg of 1-butyl-3-aminopropyl imidazolium tetrachloroferrate was added and the mixture was then ultrasonicated for 60 minutes, the mixture was stirred at 30°C for 1h. The final product was washed several times with deionized water and methanol.

Instrumentation measurements

The morphology characterization of GO and the MIL-GO nanocomposites were obtained on a JEM-2010 transmission electron microscope (TEM, JEOL Ltd., Japan), a HITACHIS-4800 scanning electron microscope (SEM, Hitachi Co. Ltd., Tokyo, Japan). FT-IR spectra analysis were performed on a Thermo Nicolet iS50 Fourier-transform (FT) infrared spectrometer (Madison, WI, USA). Raman spectra analysis were performed on a Nicolet 6700/NXR FT-Raman spectrometer (Thermo Electron, USA) with a laser excitation of 532 nm. The X-ray diffraction (XRD) patterns were recorded with a Bruker D8-Advance diffractometer using Cu-Kα (λ = 0.15418 nm) in the 10-1000(2θ) range with a step size of 0.020. Field-emission scanning electron microscopy (FESEM) images were recorded on a Hitachi SU-8010 apparatus. The Brunauer-Emmett-Teller (BET) specific surface area was ascertained by a nitrogen adsorption data measured at 77K on a Quantachrome NOVA4000 surface area analyzer. X-ray photoelectron spectroscopy (XPS) patterns were recorded on a Thermo Scientific Escalab 250Xi spectrometer with Al-Kα (1486.6V). HPLC analyses were performed using ultra-fast liquid chromatography (LC-30A, Shimadzu, Japan) with an auto sampler with deuterium lamp as the light source (228 nm) and Separation column for the eight PAEs was Shimadzu shim-pack XR-ODS III (1.6 μm). The injection volume of phenols and standard were 5 μL, and the flow rate was 0.5 mL/min. Binary mobile phase were used in the separation, optimized using a of ultrapure water (solvent A) and acetonitrile (solvent B). The gradient elution program was: 0 - 3.5 min = 35% B, 3.5 - 4 min = 35 - 70% B, 4 - 6.0 min = 70% B, 6.0 - 7 min = 70 - 60% B, 7 - 9 min = 60% B, 9 - 9.0 min = 60 - 100% B, 9.0-15 min = 100% B.

SPE procedure and real sample preparation

20.0 mg of the GO-MILs nanocomposite was packed into a standard filter, to acted as a homemade SPE column. The column was preconditioned with 2 mL of methanol and 2 mL of water. 10 mL of the sample solution was passed through the column at a flow rate of 0.5 mL/min. Then, the adsorbed phenols were eluted with 1.0 mL of methanol and concentrated to dryness under a steam of nitrogen before detection. Fused silica and glass materials were used for the entire procedure to avoid any possible interferences. The quantification was based on external calibration with areas relative to the internal standard areas (at least eight calibration standard solutions, r 2 was

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always above 0.98). The internal standard used for the EDCs was d16-bisphenol-A. The concentrations determined were corrected for average blank value and relative recovery were recorded.

Results and Discussion

FT-IR analysis

Figure 1a shows the FTIR spectra of GO and figure 1b shows the FTIR spectra of GO-(Bapim)FeCl₄ nanocomposites. The peak at approximately 3500 cm^{-1} is caused by stretching vibration of the -OH group on the surface of GO, the peak at 1729.97 cm^{-1} which are caused by the C-O stretching vibration of the carboxyl group, and that at 1623.14 cm^{-1} is belongs to the C=C bonds in the aromatic groups. In figure 1b, the peak at 1167.11 cm^{-1} is attributed to ring in-plane asymmetric stretching of the imidazolium ring. The obvious peak at 1617.68 cm^{-1} and the peak at 1652.15 cm^{-1} associated with -CONH group. These results suggest that the MILs have been successfully grafted onto the GO surface to form GO-(Bapim)FeCl₄ nanocomposites. Thus, FTIR results proved that the functional amine of ionic liquids were chemically bond with carboxyl group on the surface of graphene oxide. Moreover, the imidazolium ring of ionic liquids with alkyl chains offer large π-π and hydrogen-bond interactions and enhance electrostatic inter-sheet repulsion which result in an increase to the interaction capacity and connectivity between GO and (Bapim)FeCl₄.
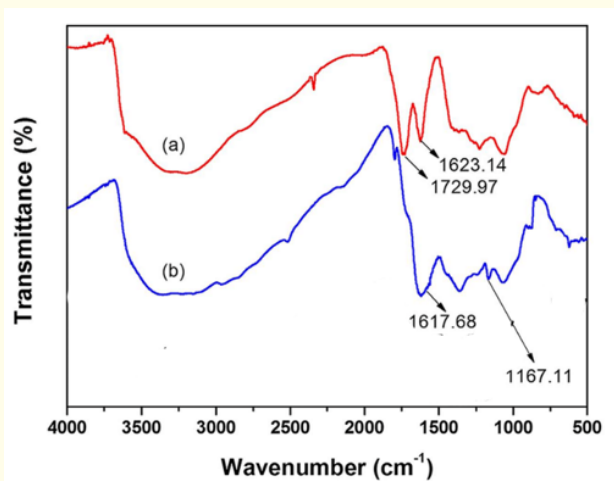

Figure 1: *FTIR spectra of (a) GO, and (b) GO-(Bapim)FeCl4 nanocomposites.*

Raman spectrum of GO and GO-(Bapim)FeCl4 nanocomposite

Figure 2a shows the Raman spectra of GO and figure 2b shows Raman spectra of GO-(Bapim)FeCl₄ nanocomposites, with a laser excitation of 532 nm was used. In the two samples clearly visible and strong peaks were noticed at approximately 1595 cm^{-1} and 1353 cm^{-1} which assigned to the G and D bands, respectively. We noticed red-shift in the D-band of the Raman spectra of GO-(Bapim)FeCl₄ nanocomposites, as shown in figure 2b. The red-shift phenomena is caused by the bonding between the C and N atoms, therefore changes the electronic structure of GO. Indeed, the intensity ratios of the two peaks (ID/IG) demonstrate the extent of defects on the GO surface caused by reaction with (Bapim)FeCl₄ and can be used to reflect the extent of covalent binding. The ID/IG ratio of GO and the GO-(Bapim)FeCl₄ nanocomposites are 1.01 and 1.07, respectively, corresponding to a slightly increased ratios of ID/IG. And thus an increase in disorder, and indicate the successful formation of amidation reaction between the GO and (Bapim)FeCl₄ to form the final product GO-(Bapim)FeCl₄ nanocomposites.
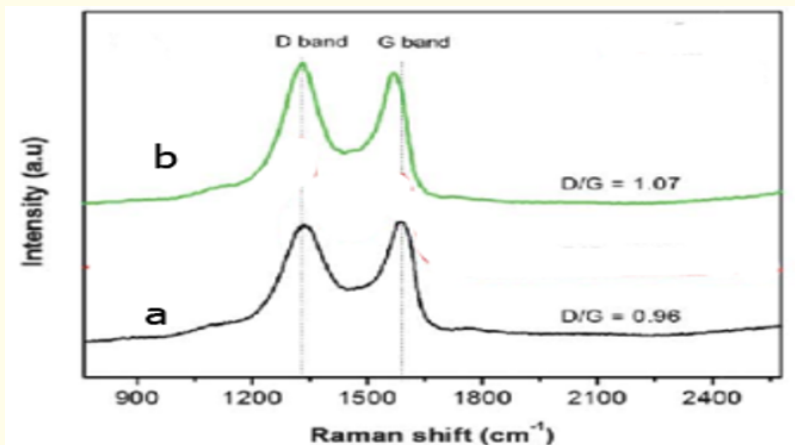
Figure 2: Raman spectra of (a) GO, and (b) GO-(Bapim)FeCl4 nanocomposites.

AFM image of GO, and GO-(Bapim)FeCl4 nanocomposites

Atomic force microscopy images (AFM) image was performed in order to examine the morphology of GO, and GO-(Bapim)FeCl$_4$ nano-composites. AFM images before and after deposition of (Bapim)FeCl$_4$ on the surface of GO is shown in figure 3. It can be seen from figure 3 a single layer of GO and a single sheet of GO-(Bapim)FeCl$_4$ of varying size. (Bapim)FeCl$_4$ are deposited on the surface of GO substrate with clear overlaps. Before deposition of (Bapim)FeCl$_4$ GO the thickness of GO vary in size from 1.0 to 1.3 nm.
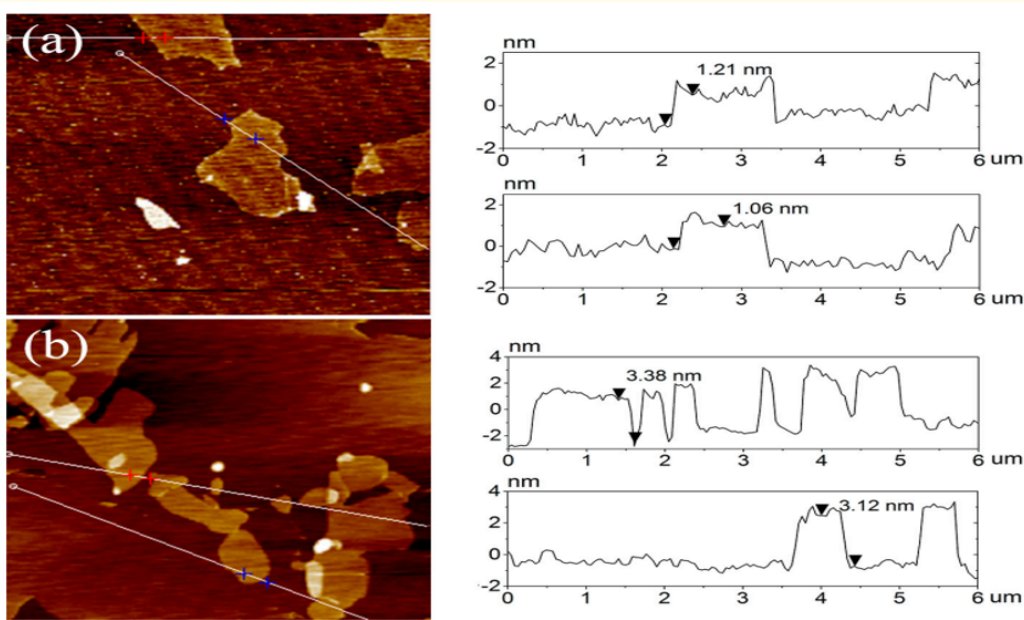

Figure 3: AFM image of (a) GO, and (b) GO-(Bapim)FeCl4 nanocomposites.

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However, after functionalization with magnetic ionic liquid (MILs), the thickness of GO single layer sheets are obviously increased to nearly 2.1 - 2.5 nm, indicating the presence of magnetic ionic liquid (Bapim)FeCl$_4$ on the surface of GO sheet.

X-ray diffraction (XRD) analysis

X-ray diffraction (XRD) analysis was applied to characterize the crystallographic structure of the fresh and used GO-(Bapim)FeCl$_4$ nanocomposites. As presented in figure 4, the XRD pattern demonstrating the successful preparation. The used GO-(Bapim)FeCl$_4$ nano-composites displays the same diffraction pattern as the fresh sample with little decrease in intensity, which indicates that the crystal structure of the GO-(Bapim)FeCl$_4$ nanocomposites is maintained after microextraction. Moreover, the discernible diffraction peaks at about 9.3^0 belonging to GO can be detected in the pattern of GO-(Bapim)FeCl$_4$ nanocomposites. This X-ray diffraction (XRD) analysis result indicates the incorporation of (Bapim)FeCl$_4$ into the GO structure.
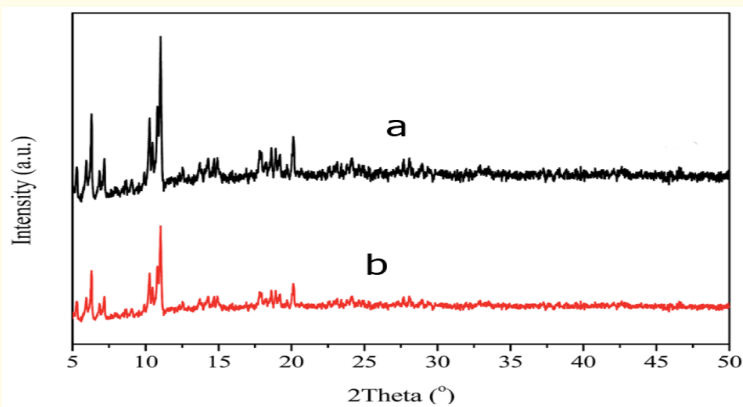

Figure 4: *X-ray diffraction (XRD) analysis of (a) Fresh GO-(Bapim)FeCl4 nanocomposites, and (b) used GO-(Bapim)FeCl4 nanocomposites.*

XPS scanning spectrum

GO and the GO-(Bapim)FeCl$_4$ nanocomposites were investigated by X-ray photoelectron spectroscopy (XPS). Figure 5a shows that only the C1s and O1s peaks exist in GO (Figure 5a bottom). The C1s peaks are obtained by coupling the five peaks at 284.8, 285.5 286.8, 287.8, and 288.7 eV, assigned for C-C, C-OH, C-O, C=O and O=C-OH, respectively. The N1s peak can be shown in GO-(Bapim)FeCl$_4$ (Figure 4a above). Relative to GO, the peak fitting of C1s in GO-(Bapim)FeCl$_4$ yields a new functional group (i.e., C-N, Figure 5c), which is indicted a successful amidation reaction between (Bapim)FeCl$_4$ and GO. As shown in figure 5, the N content increases from 0.68% to 5.10%, while the O content decreases, demonstrating a covalent amidation bonding between GO and the (Bapim)FeCl$_4$. Similar results were obtained with the used GO-(Bapim)FeCl$_4$ sample after microextraction of the phenolic endocrine disrupting chemicals.

SEM images and TEM images

Figure 6a and 6b shows TEM images of (a) GO, (b) GO-(Bapim)FeCl$_4$ nanocomposites, respectively; and Figure 6c and 6d depict SEM images of (c) GO and (d) GO-(Bapim)FeCl$_4$ nanocomposites. As revealed in TEM and SEM images GO possesses a wrinkled single-layer structure with semitransparent flake-like shape. After being grafted by the MILs, the GO-(Bapim)FeCl$_4$ nanocomposites still maintain the lamellar structure shown in figure 6b and 6c, which ensures that they retain a high specific surface area and high adsorptive performance. An obvious change in the SEM images can be observed after reaction of GO with (Bapim)FeCl$_4$, the large GO sheets are reduced to small pieces, thus giving the appearance of holes with different sizes. This appearance phenomenon can explain the novel nanocomposites structure of GO-(Bapim)FeCl$_4$, that have the advantage of the high adsorptive capacity of phenolic endocrine disrupting chemicals.

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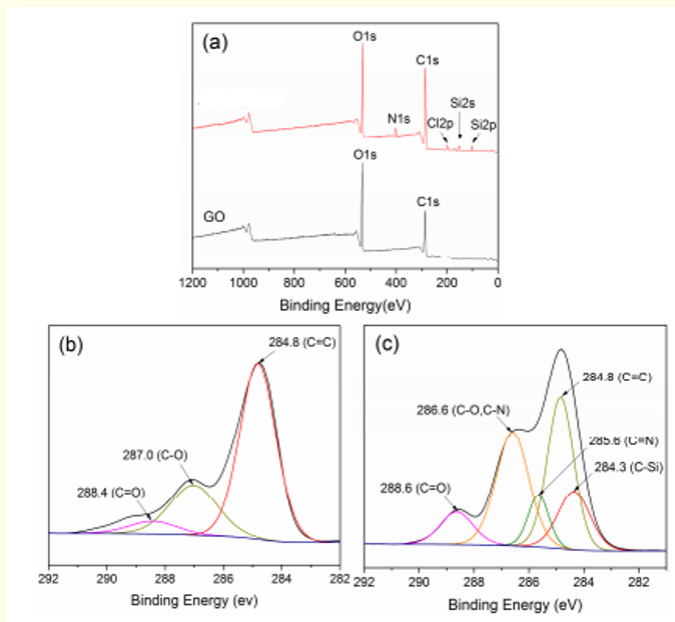
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Figure 5: *XPS spectra of GO and GO-(Bapim)FeCl4 nanocomposites (a) survey spectra; (b) C 1s of GO; (c) C 1s of GO-(Bapim)FeCl4 nanocomposites.*
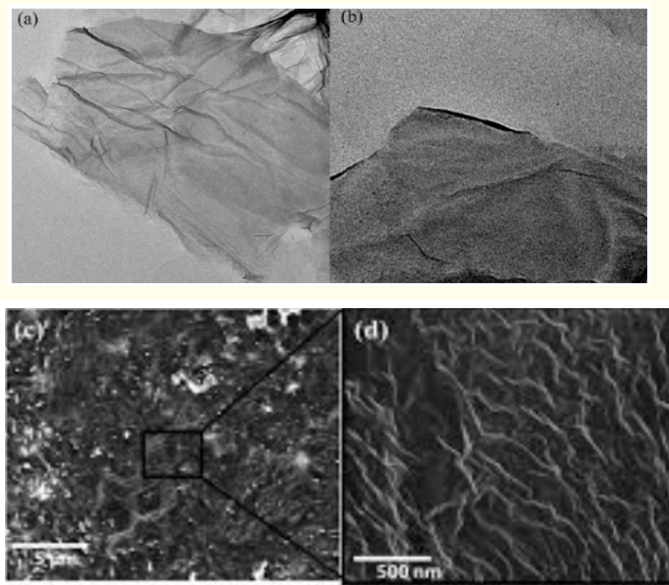

Figure 6: *TEM images of (a) GO, (b) GO-(Bapim)FeCl4 nanocomposites; and SEM images of (c) GO and (d) GO-(Bapim)FeCl4 nanocomposites.*

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The removal of phenolic endocrine disruptors by GO-(Bapim)FeCl4 nanocomposite

Figure 7 shows HPLC chromatograms of Eight phenolic endocrine disruptors standard and the extracts of a sample from GO-(Bapim) FeCl$_4$ nanocomposites after recovery As shown in table 1, we find that the GO-(Bapim)FeCl$_4$ nanocomposites possess a superior nano-extraction capacity, mainly for E3, E2, EE2 and CPRG, with the removal efficiencies were above 95% even to 100%. The results indicates a decreased in carbon chain length of MILs benefits the extraction, and the primary influence to the adsorptive performance of the GO-MIL nanocomposites was the nature of the anion, the hydrophobic effect restrained the extraction process in the aqueous system. The results indicated that GO-MIL adsorption capacities especially for BPA, DES, OP, and NP are significantly weaker, with the removal efficiencies were between 80% to 87.
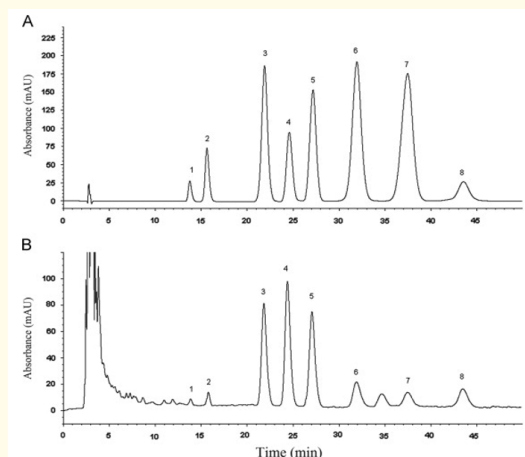

Figure 7: HPLC chromatograms of: (A) Eight phenolic endocrine disruptors standard and (B) The extracts of a sample from GO-(Bapim) FeCl4 nanocomposites after recovery. Peaks (1) BPA, (2) DES, (3) E3, (4) E2, (5) EE2, (6) OP, (7) NP, and (8) CPRG.

pH	Recovery % of phenolic EDCs using sing GO-(Bapim)FeCl$_4$ composite[a] ± S.D							
	BPA	OP	NP	DES	CPRG	E2	E1	EE2
3	55 ± 1.6	51 ± 2.4	52 ± 1.2	87 ± 1.7	57 ± 1.6	59 ± 2.1	57 ± 2.1	57 ± 1.5
4	56 ± 3.2	54 ± 2.0	57 ± 1.7	54 ± 1.1	59 ± 1.2	57 ± 1.4	57 ± 1.1	59 ± 1.8
5	62 ± 2.2	67 ± 2.3	67 ± 1.6	67 ± 1.8	67 ± 1.9	67 ± 1.8	67 ± 2.0	67 ± 1.2
6	61 ± 1.8	67 ± 22	67 ± 1.8	67 ± 1.9	67 ± 1.7	67 ± 1.4	67 ± 0.1	67 ± 2.6
7	72 ± 1.7	77 ± 1.0	77 ± 1.3	77 ± 2.4	77 ± 1.8	77 ± 2.0	77 ± 1.8	77 ± 1.8
8	82 ± 1.8	80 ± 1.8	84 ± 1.9	85 ± 1.3	87 ± 2.7	87 ± 3.0	87 ± 1.4	87 ± 1.1
9	85 ± 1.6	80 ± 1,8	85 ± 1.8	87 ± 1.0	99 ± 1.9	98 ± 4.1	99 ± 1.6	95 ± 2.6
10	87 ± 1.3	80 ± 1.4	84 ± 1.6	86 ± 1.1	97 ± 2.1	97 ± 1.5	97 ± 2.0	97 ± 0.6
11	85 ± 1.2	86 ± 1.5	86 ± 1.3	87 ± 1.5	98 ± 2.6	97 ± 1.7	95 ± 1.5	97 ± 1.4

Table 1: The compared adsorption capacity of phenolic EDCs from aqueous sample with GO-MILs nanocomposites at different pH values. Phenolic EDCs concentration was 20 ng/mL, and Incubation Time, 10 minutes.
a: Mean of three determinations.

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The influence parameters for pH values, elution time, and salt concentration were evaluated to find the optimal extraction performance of the GO-(Bapim)FeCl$_4$ for the extraction of eight phenolic EDCs. In our laboratory, the extraction capability of GO-(Bapim)FeCl$_4$ for the phenolic EDCs were tested in the range of 3.0-11. As shown in table 1, the adsorption capacity (mg/g) clearly increased as the pH increased from 3.0 to 9.0. Therefore, a pH of 9.0 was chosen for the next adsorption tests. It is noteworthy, that HPLC peak areas decreased as the pH became greater than 9.0. Moreover, HPLC chromatogram revealed that phenolic EDCs are relatively independent of changes to the sample solution pH in the range of 2.0 - 10, because they exist as neutral molecules. It is important to mention that the pH value primarily influences the charge of the ionic liquids and other functional groups such as hydroxyl, carboxyl groups and epoxide on the surface of GO. Table 2 shows the effect of elution time on the extraction performance by changing the washing time from 1 minute to 5 minutes our experimental results revealed that the best elution time was 4. Thus, elution time was set to 4 minutes to ensure balance between optimum time and efficiency. As indicated in table 3, the salt concentration is a significant factor that influences the extraction capabilities, with 1% (w/v) sodium chloride (NaCl) added to the sample solution, the extraction performance of most of the eight phenolic EDCs reached a maximum. This result may be occurred due to the salt-out effect, which usually promotes extraction. However, when the NaCl concentration exceeds 2% (w/v), the mass transfer process in the solid/liquid interface becomes inhibited from the increased viscosity, leading to a reduction of the diffusion rate of the phenolic EDCs, which decreases the extraction efficiency. For this reason, 1% (w/v) NaCl was added to the phenolic EDCs sample solution.

Elution time, min	Recovery % of phenolic EDCs using sing GO-(Bapim)FeCl$_4$ a ± S.D							
	BPA	OP	NP	DES	CPRG	E2	El	EE2
1	61 ± 1.4	67 ± 22.	67 ± 1.5	67 ± 1.6	67 ± 1.2	67 ± 1.3	67± 0.9	67 ± 2.0
2	72 ± 1.3	77±1.6	77 ± 1.7	77 ± 2.1	77 ± 1.3	77 ± 1.0	77± 1.5	77 ± 1.3
3	82 ± 1.5	80 ± 1.6	84 ± 1.4	85 ± 1.8	87 ± 2.1	87 ± 3.0	87± 1.5	87 ± 1.6
4	85 ± 1.6	80 ± 1.8	85 ± 1.8	87 ± 1.0	99 ± 1.9	98 ± 1.1	99± 1.9	95 ± 2.0
5	85±1.6	80 ± 1.9	85 ± 1.2	87 ± 1.3	99 ± 1.6	98 ± 2.1	99± 1.5	95 ± 2.1

***Table 2:** The compared adsorption capacity of phenolic EDCs from aqueous sample with MGO and GO-MILs nanocomposites at different time interval. phenolic EDCs concentration was 20 ng/mL, and Incubation Time, 10 min.*
a: Mean of three determinations

Salt conc.% (w/v)	Recovery % of phenolic EDCs using sing GO-(Bapim)FeCl$_4$ a ± S.D							
	BPA	OP	NP	DES	CPRG	E2	E1	EE2
1%	87 ± 1.2	87 ± 2	87 ± 1.2	87 ± 1.4	87 ± 1.7	87 ± 2.1	87 ± 2.1	87 ± 1.6
2%	83 ± 1.2	87 ± 2	84 ± 1.2	81 ± 1.4	84 ± 1.7	85 ± 2.1	87 ± 2.1	83 ± 1.6
3%	72 ± 1.2	76 ± 2	79 ± 1.2	78 ± 1.4	80 ± 1.7	81 ± 2.1	80 ± 2.1	80 ± 1.6
4%	67 ± 1.2	64 ± 2	66 ± 1.2	65 ± 1.4	62 ± 1.7	63 ± 2.1	67 ± 2.1	64 ± 1.6
5%	57 ± 1.2	58 ± 2	57 ± 1.2	66 ± 1.4	67 ± 1.7	67 ± 2.1	59 ± 2.1	58 ± 1.6

***Table 3:** The compared adsorption capacity of phenolic EDCs from aqueous sample with MGO and GO-MILs nanocomposites at different salt concentration.*
a: Mean of three determinations.

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Validation of the method

The analytical parameters for the GO-(Bapim)FeCl₄ nanocomposites nano-extraction of the phenolic endocrine disruptors, such as linearity, correlation coefficients (r2), limits of detection (LODs), limits of quantitation (LOQs) and repeatability were tested under the optimal experimental conditions by using a series of spiked natural water samples. As indicated in table 4, the linearity of the eight phenolic endocrine disruptors ranged from 2 ng mL^{-1} to 200 ng mL^{-1}, with the correlation coefficients exceeding 0.99. The LODs and LOQs were defined as the corresponding concentration equivalent to three and ten times the signal-to-noise ratios, in our results ranged from 0.02 ng mL^{-1} to 0.88 ng mL^{-1} and from 0.06 ng mL^{-1} to 2.94 ng mL^{-1}, respectively. The sensitivity of the method, with the use of a UV detector, is quite satisfying, mainly because the detector is easily available to most analytical laboratories. The reproducibility of the method was determined by intra-day RSDs (n = 3) and inter-day RSDs (n = 3) at a spiked sample concentration of 50 ng mL^{-1}. The two RSD values were always less than 8.0%. Indeed, all of our results indicated a high sensitivity and good reproducibility of this new method.

Analytes	Linear range (ng/mL)	Calibration equations	r2	LODs (ng/mL)	LOQ (ng/mL)	Intra-day RSDs (%)	Inter-day RSDs (%)
BPA	10-200	y = 25 ± 1x - 57	0.9944	0.88	2.94	8	8.1
OP	2-100	y = 752 ± 16x + 1012	0.9976	0.1	0.09	3.8	5.6
NP	2-200	y = 436 ± 6x-27	0.9989	0.08	0.16	3.9	6.2
DES	5-200	y = 246 ± 7x-1186	0.9967	0.03	0.32	6.4	7.3
CPRG	5-200	y = 264 ± 7x-48	0.9971	0.08	0.28	5.0	7.1
E2	5-200	y = 182 ± 6x + 230	0.9957	0.12	0.39	6.2	6.6
E1	10-200	y = 46 ± 2x + 2070	0.9948	0.24	0.8	6.6	7.2
EE2	10-200	y = 48 ± 2x - 26	0.9932	0.44	1.42	4.6	5.8

Table 4: *Analytical parameters of the GO-(Bapim)FeCl₄ method for the nanoextraction of eight of phenolic endocrine disruptors in spiked water.*

Conclusions

This research highlights the preparation of novel types of GO-MILs nanocomposite for use in SPME of phenolic endocrine disruptors from aqueous solution. In our research, a magnetic ionic liquid modified graphene oxide nanocomposites were prepared through a direct amidation reaction between GO and MIL. The prepared nanocomposites were used as the nanoadsorbent in a fixed-bed column, which possessed the advantages of low column pressure and high nanoadsorption capacity. Moreover, the system was successfully applied to the microextraction of eight of phenolic endocrine disruptors aqueous samples with good reproducibility, wide linear range and low LODs using a standard HPLC-UV detector. The proposed method provides a reliable method for the removal and determination of phenolic endocrine disruptors in aqueous solution, which can be applied in water treatment and the regulation of supplies.

Conflicts of Interest

The authors declare no conflict of interest regarding the publication of this paper.

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