

## Reduced Graphene Oxide-Cadmium Sulfide Quantum Dots Nanocomposite Based Dispersive Solid Phase Microextraction for Ultra-Trace Determination of Carbamazepine and Phenobarbital

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This research presents a fast, sensitive, and selective ultrasound-assisted dispersive solid phase microextraction technique for simultaneous preconcentration and determination of ultra-trace amount of carbamazepine and phenobarbital. Reduced graphene oxide sheets decorated with cadmium sulfide quantum dots was synthesized, characterized, and used as a high capacity adsorbent. A high performance liquid chromatography with UV detector (HPLC-UV) instrument with 58:42 composition of methanol:acetic acid/acetate buffer (pH = 5; 0.05 mol L<sup>-1</sup>) as the mobile phase and set on the wavelength of 230 nm was used to separate and quantify the analytes. In this regard, different parameters affecting adsorption and desorption of the analytes on the surface of the nanocomposite were studied and optimized to maximize the efficiency of the method. The method was linear in the ranges of 0.5-180 and 0.5-140 ng mL<sup>-1</sup> (correlation coefficient (r) > 0.999) with limits of detection of 0.19 and 0.24 ng mL<sup>-1</sup> for carbamazepine and phenobarbital, respectively. Eventually, to evaluate the efficiency of the proposed method for the determination of pharmaceuticals in biological samples, different real samples including breast milk, urine and human plasma were tested. Obtained recoveries values were within the range of 96.3 ± 2 to 103.7 ± 3.3% which showed satisfactory efficiency.

**Keywords:** cadmium sulfide quantum dots, reduced graphene oxide, microextraction, preconcentration, carbamazepine, phenobarbital

### Introduction

Ultra-trace determination of pharmaceuticals, particularly in different food, beverage, environmental and biological samples with a variety of matrices, is an attractive field of research for many applications such as medical and pharmaceutical research.<sup>1</sup> Up to now, lots of effort has been made to propose highly efficient methods to preconcentration and determination of the low concentration of different drugs and pharmaceuticals. Solid-phase microextraction (SPME) is one of the most powerful techniques for the mentioned application which provides a simple preconcentration procedure that reduces the requirement for complicated equipment and also organic solvents consumption.<sup>2</sup> At the moment, several SPME's variations are available such as in-tube, fiber, and dispersive solid-phase microextraction (DSPME).<sup>3</sup> Among them, DSPME is one the simplest and cheapest

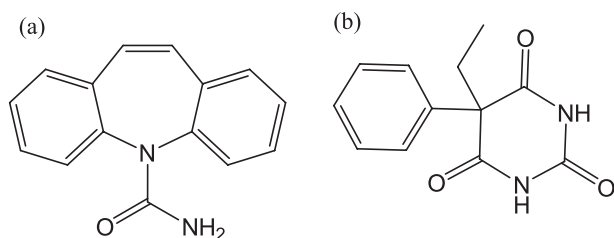
method using disperse solid adsorbents to extract and enrich analytes from samples.

In order to achieve outstanding analytical features for DSPME, choosing the adsorbents is a key parameter.<sup>4</sup> The adsorbent should provide the requirements for a good sample preconcentration, such as high adsorption efficiency and easy desorption of pre-adsorbed analyte.<sup>5</sup> Therefore, great efforts are continuously being made to develop new materials for this purpose, i.e., graphene oxide (GO), reduced graphene oxide (RGO), magnetic nanomaterial,<sup>6</sup> semiconductor nanoparticles,<sup>7</sup> etc. Carbon-based nanomaterials are well known as high potential adsorbents and they are frequently used as the main adsorbent or as a support material to enhance the adsorption capacity of different nanoparticles.<sup>8</sup> Quantum dots (QDs) owing to their unique properties, for instance size-dependent fluorescence and small size, has been widely used in different research areas. It is expected that QDs show a great potential to facilitate a successful extraction owing to their high surface area to volume ratio as well as their chemical composition.<sup>9,10</sup>

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Cadmium sulfide quantum dots (CdS QDs) is mostly used as fluorescence probe or catalyst for photodegradation of drugs and pollutant due to its optical properties.<sup>11,12</sup> However, in both cases, adsorption of the analyte to the CdS QDs is the critical step.<sup>13,14</sup> Therefore, decorating the large surface area of RGO sheets with tiny cadmium sulfide to make RGO-CdS QDs nanocomposite can significantly increase the surface area and adsorption site which leads to the DSPME efficiency improvement. Besides, the simplicity of synthesis of this nanocomposite through the facile hydrothermal approach is an asset, which makes the proposed technique simpler, cheaper, and more environmentally friendly compared to the other reported technique such as liquid-liquid microextraction.

Carbamazepine (CBZ, Figure 1a) and phenobarbital (PB, Figure 1b) are anticonvulsant agents, which are commonly used as anti-epileptic drugs (AEDs). Both are listed as essential medicines by the World Health Organization (WHO) and recommended for treating epilepsy especially in developing countries. CBZ is also used to treat schizophrenia and neuropathic pains. PB has a wider range of application for verities of surgery as a painkiller and treat anxiety and trouble sleeping. PB was also introduced as a safe alternative to diazepam to treat the delirium tremens. However, both are long-acting drugs that are slowly absorbed with frequently reported cases of toxicity and overdose.<sup>15-17</sup> Also, behavioral side effects and persistent reduction in intelligence quotient have been observed in several studies<sup>18,19</sup> focusing on children or infant of the lactating mother, which PB was prescribed as their treatment, since PB can easily cross the placenta and be excreted into breast milk.<sup>17</sup> Therefore, a technique with the ability to assay trace concentration of PB and CBZ in biological fluids is needed. Up to now different methods and procedure have been reported<sup>18,20-26</sup> for determination of PB and/or CBZ individually, together or along with other drugs, which were unable to show great analytical performance or suffered from other limiting factors, i.e., complicated procedures, requiring high volume of organic solvents, time-consuming, and costly.



**Figure 1.** Chemical structure of (a) carbamazepine and (b) phenobarbital.

In this present research, a simple, cheap, and rapid dispersive solid-phase microextraction (DSPME) with the aid

of ultrasonic was proposed for simultaneous determination of phenobarbital and carbamazepine. A high performance liquid chromatography with a UV detector (HPLC-UV) was used for analysis of the samples. Also, the easily obtainable and high capacity reduced graphene oxide-cadmium sulfide quantum dots (RGO-CdS QDs) nanocomposite was used as an adsorbent for DSPME. The optimized method was used to preconcentrate the abovementioned analytes in breast milk, urine, and human plasma.

## Experimental

### Reagents and solutions

All chemicals used in this research were of analytical grade and used without any further purification. Carbamazepine and phenobarbital analytical standards were obtained from Sigma-Aldrich (Steinheim, Germany). Graphite powder, hydrogen peroxide, potassium permanganate, sodium hydroxide, cadmium acetate ( $\text{Cd}(\text{CH}_3\text{COO})_2 \cdot 2\text{H}_2\text{O}$ ), dimethyl sulfoxide, boric acid, glacial acetic acid, phosphoric acid, and sulfuric acid were purchased from Merck (Darmstadt, Germany). HPLC grade methanol was purchased from Daejung Chemicals (Siheung, Korea). The drug-free human plasma sample was obtained from the Iranian Blood Transfusion Organization (IBTO) Damghan branch (Damghan, Iran). A volunteered healthy lactating woman kindly donated the human breast milk sample. All procedures were in accordance with the ethical standard of Damghan University's ethical committee. Urine samples were collected from a healthy male volunteer (aged 28) prior to having breakfast. All the real samples were frozen ( $-20^\circ\text{C}$ ) in sterile condition until analysis.

Double distilled water was used to prepare all the solutions. Stock solutions of CBZ and PB ( $100\text{ mg L}^{-1}$ ) were prepared by dissolving 10.0 mg of the compounds in 100 mL methanol and water, respectively, and kept at  $4^\circ\text{C}$ . Daily dilution of these solutions provides the working solutions. Britton-Robinson buffer was prepared by adding 53.5 mL NaOH ( $0.2\text{ mol L}^{-1}$ ) into a mixture solution containing  $0.04\text{ mol L}^{-1}$  of  $\text{H}_3\text{BO}_3$ ,  $\text{CH}_3\text{COOH}$ , and  $\text{H}_3\text{PO}_4$  to adjust the pH to 7 in a total volume of 100 mL.<sup>27</sup> The acetic acid/acetate ( $0.05\text{ mol L}^{-1}$ ) buffer was prepared by the addition of  $0.1\text{ mol L}^{-1}$  NaOH solution to 50 mL  $0.1\text{ mol L}^{-1}$  of  $\text{CH}_3\text{COOH}$  to adjust pH = 5.0 and the mixture was brought up to 100 mL with double distilled water.

### Instruments

A Knauer (Berlin, Germany), Smartline high-

performance liquid chromatography equipped with a Smartline UV Detector 2500 and Rheodyne six-port injector valve with a 20  $\mu\text{L}$  loop was used for analysis. Chromatographic separation was carried out on an Eurospher 100-5 C18, 250  $\times$  4.6 mm column (Knauer, Berlin, Germany). ChromGate chromatography software was used to process the data. Chromatographic conditions of analysis were selected based on our last published method<sup>25</sup> with some modifications and its determined to be 58:42 composition of methanol to acetic acid/acetate buffer (pH = 5; 0.05 mol L<sup>-1</sup>). Afterward, to select the detector wavelength, a UV-Vis measurement was performed on each analyte solved in the mobile phase, and a single wavelength with an acceptable response for both analytes has been selected ( $\lambda = 230$  nm).

A D8 advanced X-ray diffractometer (XRD, Bruker, Hamburg, Germany) with a Cu K $\alpha$  radiation source (35 kV, 30 mA, and  $\lambda = 0.1542$  nm) was used. Chemical analysis and morphological study of RGO-CdS QDs and RGO were investigated by using an electron dispersive X-ray spectroscopy (EDX) system attached to a field emission scanning electron microscopy (FESEM) Tescan Mira 3 XMU (TESCAN, Brno, Czech Republic) at an accelerating voltage of 15 KV. A 780 Metrohm pH meter (Herisau, Switzerland), a Z-300 Hermle centrifuge (Wehingen, Germany), and a DT510H Bandelin ultrasonic bath (Vorpommern-Greifswald, Germany) were used also in this research.

#### Preparation of RGO-CdS QDs nanocomposite

A one pot, simple and facile synthesis approach for simultaneous reducing the graphene oxide and depositing CdS QDs on it was applied. The GO nanosheets were synthesized from graphite powder by the Hummers' method<sup>28,29</sup> (brief details are given in Supplementary Information (SI) section). The synthesis of RGO-CdS QDs was followed by mixing 80 mg of obtained GO along with 0.212 g Cd(OAc)<sub>2</sub>·2H<sub>2</sub>O into 50 mL dimethyl sulfoxide (DMSO) and sonication (40 kHz, 500 W) for 30 min. Then, the mixture was transferred to a Teflon-lined stainless-steel autoclave and heated at 180 °C for 12 h and centrifuged (5000 rpm, 10 min) after cooling. The precipitated was washed several times with acetone and then methanol to remove any residual compounds. Finally, the obtained powder was dried at 80 °C for 1 h. In order to ascertain the successful reduction of GO and synthesis of RGO-CdS QDs, all steps for the preparation of nanocomposite were also carried out on the graphene oxide without the presence of cadmium acetate.

#### Microextraction procedure

The following steps were performed to extract and determine CBZ and PB: (i) appropriate amount of standard solutions (100 mg L<sup>-1</sup>) of CBZ and PB along with 2 mL of Britton-Robinson buffer (pH 7.0) were added to a 10 mL volumetric flask and made up to the volume; (ii) 7.5 mg of RGO-CdS QDs was added to the solution and left it to adsorb the analytes with the assistance of ultrasound for 120 s; (iii) the pre-adsorbed analytes onto the RGO-CdS QDs were precipitated with the aid of centrifuge (60 s, 3000 rpm) and the supernatant solution was decanted; (iv) 100  $\mu\text{L}$  of methanol was added to the adsorbed nanomaterial as an eluent to desorb the pre-adsorbed analytes with the help of ultrasound for 180 s; (v) finally, the eluent was isolated from the sorbent by the centrifuging (60 s, 3000 rpm), then 20  $\mu\text{L}$  of the supernatant was directly injected into the HPLC-UV. The peaks area at the retention times of 4.4 and 6.1 min were measured to evaluate the extracted PB and CBZ, respectively.

#### Real samples

The urine sample was analyzed directly as followed: 1 mL of the urine sample was spiked with analytes (to make a final concentration of 0, 30, 80, 130 ng mL<sup>-1</sup>) and centrifuged for 5 min at 4000 rpm. Analysis continued with adding 2.0 mL of Britton-Robinson buffer (pH = 7) to the supernatant and made up to 10.0 mL with double distilled water. Afterward, the optimized DSPME procedure was performed as previously described.

Breast milk and human plasma samples need to be deproteinized before analysis. For this purpose, 1 mL of acetonitrile was added to equal volume of spiked samples (1 mL) with different concentrations (within the linear range of the calibration curve) of analytes (to make a final concentration of 0, 30, 80 and 130 ng mL<sup>-1</sup> for milk and 0, 10, 40, 90 ng mL<sup>-1</sup> for plasma). The collected supernatant after centrifuge (4.0 min, 4000 rpm) were heated under a stream of nitrogen at 50 °C to reach half of the initial volume. The deproteinized milk and human plasma solutions were stored for further analysis.

For analysis of milk sample, 2 mL of Britton-Robinson buffer (pH = 7) was added to the abovementioned deproteinized solution and makeup to 10 mL with double distilled water, and the optimized DSPME procedure was applied. For human plasma, considering its severe matrix effects, a standard addition technique was applied for the determination of CBZ and PB. Hence, the obtained deproteinized plasma samples were mixed with 2 mL of Britton-Robinson buffer (pH = 7) in a 10 mL test tube and

then a standard addition method was applied by adding appropriate amount of analytes standard solutions and followed by the optimized DSPME procedure.

### Statistical analysis

In the optimization and evaluation process of the method, some usual statistical analysis was used in each experiment. All studies were performed in three replicates and the results presented as error bars by measuring the standard deviation (SD). In addition, to assure the obtained data in real samples studies, the results were appraised by relative standard deviation (RSD) and Student's *t*-test (at the 95% confidence level and two degrees of freedom).

## Results and Discussion

### Graphene-cadmium sulfide quantum dots nanocomposite characterization

The synthesized RGO-CdS QDs nanocomposite was characterized by different techniques including XRD, Raman, FESEM, and EDX and the results were compared to synthesized RGO. XRD pattern (Figure 2a) of RGO-CdS QDs shows three main peaks at the scattering angle of 26.65, 43.92, and 51.95° while RGO shows two main peaks at 24.29 and 43.16°. These results indicate that cadmium sulfide quantum dots decorated on graphene sheets in the blende form. By using the Scherrer equation, the average crystallite size of CdS quantum dots was calculated to be 5.27 nm. Furthermore, Raman spectrum of RGO-CdS QDs, RGO, and GO have been shown in Figure 2b. The typical D and G bands for graphene-based compounds appeared in 1475 and 1725 cm<sup>-1</sup> in GO profile and with a little shift and intensity decrement, due to the

chemical reduction, in RGO and RGO-CdS QDs profiles. Also, RGO-CdS QDs profile shows three peaks presented at 495, 910, and 1206 cm<sup>-1</sup> which correspond to the 1LO, 2LO, and 3LO, (first-, second-, and third-order longitudinal optical phonon modes), respectively.<sup>30</sup>

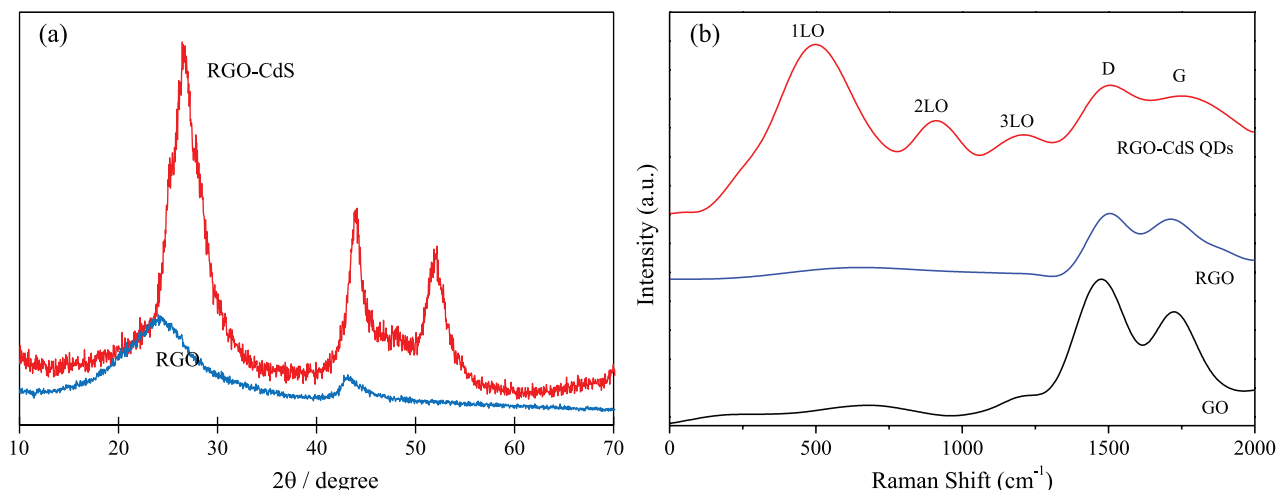
Morphological and chemical studies were also performed on RGO-CdS QDs and RGO samples by FESEM and EDX. FESEM pictures (Figure 3) obviously show the cadmium sulfide quantum dots on graphene expanded sheet and their size is almost the same as the estimated size calculated from the XRD pattern. EDX analysis (Figure 3) confirms the deposition of CdS quantum dots on the RGO. Additionally, to gain a better understanding of surface chemistry of RGO-CdS QDs, zeta potential analysis (Figure S1, SI section) in different pHs was performed which showed that adsorbent has a negative surface charge after pH = 5.

### Optimization of the microextraction condition

The proposed method was optimized with regard to desorption solvent, pH, type and amount of buffer, extraction time and amount of adsorbent using a univariate approach, to achieve the best extraction efficiency for determination of CBZ and PB.

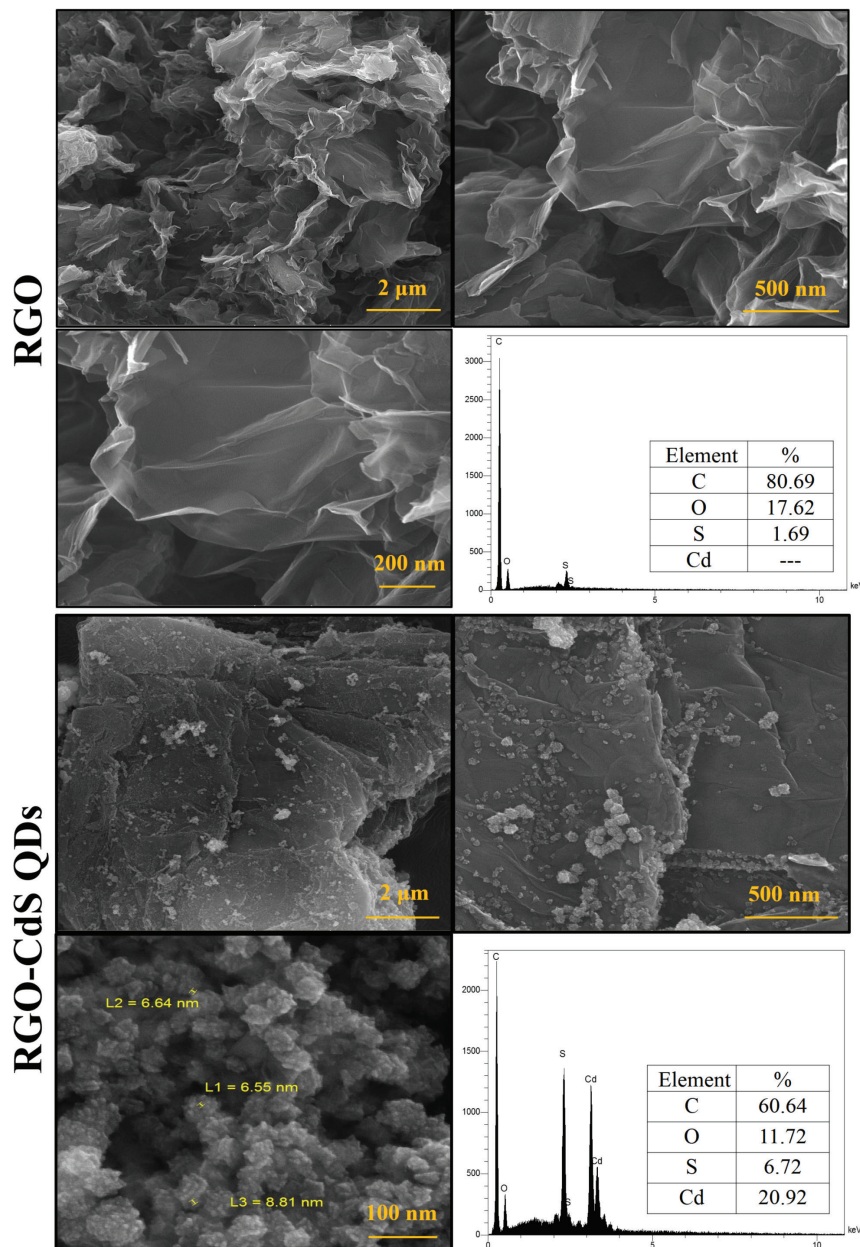
### Type and volume of eluent

Eluent has a huge influence on the efficiency of extraction. Therefore, a series of experiments was accomplished by using some common organic solvents such as acetonitrile, tetrahydrofuran, ethanol, and methanol. Based on the results (Figure 4a), among them, methanol has the maximum ability to desorb the analytes from the RGO-CdS QDs adsorbent. In further experiments the effect of the eluent volume was investigated in the range of 100-300 μL. A decrease in the analytical signal was



**Figure 2.** (a) XRD pattern and (b) Raman spectrum of synthesized nanomaterials.





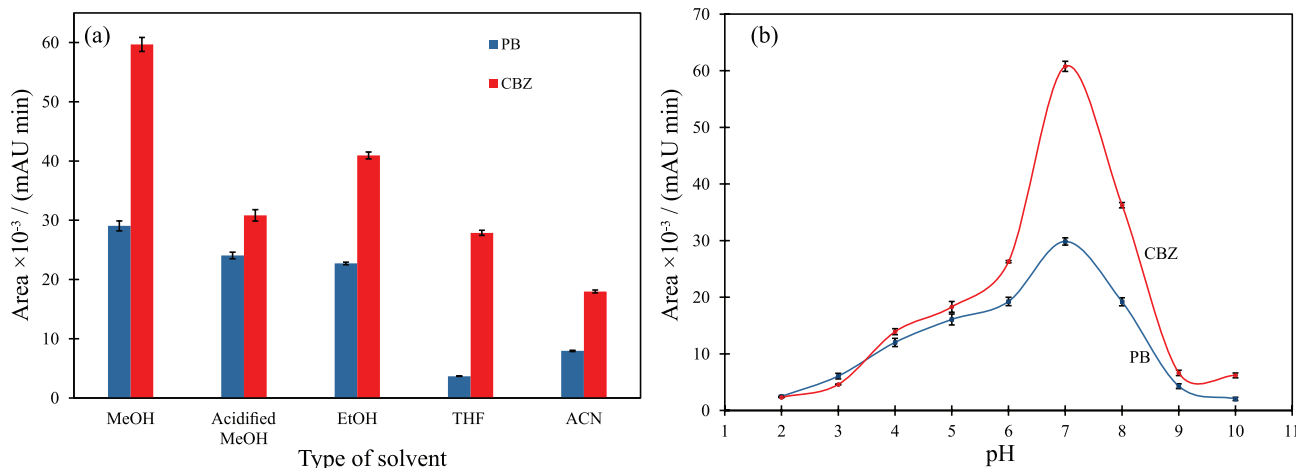
**Figure 3.** FESEM pictures and EDX analysis of synthesized RGO and RGO-CdS QDs.

observed with increasing the volume, and the volume of 100  $\mu\text{L}$  showed the best results. The volume lower than 100  $\mu\text{L}$  was not injected, due to the difficulty of the repeatable and accurate withdraw of eluent. Therefore, 100  $\mu\text{L}$  was chosen as the optimum volume.

#### Effect of pH and buffer

Influence of sample solution pH was investigated in the range of 2.0-10.0 owing to its impact on the adsorption step. As depicted in Figure 4b, for both analytes, the best results were observed by adjusting the pH at 7.0. Under these conditions, the nitrogen atom in the chemical structure of the analytes can interact with cadmium in RGO-CdS QDs

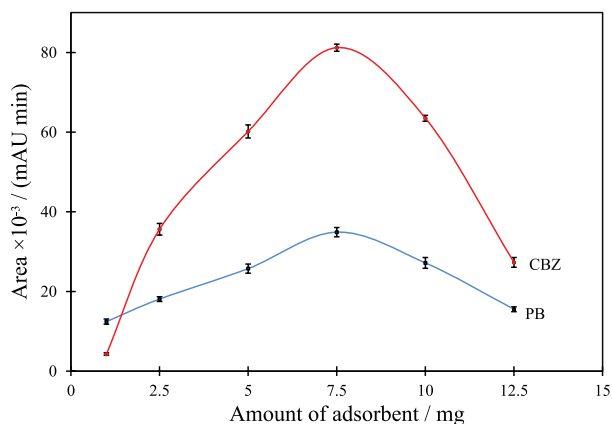
and adsorb on its surface. At  $\text{pH} < 7$ , by increasing the hydronium ion concentration the nitrogen atom in analytes chemical structure is protonated and the adsorption process disrupted. On the other hand, by an increment of hydroxyl ion concentration at  $\text{pH} > 7$ ,  $\text{OH}^-$  can react with cadmium in RGO-CdS QDs which can disrupt the analytes adsorbent interaction. Therefore, the value of  $\text{pH} = 7$  was selected as optimum for further experiments. Additional experiments on the type of buffer using phosphate buffer and Britton-Robinson buffer were carried out and according to the results, using 2 mL of Britton-Robinson buffer leads to achieve the better results, therefore this value was selected for the upcoming experiments.



**Figure 4.** Effect of (a) solvent type; (b) pH, on the extraction of analytes. Conditions: 10 mL solution of CBZ and PB (50 ng mL<sup>-1</sup>), 5 mg sorbent, 200  $\mu$ L methanol as eluent (error bars represent the SD, n = 3).

#### Effect of adsorbent amount

The influence of RGO-CdS QDs amount was studied in the range of 1.0-12.5 mg to achieve the highest extraction efficiency. Based on the obtained results (Figure 5) the extraction efficiency was increased by the increment of RGO-CdS QDs amounts from 1.0 to 7.5 mg and then decreased. As the amounts of RGO-CdS QDs increase, more analytes can adsorb by the adsorbent, whereas the higher amount of adsorbent also led to difficult desorption of analytes using a small volume of the eluent. Therefore 7.5 mg of RGO-CdS QDs was considered as the optimum value.



**Figure 5.** Effect of amount of adsorbent on the extraction. Conditions: 10 mL solution of CBZ and PB (50 ng mL<sup>-1</sup>), 2 mL of Britton-Robinson buffer (pH = 7), 200  $\mu$ L methanol as eluent (error bars represent the SD, n = 3).

#### Effect of ultrasonic time

The mass transfer phenomenon is a time-dependent process, so in order to obtain the best efficiency in preconcentration, the ultrasonic assisted adsorption and desorption time should be performed at the optimized time. The influences of adsorption time (the moment of RGO-CdS QDs addition until the end of sonication) and desorption

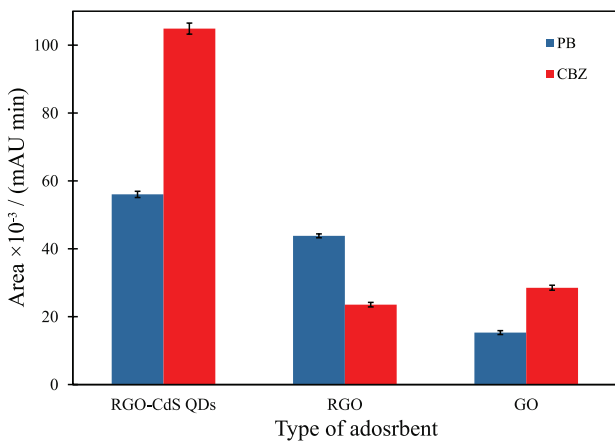
time (time interval from the injection of eluent to the loaded adsorbent to end of sonication) was studied in the range of 0-250 s. The obtained results (Figure S2, SI section) showed the best extraction efficiency was obtained at 120 and 150 s for adsorption and desorption time, respectively. These results demonstrated that the adsorption/desorption process is very fast due to the infinitely large surface area between dispersed solid RGO-CdS QDs which is in contact with the aqueous/organic phase.

#### Comparative study on adsorbents

To prove the advantage of RGO-CdS QDs over the GO and RGO as an adsorbent an equal amount (7.5 mg) of each adsorbent was added to the sample solutions and the optimum procedure was performed. The results (Figure 6) demonstrate the superiority of RGO-CdS QDs over the GO and RGO due to its surface modification by CdS quantum dots. The obtained results present the final analytical signal of the preconcentration method with different sorbents, which were influenced by efficiency of adsorption and desorption steps. Due to the greater hydrophobicity of RGO compared to other sorbents and the relatively higher hydrophobicity of CBZ, a stronger interaction between them can be formed in adsorption step. This interaction cannot be easily breakdown in the desorption step, which can explain the non-identical behavior of RGO over than other sorbents.

#### Analytical features

The proposed method under the optimized condition was evaluated by investigation of different analytical figure of merits, i.e., linearity, the limit of detection (LOD), precision (RSD in percentage), selectivity, enrichment factor (EF), and consumptive index (CI). A linear calibration curve



**Figure 6.** Comparative study on RGO-CdS QDs, GO and RGO as an adsorbent on the extraction of analytes. Conditions: 10 mL solution of CBZ and PB (50 ng mL<sup>-1</sup>), 2 mL of Britton-Robinson buffer (pH = 7), 7.5 mg sorbent, and 100 μL methanol as eluent (error bars represent the SD, n = 3).

obtained in the range of 0.5-180 and 0.5-140 ng mL<sup>-1</sup> with the regression equation of  $y = 1975.5x + 542.09$  ( $R^2 = 0.9994$ ) and  $y = 1064.9x + 114.65$  ( $R^2 = 0.9995$ ) for CBZ and PB, respectively, where  $y$  is the chromatogram peak area (mAU min) and  $x$  is the concentration of analytes (ng mL<sup>-1</sup>). The LOD values (signal-to-noise ratio (S/N) = 3.0, and eight replicates) were achieved to be 0.19 and 0.24 for CBZ and PB, respectively. The RSD (n = 8) of the method for 10 and 140 ng mL<sup>-1</sup> of CBZ and 10 and 100 ng mL<sup>-1</sup> of PB was obtained to be 2.8, 1.1, 3.2, and 1.9%, respectively. The slope ratio between the extracted and unextracted analytes concentration is defined as EF. This value was calculated to be 57 and 82 for CBZ and PB, respectively. The consumed sample volume (mL) to reach a unit of EF<sup>25</sup> is defined as CI which was found to be 0.175 and 0.121 mL for CBZ and PB, respectively.

#### Effect of foreign ions and compounds

A technique can be considered selective if the influence of matrix interferences on the results was negligible. Hence, the effect of some interfering ions and pharmaceutical compounds was investigated by analyzing the CBZ and PB solution (50 ng mL<sup>-1</sup>) containing foreign species at different concentration levels. The tolerance ratio is the maximum concentration of species, which causes a relative error of less than ± 5% in analytical signals. The results are presented in Table 1, which have shown an acceptable selectivity in the presence of manifold concentrations of interferences.

#### Determination of CBZ and PB in biological samples

Eventually to check the proficiency of the optimized

**Table 1.** Effect of interfering ions and compounds on preconcentration efficiency

Foreign species	Tolerance ratio
Al <sup>3+</sup> , Pb <sup>2+</sup> , Cd <sup>2+</sup> , Sn <sup>2+</sup> , Ca <sup>2+</sup> , Ag <sup>+</sup> , Na <sup>+</sup> , K <sup>+</sup> , S <sup>2-</sup> , SO <sub>4</sub> <sup>2-</sup> , Cl <sup>-</sup> , Br <sup>-</sup> , H <sub>2</sub> PO <sub>4</sub> <sup>-</sup> , CN <sup>-</sup> , <sup>a</sup> methyl dopa, ethambutol, fluvoxamine, caffeine, urea, glucose, glycine	1000
Cr <sup>3+</sup> , <sup>a</sup> Ni <sup>2+</sup> , <sup>a</sup> Cu <sup>2+</sup> , Fe <sup>2+</sup> , CN <sup>-</sup> , <sup>b</sup> I <sup>-</sup>	500
Cr <sup>3+</sup> , <sup>b</sup> Ni <sup>2+</sup> , <sup>b</sup> CO <sub>3</sub> <sup>2-</sup> , EDTA	250

<sup>a</sup>For CBZ; <sup>b</sup>for PB. EDTA: ethylenediaminetetraacetic acid.

DSPME procedure, three biological samples (urine, breast milk, and human plasma) were subjected to the proposed method for the determination of CBZ and PB. For this purpose, a detailed method was described in Experimental section. In brief, human plasma and breast milk should be deproteinized prior to the analysis, however, urine sample does not require any pretreatment. Additionally, determination of analytes in the plasma sample was studied with the aid of standard addition method. All measurements were done in three replicates and the obtained results were evaluated by SD and Student's  $t$ -test (95% confidence level). The results (Tables 2 and 3) show high efficiency of the optimized method to preconcentrate and determine the analyte in different concentration level and verities of matrix.

#### Comparison of the proposed DSPME method

The analytical performance of the proposed RGO-CdS QDs based DSPME technique was compared with some previously published solid-phase extraction methods for the determination CBZ and/or PB and details are summarized in Table 4. The comparison shows that the suggested method is comparable or even better than most of previously reported methods in the term of LODs, RSD, and the lowest quantifiable concentrations while using a smaller amount of adsorbent. Low adsorbent consumption in the procedure makes it an environmentally friendly and inexpensive method.

## Conclusions

In the present study, an ultrasound-assisted dispersive solid phase microextraction technique was presented for simultaneous preconcentration and determination of carbamazepine and phenobarbital. Graphene-cadmium sulfide quantum dot nanocomposite was synthesized, characterized and its application as a high capacity and powerful adsorbent was studied. The proposed method presents outstanding analytical features as well as other

**Table 2.** Determination of carbamazepine (CBZ) and phenobarbital (PB) in biological samples

Real sample	Phenobarbital				Carbamazepine			
	Concentration of PB / (ng mL <sup>-1</sup> )		RR / %	<i>t</i> -test	Concentration of CBZ / (ng mL <sup>-1</sup> )		RR / %	<i>t</i> -test
	Added	Found <sup>a</sup>			Added	Found <sup>a</sup>		
Breast milk	0	ND	–	–	0	ND	–	–
	30	31.1 ± 0.6	103.7	3.25	30	30.9 ± 1.7	103.1	0.95
	80	80.2 ± 2.5	100.3	0.17	80	78.6 ± 2.6	98.2	0.93
	130	129.8 ± 2.2	99.8	0.18	130	128.9 ± 3.3	99.2	0.57
Urine	0	ND	–	–	0	ND	–	–
	30	28.9 ± 1.0	96.3	1.8	30	29.9 ± 0.5	99.7	0.31
	80	80.3 ± 2.7	100.4	0.21	80	78.2 ± 2.8	97.7	1.1
	130	128.5 ± 2.0	99.8	1.3	130	127.7 ± 2.2	98.2	1.75

<sup>a</sup>Average of three measurements ± standard deviation. RR: relative recovery; r: correlation coefficient; ND: not detected.

**Table 3.** The determination of CBZ and PB in human plasma samples

Analyte	Concentration of analyte / (ng mL <sup>-1</sup> )		RR / %	Standard addition calibration curve	r	<i>t</i> -test
	Added	Found <sup>a</sup>				
Phenobarbital	0	ND	–	$y^b = 901.6x^c + 367.4$	0.9992	1.64
	10	9.9 ± 0.8	99.1	$y = 1070.2x + 10579$	0.9986	0.17
	40	40.1 ± 1.8	100.3	$y = 1074.4x + 4304.9$	0.9975	0.11
	90	90.1 ± 1.7	100.1	$y = 1098.3x + 9897.9$	0.9985	0.16
Carbamazepine	0	ND	–	$y = 901.6x + 155.2$	0.9992	3.26
	10	10.1 ± 0.6	101.5	$y = 2025.4x + 20564$	0.9981	0.46
	40	41.4 ± 1.7	103.5	$y = 1828.2x + 7563.5$	0.9955	1.37
	90	89.7 ± 2.6	99.7	$y = 1928.9x + 17309.2$	0.9964	0.17

<sup>a</sup>Average of three measurements ± standard deviation; <sup>b</sup>area (mAU min); <sup>c</sup>concentration of analytes (ng mL<sup>-1</sup>). RR: relative recovery; r: correlation coefficient; ND: not detected.

**Table 4.** Comparison of the proposed method with previously published methods

Extraction method	Sorbent	Analyte	Real sample	Linear range / (ng mL <sup>-1</sup> )	LOD / (ng mL <sup>-1</sup> )	Sorbent / mg	Reference
MEPS	C18	CBZ PB	plasma	100-15000 200-40000	15	NR	23
SPME	SG/MWCNT	PB	wastewater	0.50-5000	0.32	50	31
DLLME-SPE	C18	CBZ	urine, plasma	2500-500000	400	NR	32
SPE	CM-SNPs	CBZ	tablet, breast milk, plasma	0.5-200	0.16	200	25
MISPE	CBZ-IP	CBZ	urine, wastewater	50-24000	25	200	20
DSPME	RGO-CdS QDs	CBZ PB	plasma, urine, breast milk	0.5-180 0.5-140	0.19 0.24	7.5	this work

LOD: limit of detection; MEPS: microextraction by packed sorbent; CBZ: carbamazepine; PB: phenobarbital; NR: not reported; SPME: solid phase microextraction; SG/MWCNT: sol-gel/multi-walled carbon nanotubes; DLLME-SPE: dispersive liquid-liquid microextraction-solid phase extraction; SPE: solid phase extraction; CM-SNPs: cation modified sulfur nanoparticles; MISPE: molecularly imprinted solid-phase extraction; CBZ-IP: carbamazepine-imprinted polymer.

advantages such as rapidity, simplicity, environmentally friendly and high sensitivity, and selectivity. It also exhibits excellent applicability in the preconcentration of analytes

in different real samples such as urine, human plasma, and breast milk with high recovery percentages.



## Supplementary Information

Supplementary data are available free of charge at <http://jbcbs.s bq.org.br> as PDF file.

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