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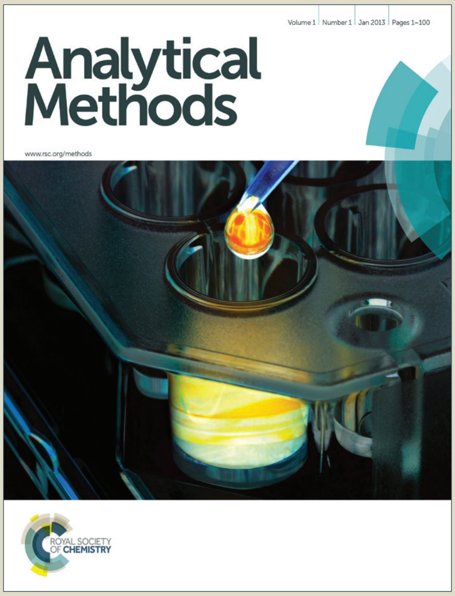

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ARTICLE

Dispersive solid-phase extraction of organophosphorus pesticides from apple, cucumber and water samples using reduced graphene oxide coated with ZnO nanocomposites as sorbent

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Ting Sun,[a] Yongri Jin,[a] Jie Yang,[a] Lanjie Li,[a] Xiaolei Shi[b*] and Xuwen Li[a*]

In this work, reduced graphene oxide coated with ZnO (RGO-ZnO) nanocomposites was synthesized by hydrothermal reduction strategy. The presence of ZnO was characterized by fourier transform infrared spectroscopy (FT-IR), X-ray diffraction (XRD), thermogravimetric analysis (TGA), scanning electron microscopy (SEM), and transmission electron microscopy (TEM). Subsequently, RGO-ZnO was used as dispersive solid-phase extraction (DSPE) sorbent for the enrichment of eight organophosphorus pesticides (OPs) in apple, cucumber and water samples prior to their determination by gas chromatography-mass spectrometry (GC-MS). Several experimental parameters affecting the extraction efficiencies, including the amount of the RGO-ZnO, extraction time, the pH of sample solution, as well as type and volume of eluent solvent, were investigated and optimized in this work. Under the optimal experimental conditions, good linearity existed in the range of 0.5–200.0 ng g^{-1} for all the analytes with the correlation coefficients (R^2) ranging from 0.9894 to 0.9983. The limits of detection (S/N = 3) of the method for the compounds were between 0.01 and 0.05 ng g^{-1}. Good reproducibilities were acquired with the relative standard deviations (RSDs) below 8.6% for both intra-day and inter-day precision. The recoveries of the method were in the range from 75.0% to 104.2%.

1 Introduction

Organophosphorus pesticides (OPs), as the most commonly applied pesticides in agriculture, are widespread used as across the world.[1,2] However, some OPs do not readily decompose and migrate from soil to water sources.[3] Due to their persistence, high toxicity and bioaccumulation along the food chain, the extensive use of OPs can cause environment pollution and ecological problem, and their presence in water and food poses increased hazard to human health.[4] Hence, sensitive and selective analytical methods for the quantification of OPs at trace levels have become increasingly important for public security and health protection. Currently, the instrumental analysis methods for OPs determination have been involved in Gas chromatography (GC),[5] high performance liquid chromatography (HPLC), gas chromatography coupled with mass spectrometry (GC-MS)[6, 7] and liquid chromatography coupled with mass spectrometry (LC-MS), enzyme-linked immunosorbent assays (ELISAs)[8] and Electrochemical biosensor,[9] due to their high sensitivity and precision for the trace analysis of OPs.[10] As we know, extraction and enrichment of OPs in samples were needed prior to instrumental analysis, because of the matrices are complex and the target analytes are in extremely low concentrations in samples. Sample preparation is one of the most important and crucial step in a whole analytical process. To date, various solid-phase extraction (SPE) is used as sample preparation methods in Ops analysis, such as cartridge,[11] solid phase microextraction (SPME),[12] headspace solid phase microextraction (HS-SPME),[13] matrix solid-phase dispersion (MSPD),[14] stir bar sorptive extraction (SBSE),[15] magnetic solid-phase extraction (MSPE).[16] A wide range of studies hitherto bears testimony to the fact that they are effective and economical for sample preparation. However, most of these methods due to the limited interface between the adsorbent and the aqueous samples, a considerable time is needed for the analytes to be extracted into the organic phase or onto the adsorbent surface.

Dispersive solid-phase extraction (DSPE) which was introduced by Anastassiades et al. in 2003,[17] can be considered as quick, easy, cheap, effective, rugged and safe (QuEChERS) sample treatment method. In this method, the loose sorbent is dispersed in the sample solution to remove the interferences and collected by centrifugation after extraction. DSPE, as an independent sample preparation method, has been widely applied for various samples. The main advantage of DSPE is the increase in active contact area between the analytes and adsorbent. Thus, the extraction efficiency

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[a] *College of Chemistry, Jilin University, Jiefang street 2519, Changchun 130021, P. R. China.*
[b] *College of Quartermaster Technology, Jilin University, Xian Street 5333, Changchun 130062, P. R. China.*
** Corresponding author: Professor Xuwen Li, E-mail address: xwli@jlu.edu.cn Tel./fax: +86-431-85167996.; Professor Xiaolei Shi, E-mail address: xlshi@jlu.edu.cn*
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is high with less extraction time and channeling or blockage, as occurs frequently in SPE cartridge or column, is avoided.[18] For DSPE method, the application of effective sorption material is critical. It is important to seek new adsorbents with a high loading capacity and selective adsorption ability.

Graphene, as a new two-dimensional carbon nanomaterial, arouse great interest among scientists in various fields. Due to its large specific surface area, chemical stability, flexibility and π-electron rich structure, *etc*. make a good candidate as a sorbent for sample pretreatment.[19, 20] However, there are some problems for directly used graphene as adsorbents. Firstly, the irreversible aggregate through van der Waals interaction due to their high surface area.[21] Secondly, tiny graphene sheets blocked of frits in SPE cartridge or column. Thirdly, pure graphene tends to aggregate in aqueous solution. These problems may hinder effective sorption behaviors and reduce the sorption capacity. One effective strategy to get graphene as individual sheet is to decorate the surface of graphene sheets by introducing nanoparticles to enhance its sorption performance. Reduced graphene oxide coated with ZnO nanoparticles (RGO-ZnO) can avoid the above-mentioned problems and maintain the advantageous characteristics of graphene sheets. Moreover, the water-solube analytes are adsorpted effectively by the hydrophilic surface of ZnO in water. Also the high density of ZnO helps the facile separation of RGO-ZnO from water after adsorption by centrifugation.[22] Basis of their remarkable properties, RGO-ZnO nanocomposites is expected to be a potential adsorbent for enrich Ops in various samples. Nowadays, RGO-ZnO nanocomposites as photocatalyst are widely used in water for pollutant adsorbent for its photocatalytic ability. [23-25] To the best of our knowledge, there is no relevant report regarding the application of RGO-ZnO nanocomposites as DSPE sorbent on sample preparation of OPs before instrumental analysis.

In this work, we first synthesized RGO-ZnO, using a facile hydrothermal reduction strategy. The nanocomposites could be produced directly from graphene oxide (GO) in a facile one-step reaction, where the reduction of GO and the deposition of ZnO on graphene occur simultaneously. This method is simple, economical and environment friendly. The identity of nanocomposites was confirmed using FT-IR, XRD, TGA, SEM and TEM. Subsequently, the effects of the adsorption and desorption conditions on the performance of RGO-ZnO were evaluated. Under the optimal experimental conditions, eight types of OPs in different samples were enriched by RGO-ZnO as DSPE sorbent prior to their determination by GC-MS.

2 Experimental

2.1 Reagents and materials

Graphite flakes (99.95%, particle size ≤ 30 μm) were purchased from Alfa Aesar (Heysham, England). Potassium permanganate ($KMnO_4$), sulphuric acid (H_2SO_4), zinc acetate ($Zn(CH_3COOH)_2{\cdot}2H_2O$) were supplied by Beijing Chemical Works (Beijing, China). All of the organic reagents employed were analytical purity. Deionized water was used throughout the work. Pesticide standards of OPs (methamidophos, dichlorvos, omethoate, dimethoate, methyl parathion, malathion, chlorpyrifos, and parathion) were purchased from the Institute for Environmental Reference Materials of Ministry of Environmental Protection (Beijing, China), which were prepared in acetone the concentration of 100 μg mL^{-1}. A stock solution containing a mixture of eight Ops at the concentration of 10 μg mL^{-1} was prepared in methanol. A series of standard solutions were prepared by the appropriate dilution of the stock solutions and stored at 4 ℃ in a freezer.

Fresh apples, cucumbers were purchased from a local supermarket (Changchun, China); lake water was collected from the South Lake (Changchun, China).

2.2 Instruments and analytical conditions

Infrared absorption spectra were obtained with a Varian 800 FT-IR spectrometer (USA). XRD spectra were carried out using an XPert Powder X-ray diffractometer (Holland). TGA was characterized by STA 409 PC Luxx (Germany). SEM images were recorded on a Hitachi SU8010 SEM instrument (Japan). TEM images were obtained with a Hitachi H-600 transmission electron microscope (Japan).

An Agilent GC-MS (7890A GC and a quadrupole MS 5975C, Palo Alto, CA) with a 30m × 0.25 mm, 0.25 μm film DB-5 bonded-phase fused silica capillary column (J&W Scientific, Inc., Folsom, CA) was used. Helium carrier gas was set at a flow rate of 1.0 mL min^{-1}, and separation of the eight Ops was carried out under a temperature programmed as follows: injector temperature at 200 ℃, column temperature with an initial temperature of 80 ℃ (1-min hold) and ramped to 180 ℃ at a rate of 20 ℃ min^{-1} for 5min, finally increased to 240 ℃ at the rate of 40 ℃ min^{-1} and held for 5 min. The ions source was set at 280 ℃ and the ionization energy was under electron ionization mode at 70 eV. Aliquots of 1 μL were injected into the GC-MS system under splitless mode, and the analytes were respectively monitored under the single ion monitoring mode at multiple mass channels. Retention times, selected quantification and identification ions of the eight OPs are shown in Supporting Information Table S1.

2.3 Preparation of graphene oxide

Graphene oxide (GO) was prepared from graphite flakes by a modified Hummers method.[26, 27] In a typical procedure, 50 ml of concentrated H_2SO_4 is heated to 90 ℃ in a 500 ml beaker followed by the addition of $K_2S_2O_8$ (10 g) and P_2O_5 (10 g). Graphite powder (12 g) is added to the solution at 80 ℃, resulting in bubbling that subsides within 30 minutes. The mixture was agitated by stirrer and kept for 4.5 h at 80 ℃ using a hotplate and then diluted with 2 L of water and left overnight. The following day the mixture is filtered and transferred to a drying dish and left overnight under ambient conditions. Then the solid was added to a 2 L conical flask before filled with 460 mL concentrated H_2SO_4 at 0 ℃. $KMnO_4$ (60 g) was added slowly and the temperature was kept under 10 ℃. This mixture is then allowed to react at 35 ℃ for 2 h. After that, 920 ml of distilled water is added, during this procedure the temperature not allow to climb above 50 ℃. The mixture is stirred for 2 h before an additional 2.8 L of water is added. Subsequently, 50 ml of 30% H_2O_2 is added to the mixture until the bubbling of gas was completed. The mixture is allowed to settle for at least a day after which the clear supernatant is decanted. The product was made free from acid by centrifuging and washing several times with 10% HCl solution and water. The desired dark brown cake was redispersed in water, which was subjected to dialysis for 1 week to

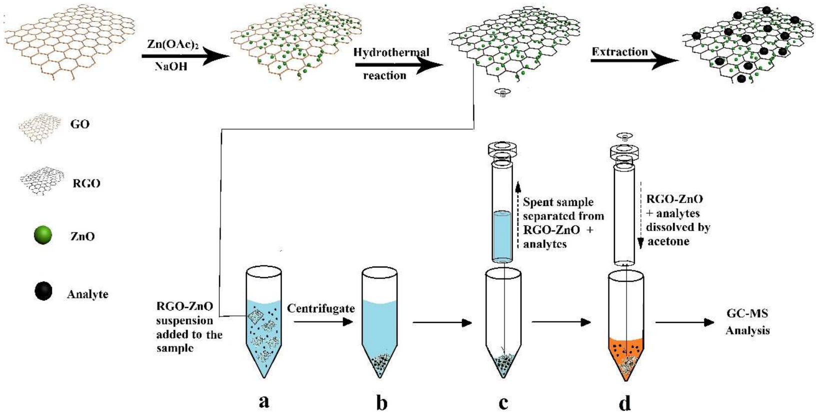

Figure 1 The overall synthetic procedure of RGO-ZnO nanocomposites and the DSPE procedure when RGO-ZnO was used as sorbent.

remove the residual salts and acids. And the brown suspension was dried under vacuum at 60 ℃.

2.4 Preparation of RGO-ZnO nanocomposites

GO (200 mg) was dispersed in 100mL of ethylene glycol and ultrasonically exfoliated in a bath sonicator for 30min to form a light-brown solution. A desired amount of zinc acetate (600 mg) was dissolved in 10 mL of ethylene glycol before adding the GO suspension. Afterward, the pH of the solution was adjusted to 9.0 with 1M NaOH solution while stirring continued for 1 h. Then the mixed solution was hydrothermally treated at 180 ℃ for 12 h. The obtained samples were washed and dried in a vacuum oven at 60 ℃. For comparison purposes, pure ZnO particles were obtained by using the same experimental conditions without the addition of GO. RGO was also prepared by solely treating GO in the hydrothermal reaction.

2.5 Sample preparation

Fresh apples and cucumbers were homogenized with a juice extractor, respectively. A 20 g portion of the homogenized samples were centrifuged at 4000 rpm for 5 min, the supernatant was collected and filtered through a 0.45 μm membrane to eliminate particulate matters, the supernatant was diluted to 40 mL with deionized water. The water sample was filtered through a 0.45μm membrane. The obtained sample solution was then subjected to the following DSPE procedure.

2.6 DSPE procedure

The DSPE procedure is shown in Figure 1. Typically, 20 mg of RGO-ZnO and 40 mL sample solution with desired initial concentration of OPs were added into a 50 mL centrifuge tube (Figure 1a), and then the tube was shaken on a platform shaker for 6 min at room temperature. After extraction the sample tube was centrifuged for 5min at 4000 rpm (Figure 1b) before the supernatant was removed by syringe and discarded (Figure 1c). The resultant sedimented phase was totally transferred to a 2 mL centrifuge tube. The enriched target analytes were desorbed from the RGO-ZnO with 1.0 mL of acetone by vigorously vortex for 1min (Figure 1d). After centrifugation for 5min at 4000 rpm, the supernatant solvent was collected by syringe. This desorption procedure was repeated another two more times. The eluates were combined together and then dried under a stream of nitrogen. The residue was redissolved with 50 μL methanol, and 1 μL was injected into the GC-MS system for analysis.

3 Results and discussion

3.1 Characterization of the RGO-ZnO nanocomposites

GO reduction during hydrothermal treatment was confirmed by FT-IR spectra. Figure 2a shows the FT-IR spectra of as-prepared composite material. On the spectrum of GO, several characteristic peaks can be observed, confirming the successful oxidation of graphite. In detail, bands at 3410, 1735, 1597, 1064 cm^{-1} are attributed to the stretching vibrations of O-H, C=O, aromatic C=C, and alkoxy C-O.[28] In contrast, the absorption at 1735 cm^{-1} (C=O stretching vibrations) was decreased very much in intensity and most bands that are related with the oxygen-functional groups almost vanished in the spectrum of RGO-ZnO, suggesting the successful reduction of oxygen-functional groups by hydrothermal treatment. XRD measurements were used to investigate the phase and crystal structure of products. The XRD pattern of graphite flakes, GO, RGO, ZnO and RGO-ZnO with the corresponding 2θ values were presented in Figure 2b. After the oxidation of graphite flakes, reflection peak shifts to the lower angle at 2θ = 10.0° (d-spacing = 8.75 Å) from the 2θ = 26.6° (d-spacing = 3.35 Å) indicates the successful oxidation of graphite.[29] In the XRD pattern of ZnO and RGO-ZnO nanocomposites, all the diffraction peaks can be perfectly indexed to the standard value of the zincite (JCPDS card No. 36-1451) phase, and no typical diffraction peaks of GO were observable after reduced by hydrothermal treatment, which is in agreement with the GO being reduced to graphene during the synthesis process. According to the spectra of RGO and RGO-ZnO, without extra diffraction peaks related to carbon, it can be concluded that the RGO sheets were completely exfoliated due to the loading of ZnO nanoparticles on their surfaces.[30] These results suggested the presence of ZnO nanoparticles on RGO sheets, which is consistent with the FT-IR results. In order to determine the mass content of RGO in the RGO-ZnO nanocomposites, TGA was performed in air at the temperature range from room temperature to 800 °C at a rate of 10 °C min^{-1}. Figure 2c illustrates the TGA curve of the RGO-ZnO nanocomposites. It

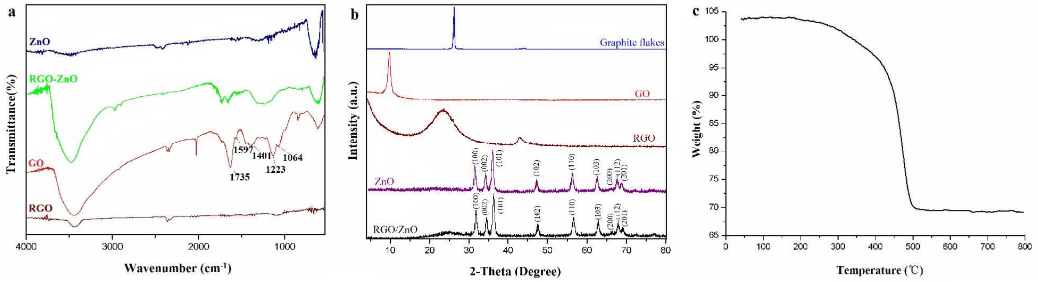

Figure 2 (a) FT-IR spectra and (b) XRD pattern of GO, RGO, ZnO and RGO-ZnO nanocomposites; (c) TGA curve of the RGO-ZnO nanocomposites.

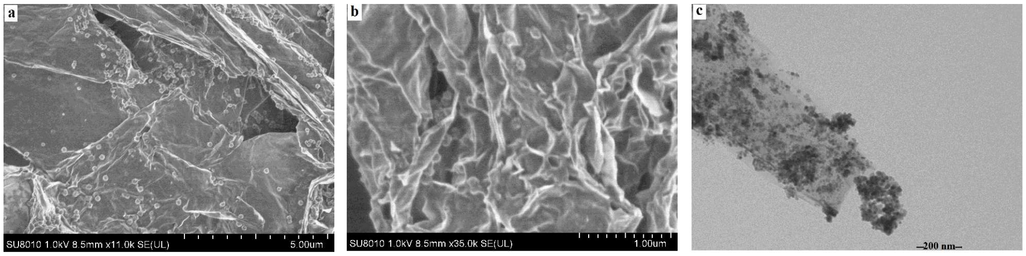

Figure 3 (a) Low-magnification SEM images; (b) high-magnification SEM images; (c) TEM image of RGO-ZnO nanocomposites.

shows weight loss from room temperature to 250 °C which may be attributed to the desorption of surface bound water.[31] An abrupt weight loss that occurs from 250 to 700 °C could be due to the removal of oxygen-functional groups and the decomposition of carbon framework from the nanocomposites.[32] According to the TGA analysis, the content of RGO is about 33.9 wt%, which are calculated from the weight loss of 250 to 700 °C.

The morphologies of RGO-ZnO were characterized by SEM and TEM. After ultrasonic with ethanol, the SEM and TEM images of RGO-ZnO were displayed that ZnO nanoparticles attached onto the surface of RGO sheets and no ZnO nanoparticles falling off from RGO sheets as seen in Figure 3, suggesting the ZnO nanoparticles grown onto the surface of RGO sheets robustly. The RGO sheets look like transparent veil decorating ZnO nanoparticles (Figure 3a), implying that the RGO sheets are quite thin. The interlayers of the RGO sheets are continuous folded and winkled surfaces (Figure 3b), avoiding the restack of RGO sheets. Figure 3c shows the TEM image of ZnO particles on the RGO sheets are nearly spherical in shape and with a typical size of about 50 nm.

3.2 Evaluation of the RGO-ZnO as DSPE adsorbent

Several parameters that could influence the DSPE efficiency, such as the amount of the RGO-ZnO, extraction time, the pH of sample solution, as well as type and volume of eluent solvent, were investigated and optimized in this work. Extraction recovery (*R*) was used to evaluate the extraction performance. *R* is expressed as follows:

$$R = \frac{CV}{C_0V_0} \times 100\%$$

Where *C* is the analyte concentration (ng mL^{-1}) in the reconstituted solvent, C_0 is the initial concentration of analyte in water sample. *V* and V_0 are the volumes of the reconstituted solvent and water sample, respectively.

3.2.1 The sorption capacity of RGO-ZnO for OPs

The sorption capacity is a key factor for a good adsorbent, since it determines the high enrichment factor. Firstly, the sorption capacity of RGO-ZnO for OPs was simply evaluated. The centrifuge tube was added with 30 mg RGO-ZnO and 5 μg of each analyte in 40 mL aqueous solution, and then the mixture was shaken on a platform shaker for 20min. After centrifugation no analytes were detected in the supernatant, implying a good sorption capacity for OPs.

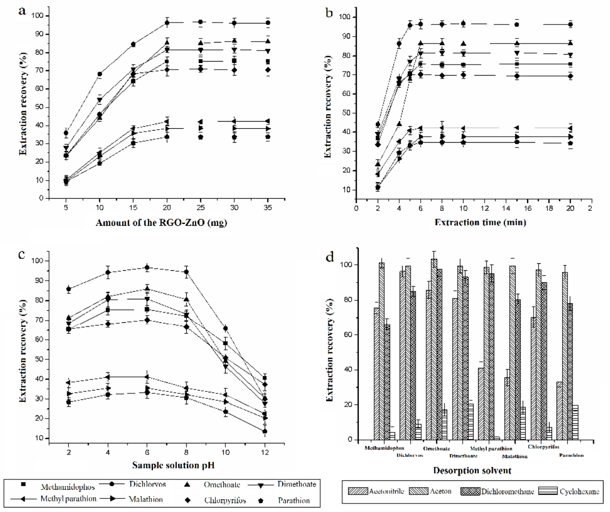

Figure 4 Effect of extraction conditions on DSPE efficiency of the eight pesticides. (a) Effect of the amount of RGO-ZnO; (b) effect of extraction time; (c) effect of the sample solution pH; (d) effect of the desorption solvent.

3.2.2 Effect of the amount of the RGO-ZnO nanocomposites

The amounts of RGO-ZnO in the range from 5 to 35 mg in 40 mL aqueous solution spiked with 50 ng mL^{-1} of each analyte were investigated to extract OPs, when acetonitrile (5 mL, three times) was used as elution solvent. It can be seen from Figure 4a, the extraction recovery of OPs increased as the amount of adsorbent increased (up to 20mg), with further increase in the adsorbent amount extraction recovery almost remained constant. According to the result, 20 mg of RGO-ZnO was selected.

3.2.3 Effect of extraction time

In DSPE procedure, extraction time is an important parameter, since they influence the efficiency of extraction. In this work, the extraction time in the range of 2-20 min, when 40mL aqueous solution spiked with 50 ng mL^{-1} of each analyte was used as the sample solution, acetonitrile (5 mL, three times) was used as elution. As shown in Figure 4b, the extraction recoveries of the analytes were increased with increased extraction time from 2 to 6 min and then no significant increase was observed after 6 min. Hence, the extraction time was employed in the following studies.

3.2.4 Effect of pH of the sample

For samples, the pH is also a key factor affecting the physicochemical properties of the analytes as well as the surface properties of the adsorbent during the adsorption procedure. Hence, the influence of sample pH on the extraction recoveries of OPs was investigated in the pH range from 2.0 to 12.0 by added hydrochloric acid or sodium hydroxide solutions. As shown in Figure 4c, the highest extraction recoveries were obtained when the sample pH was 6.0, while a significant decrease with the further increased the pH value (≥8.0). This was probably due to the instability of the OPs under alkaline solution. Therefore, the sample solution pH was adjusted to 6.0 before extraction.

3.2.5 Effect of washing solution

In this work, the effect of washing solution was also evaluated. After extraction, the RGO-ZnO adsorbents were washed by 10 mL of water. According to the results, no obvious variations in the recoveries of OPs were observed. However, washing with water make the DSPE procedure more complicated. Hence, washing solution was not selected in this work.

3.2.6 Effect of elution solvent

Selection of the kind of elution solvent is of vital importance for the extraction efficiency of the analytes. In this work, four types of eluent solvents with different polarity including acetonitrile, acetone, dichloromethane and cyclohexane were tested. The adsorbent loaded 40 mL sample solution with 2μg spiked of each analyte, then eluted with 5 mL (each time) of elution solvent three times. According to the results shown in Figure 4d cyclohexane had poor eluting capability compared with the other elution solvents. Acetonitrile improved the extraction recoveries, but it had poor eluting ability for the methyl parathion, malathion and parathion.

Table 1 Parameters of the proposed method for the eight pesticides.

Pesticides	Linear range (ng g^{-1})	R^2	LODs (ng g^{-1})	LOQs (ng g^{-1})	Intra-day RSDs (%)	Inter-day RSDs (%)
Methamidophos	0.5-200.0	0.9972	0.03	0.10	4.6	8.6
Dichlorvos	0.5-200.0	0.9903	0.03	0.10	5.2	7.5
Omethoate	0.5-200.0	0.9894	0.03	0.17	4.9	7.4
Dimethoate	0.5-200.0	0.9934	0.05	0.18	3.7	7.9
Methyl parathion	0.5-200.0	0.9982	0.05	0.15	5.6	8.3
Malathion	0.5-200.0	0.9975	0.05	0.18	4.2	8.6
Chlorpyrifos	0.5-200.0	0.9983	0.01	0.05	3.0	7.5
Parathion	0.5-200.0	0.9970	0.05	0.17	6.7	8.2

Table 2 Comparison of the proposed method with other methods for the determination of OPs.

Methods	Samples	Extraction time (min)	Linear range (ng g^{-1})	LODs (ng g^{-1})	Precision RSDs (%)	Ref.
Graphene-based SPE-GC-MS	Apple juices	16.7	0.5-200 ng mL^{-1}	0.04-0.35	3.3–9.2	20
MSPD-GC-NPD[a]	Fruit juices	15	5-100 ng mL^{-1}	10-200	1.1- 4.2	32
MIP[b]-MSPD-GC	Apple, pear	4.0h	-	0.3-1.6	1.2–4.8	33
MSPE-GC-MS	Tomato, rape	20	1-100	0.005-0.03	4.4-12.5	34
DSPE-GC-MS	Peanut oil	10	5-200 ng mL^{-1}	0.7-1.6	2.68-8.48	35
QuEChERS-GC-FPD[c]	Morinda roots		20-1280	5-20	1.01-9.26	36
SDME[d]-GC-FPD	Orange juice	15	10-500 ng mL^{-1}	0.98-2.2	6.3-14.3	37
DSPE-GC-MS	Apple, cucumber, water	6	0.5-200	0.01-0.05	3.7-8.6	This study

[a] NPD: nitrogen phosphorus detector
[b] MIP: molecularly imprinted polymer
[c] FPD: flame photometric detector
[d] SDME: single-drop microextraction

Table 3. Recoveries of real samples spiked with the target analytes (*n* = 5)

Pesticides	Water			Apple			Cucumber		
	Spiked (ng g^{-1})	Recovery (%)	RSD (%)	Spiked (ng g^{-1})	Recovery (%)	RSD (%)	Spiked (ng g^{-1})	Recovery (%)	RSD (%)
Methamidophos	0.0	nd[a]		0.0			0.0		
	1.0	79.1	9.6	1.0	75.7	9.3	1.0	80.0	10.6
	10.0	93.7	4.6	10.0	90.2	6.1	10.0	93.3	6.3
Dichlorvos	0.0	nd[a]		0.0			0.0		
	1.0	86.2	8.2	1.0	83.5	8.9	1.0	83.2	8.2
	10.0	102.2	5.2	10.0	94.6	6.0	10.0	95.6	4.9
Omethoate	0.0	nd[a]		0.0			0.0		
	1.0	82.1	8.9	1.0	82.4	8.3	1.0	103.2	11.1
	10.0	95.2	4.9	10.0	89.7	7.6	10.0	89.3	6.7
Dimethoate	0.0	nd[a]		0.0			0.0		
	1.0	88.5	8.7	1.0	104.2	10.7	1.0	89.0	13.3
	10.0	96.1	3.7	10.0	93.9	9.1	10.0	93.1	9.2
Methyl parathion	0.0	nd[a]		0.0			0.0		
	1.0	86.4	11.7	1.0	80.6	12.2	1.0	83.9	10.6
	10.0	95.3	5.6	10.0	95.4	8.0	10.0	96.1	8.2
Malathion	0.0	nd[a]		0.0			0.0		
	1.0	88.2	9.6	1.0	82.3	11.2	1.0	85.7	10.7
	10.0	97.9	4.2	10.0	94.3	4.1	10.0	93.7	6.6
Chlorpyrifos	0.0	nd[a]		0.0			0.0		
	1.0	82.8	12.3	1.0	78.7	10.8	1.0	75.0	11.7
	10.0	97.3	3.0	10.0	95.1	7.5	10.0	95.5	8.9
Parathion	0.0	nd[a]		0.0			0.0		
	1.0	79.1	8.3	1.0	77.0	9.0	1.0	78.8	8.1
	10.0	102.5	6.7	10.0	89.8	6.6	10.0	90.2	5.3

[a] nd: not detected.

66.3% and 78.2% for methamidophos and parathion. When using acetone as the eluent, the highest extraction recoveries in the range from 95.8% to 103.4% were obtained. Thus, acetone was chosen as elution solvent. The effect of elution solvent volume on the extraction recovery was also investigated. It was found that the extraction with 1.0 mL acetone three times (1.0 mL × 3) could completely elute the anlytes from the adsorbent.

3.3 Reusability of the adsorbent

In order to investigate the reusability of RGO-ZnO, the adsorbent was washed with acetone twice (each time vortex for 1 min with 5 mL acetone) and with 5 mL water after desorption. Then the adsorbent was reused for the next DSPE of the OPs according to the section 2.6. The extraction recoveries of the first cycle and the tenth cycle were varied from 96.1% to 103.3% and 85.4% to 97.8% ， respectively. It suggested that the adsorbent can be reused at least 10 times without significant decrease of the adsorption capacity.

3.4 Validation of the DSPE-GC-MS method

Under the optimal experimental conditions, several important parameters including the linearity, precision, limit of detection (LOD), and limit of quantification (LOQ) of the proposed method were studied. Pesticide-free water was used as blanks for the method validation. To establish the calibration curve, a series of sample solutions containing OPs from 0.5 ng g^{-1} to 200 ng g^{-1} were prepared. The linear ranges and correlation coefficients were summarized in Table 1. Good linearities were obtained for the analytes with correlation coefficients (R^2) of all better than 0.9894 in the responding linear range. The LOD and LOQ values were calculated at a signal-to-noise ratio (S/N) of 3 and 10, which were in range from 0.01 to 0.05 ng g^{-1} and from 0.05 to 0.18 ng g^{-1}, respectively. Moreover, precision of the proposed method was evaluated as intra-day and inter-day precision by analyzing the spiked samples at 10 ng g^{-1} of each anlytes in the same day and on three consecutive days. The precision results, expressed as the relative standard deviation (RSD) of peak areas, are shown in Table 1. It is found that the acquired RSD values were less than 8.6% for both intra-day and inter-day precision. Thereby, the proposed method was reliable.

Table 2 compares the performance of the proposed method in this study and other previously reported methods. The results show that the DSPE method has a shorter extraction time, wider linearity and a comparable reproducibility in contrast with the methods mentioned above. In addition, this method has lower LODs compared with the other method except for MSPE. These results confirmed that this method is faster, more sensitive and efficient.

3.5 Analysis of real samples

The proposed method was applied to the determination of OPs in apples, cucumbers and water. The results are summarized in Table 3, it can be seen no residues of the pesticides were detected in these samples. To estimate the effect of the matrices, all of the samples were spiked at 1 and 10 ng g^{-1} concentration levels of OPs to determine the recovery of the targeted analyte. For each concentration level five replicates were performed and the relevant data are listed in Table 3. The satisfactory recoveries of OPs from three real samples were in the range from 75.0% to 104.2% with RSDs between 3.0% and 13.3%. As can be seen, these real sample matrices had little interference with the performance of RGO-ZnO and it can also be concluded that the proposed method is highly precise and efficient.

4 Conclusions

In this work, the developed DSPE-GC-MS method is provided a sensitive and efficient approach for the determination of the OPs in apples, cucumbers and water samples by using RGO-ZnO nanocomposites as sorbent. RGO-ZnO nanocomposites were synthesized via a simple one-step hydrothermal strategy. This material can avoid the aggregation of graphene sheets and maintain the advantageous characteristics of graphene. Moreover, the ZnO nanoparticles endow the material with effective adsorption of water-solube analytes as well as facile separation of RGO-ZnO from water after adsorption by centrifugation. As a good adsorbent RGO-ZnO exhibited excellent extraction capability for OPs with low detection limit and satisfactory recoveries in aqueous samples. The results suggest that RGO-ZnO has a potential application in the determination of some other trace analytes.

References

1 D. Du, J.Wang, J. N. Smith, C. Timchalk and Y. Lin, *Anal. Chem.,* 2009, **81**, 9314.
2 G. Ding, P. Wang, Y. Tian, J .Zhang, Y. Gao, X. Wang, R. Shi, G. Wang and X.Shen, *Environ. Sci. Technol.* 2012, **46** 2911.
3 H. Naeeni, Y. Yamini and M. Rezaee, *J. Supercrit. Fluids,* 2011, **57**, 219.
4 G. Pagliuca, T. Gazzotti, E. Zironi and P. Sticca, *J. Chromatogr. A*, 2005, **1071**, 67.
5 G. Cinelli, P. Avino, I. Notardonato and M.V. Russo, *Anal. Methods,* 2014, **6**, 782.
6 I. Notardonato, P. Avino, G. Cinelli and M.V. Russo, *RSC Advances,* 2014, **4**, 42424.
7 M.V. Russo, L. Campanella and P. Avino, *J. Chromatogr. B*, 2002, **780**, 431.
8 J.Y. Shim, Y.A. Kim, E.H. Lee, Y.T. LEE and H. LEE, *J. Agric. Food Chem.,* 2008, **56**, 11551.
9 D. Liu, W. Chen, J. Wei, X. Li, Z. Wang and X. Jiang, *Anal. Chem.,* 2012, **84**, 4185.
10 P.S. Chen and S.D.Huang, *Talanta,* 2006. **69**, 669.
11 C. Molina, M. Honing and D. Barcelo *Anal. Chem.,* 1994**, *66***, 4444.
12 W. Chen, K. Poon and H.W. Michael, *Environ.Sci.Technol.,* 1998, **32**, 3816.
13 D.A. Lambropoulou and T. A. Albanis, *J. Agric. Food Chem.*, 2002, **50**, 3359.
14 B. Albero, C.S. Brunete, J.L. Tadeo, *J. Agric. Food Chem.,* 2003, **51**, 6915.
15 C. Bicchi, C.Cordero, C. Iori, P. Rubiolo, P. Sandra, J. H. Yariwake and V.G. Zuin, *J. Agric. Food Chem.*, 2003, **51**, 27.
16 S. Yan, T. Qi, D. Chen, Z. Li, X. Li, S. Pan, *J. Chromatogr. A,* 2014, **1347,** 30.
17 M. Anastassiades, S.J. Lehotay, D. Stajnbaher and F.J. Schenck, *J. AOAC Int.* 2003, **86**, 412.
18 I. P. Roman, A. Chisvert and A. J. Canals, *J. Chromatogr. A,* 2011, **1218**, 2467.
19 Q. Liu, J. Shi, L. Zeng, T. Wang, Y. Cai and G. J. Jiang, *J. Chromatogr. A,* 2011, **1218**, 197.
20 Q. Han, Z. Wang, J. Xia, X. Zhang H. Wang and M. Ding, *J. Sep. Sci.,* 2014, **37**, 99.
21 Q. Liu, J.B. Shi, J.T. Sun, T. Wang, L.X. Zeng and G.B. Jiang, *Angew. Chem. Int. Ed.*, 2011, **50**, 5913.
22 J. Wang, T. Tsuzuki, B. Tang, X. Hou, L. Sun and X. Wang, *ACS Appl. Mater. Interfaces,* 2012 , **4**, 3084.
23 P.Roy, A.P. Periasamy, C. Liang and H. Chang, *Environ. Sci. Technol.,* 2013, **47**, 6688.
24 S. Liu, H. Sun ,A. Suvorova and S. Wang, *Chemical Engineering Journal,* 2013, **229**, 533.
25 D. Fu, G. Han, F. Yang, T. Zhang, Y. Chang and F. Liu, *Applied Surface Science,* 2013, **283**, 654.
26 W. S. Hummers and R. E.Offeman, *J. Am. Chem. Soc.*, 1958, **80**, 1339.
27 V.C. Tung, L.M. Chen, M.J. Allen, J.K. Wassei, K. Nelson, R.B. Kaner, and Y. Yang, *Nano Lett.,* 2009, **9**, 1949.
28 W. F.Chen and L. F.Yan, *Nanoscale,* 2010, **2**, 559.
29 C. Zhu, S. Guo, Y. Fang and S. Dong, *ACS Nano,* 2010, **4**, 2429.
30 H.R. Pant, C.H. Park,P. Pokharel, Leonard D. Tijing, D.S. Lee and C. S. Kim, *PowderTechnology,* 2013, **235**, 853.
31 J.L. Wang, Z.X. Shi, J.C. Fan, Y. Ge, J. Yin and G. X. Hu, *J. Mater. Chem.,* 2012, **22**, 22459.
32 C. Bosch-Navarro, E. Coronado, C. Martí-Gastaldo, J. F. Sánchez-Royo and M. G. Gòmez, *Nanoscale,* 2012, **4**, 3977.
33 X. Wang, X. Qiao, Y. Ma, T. Zhao and Z. Xu, *J. Agric. Food Chem.,* 2013, **61**, 3821.
34 L. Wang, X. Zang, Q. Chang, C. Wang and Z. Wang, *Anal. Methods,* 2014, **6**, 253.
35 R. Su, X. Xu, X.H. Wang, D. Li, X.Y. Li, H.Q. Zhang and A.M. Yu, *J. Chromatogr. B,* 2011, **879**, 3423.
36 H.M. Liu, W.J. Kong, Y. Qi, B. Gong, Q. Miao, J.H. Wei and M.H. Yang, *Chemosphere,* 2013, **95**, 33.
37 E. Zhao, L. Han, S. Jiang, Q. Wang, Z. Zhou, *J. Chromatogr. A,* 2006, **1114,** 269.