Controlled adsorption and release of amoxicillin in GO/HA composite materials

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Abstract

Present work is aimed at preparing hybrid composites based on graphene oxide and hydroxyapatite as a matrix (GO/HA), which can be used as a template for drug delivery after surgical operation. Composite materials were synthesized through an in situ precipitation as simple and save-time method. The obtained materials were characterized using infra-red spectroscopy (FT-IR), scanning electron microscopy (SEM), thermogravimetric analysis (TGA), and X-ray diffraction (XRD). Hybrid materials were loaded with amoxicillin and the drug delivery during various periods of time was assessed with UV spectroscopy. During loading periods, it was noted that GO has a much larger cumulative drug concentration compared to hydroxyapatite and other composites. Similarly, and during the release process, amoxicillin is released much faster during the earlier stage of immersion before reaching a stationary state towards long periods of time. This variation was found to be dependent on the amount of GO in the HA matrix, as well as on the number of oxygen available on the GO sheets and the HA surfaces.

Keywords Hydroxyapatite · Graphene oxide · Drug delivery · Drug loading · Composites

1 Introduction

The bone infection caused by microorganism on orthopaedic surgeries is the costliest complications, and is one of the major increased morbidity in the world [1]. This infection is associated with patient discomfort, longer hospitalization and additional surgeries for the treatment of infected patients [2–5]. Treatment of bone infection at surgical site is actually more demanded to prevent bacterial infections. Traditionally treatment of bone infection is ensured by administration of antibiotic systematically, but unfortunately, the drawbacks of this delivery route is the poor drug diffusion in the blood [6]. However, local delivery of antimicrobial drug on material as carrier, which is able to release the drug needed in the specific organ with a very defined concentration. Therefore, this is a viable solution to ensure a treatment of infection after orthopaedic surgeries [7].

Due to its fascinating properties, graphene has been accompanied by increasing research attention to explore this new material for energy conversion and storage systems [8, 9] to photocatalysis [10–13], through drug delivery application. Graphene (Gr), is a single layer at atomic thick, and a two-dimensional (2D) sp^2 hybridized carbon material arranged in a honeycomb crystal lattice. Graphene had a wide variety of promising biomedical applications such as anti-bacterial paper, cancer targeting, photo-thermal therapy, drug delivery, biological imaging, and tissue engineering [14, 15]. Graphene has been widely used in his oxidized structure called graphene oxide (GO) due to the abundant hydroxyl, epoxide, and carboxylic groups on the basal plane and edges of graphene oxide surface respectively, which lead to the high dispersion of GO in water [16]. Consequently, makes their biomedical application in drug loading and delivery possible [17–19]. In addition the large specific surface area ($2630 \text{ m}^2 \text{ g}^{-1}$),

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the good biocompatibility, and high capability of loading drugs through physical adsorption, make GO an excellent carrier for loading and delivery for a variety of therapeutic drugs, such as doxorubicin, docetaxel, and camptothecin [20–22]. However, controlled drug release rate of GO is essentially very demanded for the performance of drug materials. In fact, if the drug release rate is faster, it can consequently kill some normal cells. In the other hand the drug release rate may not be slow, which can reduce their effective to treat bone infection [23].

Hydroxyapatite (HA) is actually an attractive biomaterial used in hard tissue engineering for restoring damaged bone, owing to its biological properties, and chemical composition similar to natural bone [24]. Also, HA have a great potential as drug delivery system. First and foremost, the geometrical structure of hydroxyapatite was found to have a vital role in adsorption of biomolecules through electrostatic attractions. The crystallographic structure of HA is formed by two horizontal layer of PO_4^{3-} tetrahedron, located at $z = \frac{1}{4}$ and $z = \frac{3}{4}$ planes, and positively charged Ca^{2+} atoms located at $z = 0, \frac{1}{2}, \frac{1}{4}$ and $\frac{3}{4}$ planes and negatively charged OH^- located at $\frac{1}{4}, \frac{3}{4}$ [25]. These two different charges of HA may be responsible for the adsorption of drugs and biomolecules on its surface. In addition, HA is very demanded for its potential application on drug delivery, thanks to the capacity to deliver anti-cancer drug [26], anti-inflammatory drug [27], anti-osteoporotic drug [28]. Therefore, the development of HA-drug systems, e.g. antibiotic, anti-microbial agent, etc., with controlled drug release seems to be an important problem, in view of the infection in bone surgery [29]. It is important to develop more advanced system able to deliver drug when required with optimal dosage within time.

Recently, few research works have introduced hydroxyapatite into graphene oxide in order to combine the advantages properties of GO and hydroxyapatite for drug delivery; and to develop composites with controlled drug release kinetics. For example, pH-sensitive drug carriers have been investigated using GO/HA composites and the release rate from these composites was found to be pH-dependent. The drug release in PBS medium (phosphate buffer solution) at a pH = 4.4 was found to be higher compared to pH = 7.4 and 9 respectively [30]. Furthermore, the porous flower like structure of the GO/HA composites revealed a reduction in drug release rate of ibuprofen (IBU), which make this nanocomposite, a promising material for drug delivery application [31]. Also, drug release of a cationic Anticancer doxorubicin was controlled on carbon dots conjugated carboxymethyl cellulose-hydroxyapatite nanocomposite which explained by a diffusion-controlled mechanism [32]. Even more, there is a serious discrepancy about the future application of hydroxyapatite particles, when they are combined with

GO, if the combination will be able to control drug release or not. The big challenge must be achieved is the new composites development functionalized by therapeutic drug to significantly improve local surgical outcomes and reduced bone infection. Despite the progress in graphene based nano-composites, to our knowledge, there are few reports on GO/HA composites as a template for drug delivery to control release drug rate. In this paper, the effect of GO loading and drugs release will be studied. The drug loading and controlled release will be investigated using Amoxicillin (Fig. 1) as the model drug.

2 Experimental details

2.1 Materials

Synthetic Graphite (powder, < 20 mm), amoxicillin, sulfuric acid (H_2SO_4), sodium nitrate (NaNO_3), potassium permanganate (KMnO_4), hydrogen peroxide (H_2O_2), chloridric acid (HCl), calcium chloride (CaCl_2), ammonia water (NH_4OH), phosphoric acid (H_3PO_4) were purchased from Sigma Aldrich Chemicals. All the chemicals were employed without further purification.

2.2 Apparatus and characterization

Fourier transform infrared (FTIR) analyses were carried out on a Thermoscientific, IS-50 FT-IR in the frequency range of $4000\text{--}400\text{ cm}^{-1}$ to identify the functional groups of the composites, with a resolution of 4 cm^{-1} . The X-ray diffraction (XRD) patterns of the powders and composites were assayed using an automated X-ray powder diffractometer (XRD, PANalytical) at a scanning rate of 0.033° per second in a 2θ range from 20° to 80° with Cu-K α radiation ($\lambda = 1.54,060\text{ \AA}$), operated at 45 kV and 40 mA. Thermogravimetric analysis (TGA) was conducted with TGA Q500. All the samples were carefully grounded to fine powder. The samples were analyzed within the temperature range $25\text{--}1000^\circ\text{C}$ at a heating rate of $10^\circ\text{C}/\text{min}$ under atmospheric environment. The morphology of the hybrids was

Fig. 1 Chemical structure of Amoxicillin drug molecule

carried out by using scanning electron microscopy (SEM) on FEI Quanta 200 EDAXR.

2.3 Synthesis of graphene oxide composites (GO)

Graphene oxide (GO) was synthesized using Hummer's method from natural graphite. Briefly, 2 g of graphite powder and 1 g of NaNO₃ were added to 50 mL of concentrated H₂SO₄ and were stirred in an ice bath, then 6 g of KMnO₄ was slowly added with vigorous stirring. The suspension was kept at 35 °C for 30 min with stirring. After that, the reaction was heated at 95 °C and stirred for 30 min after addition of 180 ml of water. Then, 450 ml of water was slowly added following by the addition of 15 ml H₂O₂ (30%) slowly. The mixture was filtered and washed with HCl (1 M, 37%) and then with distilled water, until the suspension was neutral and dried at 60 °C for 24 h [33].

2.4 In situ synthesis of HA in graphene oxide suspension

Predetermined amount of graphene oxide was dispersed by ultra-sonication in 50 ml of water for 2H. Hydroxyapatite was prepared in the presence of GO solution at different weight fraction of GO by in situ precipitation (Table 1).

CaCl₂ precursors (10.25 mM) were dissolved in 50 ml of water and the solution was added drop wise to GO solution under stirring. After that, 50 ml of diluted H₃PO₄ (6.14 mM) was added to the previous solution under agitation during 30 min. The pH of the solution was then adjusted to 10 by adding ammonia solution (NH₄OH). The solution was then stirred for 24 h at 37 °C. the resulting precipitate was centrifuged and washed four times with distilled water and ethanol. The obtained powder was dried at 80 °C for overnight.

2.5 In vitro tests: drug loading, release and biomimetic mineralization

Loading of amoxicillin (AMX) was carried out by dispersing GO/HA (a, b, c, d, e) in aqueous solution at pH = 7.42 at 37 °C for 24H in dark environment. The liquid supernatant

was separated from solid samples using centrifugation operating at 10000 rpm for 5 min. The drug loading was calculated according to the Eq. (3). The release of AMX from various composite samples (GO/HA) was carried out by dispersing pre-weighed samples in PBS medium at pH = 7.42, T = 37 °C (in an incubator) at various period of times (1, 2, 4.5, 5.5, 24, 48, 72 and 96H). Periodically, 4 ml of dissolution medium were collected and replaced by the same volume. The PBS solutions were analysed at 272 nm by UV/VIS Spectrometer Lambda-850. The experiments (loading and release) were repeated in triplicate to get mean and the standard deviation. The amount of the AMX released was calculated using a linear regression. The cumulative release of AMX was calculated from Eq. (2):

$$\text{Drug loading (\%)} = \frac{m_0 - m_t}{m_0} \times 100 \quad (1)$$

Cumulative drug release (%)_t

$$= \left(\frac{m_t}{m_0} \times 100 \right)_t + (\% \text{ drug release})_{t-1} \quad (2)$$

m₀ is the initial mass of drug used in loading (1 mg/ml), m_t is the mass of AMX collected in aqueous or PBS solution, t is the time of collection.

To investigate the apatite forming ability, cylindrical shaped samples weighing 0.1 g prepared by uniaxial press were soaked in 30 mL of SBF [34] solution in a polyethylene plastic container and were placed in an incubator at 37 °C (± 0.5 °C). After 21 days of immersion, the samples were collected from the SBF solution, rinsed with water and dried at room temperature. SEM observation was used to check the apatite formation on the surfaces of cylindrical shaped samples.

3 Results and discussion

3.1 Preparation and characterization of GO/HA composites

Scheme 1 illustrates the synthesis process of the composites GO/HA (a, b, c, d, and e). The hybrid materials were prepared using in situ precipitation method. First, GO was ultrasonically well dispersed in water. Then, in situ precipitation of HA was achieved. During the reaction process, polar functional groups of GO surface act as nucleation and growth sites of HA. First, calcium ions (Ca²⁺) with positive charge was strongly attracted to GO surfaces via electrostatic interaction. Then, phosphate ions (PO₄³⁻) will counterbalance the electronic charge of the surface. The increase of the pH of the solution to

Table 1 Samples acronyms of the prepared nanocomposites

Samples	wt% of GO
HA	0
GO/HA (a)	0.5
GO/HA (b)	1
GO/HA (c)	1.5
GO/HA (d)	2
GO/HA (e)	3
GO	100

Scheme 1 The experimental process of synthesized composites

10–10.5, will spontaneously induced nucleation and growth of HA on GO surfaces within time.

3.2 FTIR and UV analysis of GO and GO/HA composites

FTIR spectra of GO and GO/HA (a, b, c, d, e) composites are displayed in Fig. 2. According to Fig. 2a, the absorbance bands at 3203 cm^{-1} and 1246 cm^{-1} were assigned to the hydroxyl groups (O–H), 1620 cm^{-1} to the sp^2 -hybridized C=C vibration stretching. Besides, the adsorption peaks produced by other functional groups such as carboxyl C=O (1730 cm^{-1}), epoxy C–O (1051 cm^{-1}) groups on the surface of GO [35]. After nucleation and growth of HA particles, the future composites were examined with FTIR. Infrared spectra of GO/HA composites showed all characteristic peaks of hydroxyapatite at 560 , 600 , 967 and 3345 cm^{-1} respectively (Fig. 2b) [36]. Moreover, according to Fig. 2b, it was observed a net reduction of pic intensity of the absorption band of C=O (1730 cm^{-1}) indicating a strong interaction between HA and GO nanosheets. It was also observed from UV–vis spectra of GO and GO/HA composite (Fig. 2d) that GO nanosheets consist of the band at 232 nm , which is characteristic of π – π^* transitions of carboxyl group (C=O) [37]. After addition of HA, the peak located at 232 nm was shifted to 310 nm , which confirmed the strong interaction between HA and GO [31, 38].

3.3 X-ray diffraction studies and thermogravimetric analysis

Phase and crystallinity of prepared composites were identified using XRD analysis. Figure 3a shows the XRD pattern of graphite and graphene oxide where graphite shows an intense peak at $2\theta = 26.5^\circ$ and GO at $2\theta = 10.1^\circ$ (due to (002) plane) which clearly indicates the oxidation of graphite affecting its crystal structure and the inter-layer spacing which has increased from 3.4 to $8.7\text{ \AA}$ [35]. Figure 3b displayed the XRD patterns of the GO/HA composites and HA powders. All samples presented diffraction peaks of HA crystal at 2θ of 25.79 , 31.9 , 39.3 , 46.36 , 49.5 , 53.26 corresponding to (002), (211), (310), (222), (313), (004) inter-reticular plans [39]. These characteristic peaks are consistent with the standard diffraction pattern of HA (JCPDF 00-001-1008). However, after formation of HA on GO sheets, the (002) plan of GO was not observed in the composites, due the lower amount of GO, which was consistent with previous studies [40].

TGA analysis were further performed to confirm the weight change of GO, HA, and GO/HA (a, b, c, d, e) (Fig. 4). The samples were heated from room temperature to $1000\text{ }^\circ\text{C}$ at a rate of $10\text{ }^\circ\text{C}/\text{min}$. The initial weight loss at around $100\text{ }^\circ\text{C}$ was attributed to the evaporation of water for all samples. According to this figure, the GO is thermally unstable and recorded two significant loss weight for at around $200\text{ }^\circ\text{C}$ and $510\text{ }^\circ\text{C}$ due to decomposition of oxygenated functional groups. The former are attributed

Fig. 2 FTIR spectra of **a** graphite and graphene oxide, **b** GO/HA corresponding composites and **c** UV-vis spectra of graphene oxide (GO) and selected GO/HA composite

Fig. 3 **a** XRD patterns of Graphite and GO, **b** XRD patterns of GO/HA composite materials

Fig. 4 TGA profiles of HA, GO and GO composite

to formation of CO, and CO₂, and the latter are attributed to the combustion of the carbon of GO [41]. The weight loss of GO at 200 °C was 20 wt%, compared to GO/HA (a) to GO/HA (e) that show a variation of weight loss from 2.5 to 4.5 wt% respectively. In addition, when the temperature reached 900 °C, no significant weight loss was recorded for HA, and GO/HA composites (a, b, c, d, and e), which due to their high thermal stability of HA. The final weight loss was also found to be GO dependent. the higher the graphene loading rate in composite materials, the more a net decrease in mass loss is recorded at 900 °C.

3.4 AMX loading and in vitro drug release

According to FTIR spectra of AMX (Fig. 5), the absorbance bands at 1770 cm⁻¹, 1640–1560 cm⁻¹, 1350–1280 cm⁻¹, 3650–3590 cm⁻¹ were corresponded to ketone, primary amine, secondary amine, and hydroxyl (broad) groups of amoxicillin respectively [42]. The characteristic bands of AMX were also observed for all composite materials (GO/HA) which confirmed the successful loading of AMX drug molecule in all samples. As mentioned above, the UV–Vis spectroscopy displayed a characteristic absorbance band at 272 nm for amoxicillin (Fig. 2c). The loading content in composite materials were assessed by UV spectroscopy and for that the calibration curve for various concentration of AMX was released.

Figure 6a shows the evolution of the concentration of AMX in GO, HA and their corresponding composites. With respect to this figure, it can be observed that virgin graphene exhibited the highest absorbance of the AMX drug molecules (82.19%) compared to HA (51.99%) and the other composites. This can be explained by the abundance

Fig. 5 FTIR spectra of GO/HA composite samples loaded with AMX drug molecule

Fig. 6 AMX loading (a) and release (b) profiles from HA, GO and GO/HA carriers

of the graphene-sheets by functional groups capable of developing favourable interactions (π – π interactions) with AMX such as carboxyl, epoxide and carboxylic acid [43]. The second parameter impacting this extraordinary absorbance of the AMX drug molecules is the higher specific surface area of graphene sheets.

In the case of HA, the average cumulative concentration of the AMX absorbed on the surface of the granules reached 51.99%. This modest value compared to that obtained in the case of the GO, is explained by the methodology adopted during synthesizing the HA. It should be noted that the preparation of the HA by the precipitation method with vigorous stirring does not adequately allowed a good control of the geometry of the grains, neither it can increase the specific surface area of the apatite particles such as the case observed with surfactant assisted particle synthesis of hydroxyapatite [29]. This will effectively reduce the exposure of functional groups able of developing favourable interactions with AMX.

For the intermediate phases, it has been observed that the cumulative concentration of AMX increased with GO addition in the prepared composites. In the case of GO/HA composites (a, 0.5 wt% of graphene), a value of 45.42% was recorded. This value is slightly lower to that observed in the case of the virgin HA and can be explained by a net reduction of the oxygenated sites necessary to interact with the AMX drug molecules. Generally, during the formation of hydroxyapatite in the presence of suspended graphene sheets, the formation of covalent bonds between HA and GO has been observed and may be responsible for this decrease. This has been also confirmed by the FTIR spectra (Fig. 2b). As the amount of GO increased in the prepared composites, the cumulative concentration of AMX increase to reach its higher value in the case of 3 wt% of GO addition. The available of both functional groups on nanosheets of GO and HA are responsible of this enhancement. In addition, it was reported earlier that the percentage of graphene filled HA should not exceed 5 wt% to not deteriorate biological cells [44–46].

Figure 6b shows the kinetics of release of AMX drug molecules in PBS medium for virgin materials (HA and GO) and their corresponding composites. According to this figure, it can be seen that the drug release is graphene oxide dependent. As a function of time, all composites presented a net increase in the AMX concentration in the PBS solution during the earlier stage of immersion (for periods of time less than 5 h) before reaching a quasi-stationary states after 5 h of immersion. The steady states are therefore dependent on the amount of graphene in composite materials. As the rate of graphene oxide was increased in the composite materials, as the concentration limit of AMX in the PBS solution become more important. Graphene oxide therefore has the highest concentration while HA has the lowest one. This phenomenon is explained by the amount of AMX absorbed on the surface of composites during drug loading as well as by the strength of the concentration gradient during the release in the PBS solution. The rapid delivery of the drug during the earlier stage is required immediately after surgery for the effective inhibition of microorganisms and then a controlled release is needed to aid long-time healing and to avoid the toxic and adverse systemic effect caused by high concentration of antibiotics administrated orally.

In order to qualify the mechanism of AMX release, several researchers suppose that drug release mechanism occurred by either drug diffusion or eroding of the matrix, which influenced the rate of release. Several models have been used to explain the mechanisms governing the drug release. Semi-empirical models were developed by Ritger & Peppas (Eq. 3) [47] and kopcha (Eq. 4) [48–50].

$$\frac{M_t}{M_\infty} = kt^n \quad (3)$$

$$M_t = A\sqrt{t} + Bt \quad (4)$$

$\frac{M_t}{M_\infty}$ is the fractional solute release, M_t is the amount of released drug at time t , M_∞ is total amount of released drug, k is a constant, and n is the diffusional exponent characteristic of the release mechanism. n gives an information of the drug-release mechanism: if $n < 0.5$, the mechanism corresponded to Fickian diffusion and if n is situated between 0.5 and 1.0, the mechanism will correspond to non-Fickian transport. For Eq. 4, A is a diffusional term and B is the erosion term. When $A/B > 1$, the diffusion phenomenon is predominant, and when $A/B < 1$ the erosion phenomenon is predominant. Table 2 represents the results of calculations of parameters n , A and B respectively. According to the calculated parameters, it can be concluded from both models, that the drug release followed the Fickian diffusion law for all samples.

3.5 Biomimetic mineralization

Two factors are consistent from the point of view of good implants grafting: good osseointegration of the biomaterials with surrounding tissues and excellent biocompatibility of the biomaterials for the growth promotion of osteoblast cells [51]. A strong bone-bonding ability between the implant and the surrounding tissues is generally due to a regular distribution and fast apatite formation. In this study, we used in vitro acellular bioactivity tests using simulated body fluid (SBF) as a standard precursor medium mimicking the biomineralization process of the new bone. In order to evaluate this capacity, GO/HA composite materials were immersed in SBF for different periods of time (1–21 days). All GO/HA composite samples showed the formation of a new apatite layers as consistently confirmed by SEM and FTIR results. Figure 7 displayed the representative surface morphologies obtained from SEM of pure HA and the GO/HA composites after being soaked in SBF for 21 days.

Table 2 Parameters of the AMX release profile fitting according to Ritger and Peppas and Korsmeyer models

Samples	Korsmeyer–Peppas n	Kopcha A/B
HA	0.39	$\gg 1$
GO	0.38	$\gg 1$
GO/HA (a)	0.45	$\gg 1$
GO/HA (b)	0.46	$\gg 1$
GO/HA (c)	0.49	$\gg 1$
GO/HA (d)	0.46	$\gg 1$
GO/HA (e)	0.34	$\gg 1$

Fig. 7 SEM micrographs of **a** GO sheets, **b** HA before soaking, **c** HA, **d** GO/HA (0.5%), **e** GO/HA (2%), **f** GO/HA (3%) after 21 days of soaking in SBF medium

Figure 7a revealed that GO appears like folded flakes, multi layered and wavy, these observations are consistent with those from Mohandes et al. [52] study. However, HA pellet displayed a smooth surface before soaking in SBF medium (Fig. 7b). In contrary, it's clearly observed that the morphology of mineralization product varies with the addition of GO into the HA matrix and varied from spherical to sheet-like apatite forms. Increasing the amount of GO in the composites promote the deposition of a dense layer of spherical-like apatite, although pure HA ceramics showed a platelet-like crystals with the typical HA morphology. In addition, it is observed some microcracks on the GO containing samples, indicating the formation of a thick deposit [53]. The apatite layer deposition is controlled by the presence of available oxygen functionalities on GO and HA surfaces. Thereafter, both of them proceed as competing nucleation points for ionic cluster precursors. For pure HA, the same situation is observed in the case of the formation of apatite on the surface of sintered ceramics after soaking in SBF, which allows for selective ion diffusion [54–56].

In the case of GO/HA composite materials, the formation of apatite layer after 21 days of immersion (Fig. 7d–f). They specifically grow on the HA particles, and interestingly, few of them appear on divers functional groups present on GO nanosheets, which remain

accessible after the in situ synthesis of hydroxyapatite. These observations suggested that HA nuclei forming on the GO/HA surface retain a high reactivity which allows them to migrate towards the GO/HA surface to initiate HA growth and resulted on a dense apatite layer. All the observations showed clearly that the presence of hydroxyapatite and GO nanosheets are recommended to obtain a rough surface after the deposition of the new apatite layers.

As known, that this rough surface is ideal for the better adhesion of cells [57]. The formation of apatite particles on the various hybrid composites surfaces is followed more closely by FTIR spectroscopy (Fig. 8). After incubation for 21 days, the FTIR pattern revealed the presence of PO_4^{3-} and OH groups. Weak PO_4^{3-} bands appeared for HA samples. After further addition of GO, strong PO_4^{3-} bands were observed. Among these bands, the band at 964 cm^{-1} is due to the symmetric stretching mode ν_1 (PO_4^{3-}), 1015 and 1089 cm^{-1} to the vibration mode ν_3 (PO_4^{3-}), 597 , 560 cm^{-1} and 472 cm^{-1} to bending modes ν_4 (PO_4^{3-}). Then, the FTIR results also confirmed the formation of apatite phase on the GO/HA composites surfaces after soaking in SBF medium [36, 58]. The most significant and interesting finding is that hydroxyapatite and graphene oxide are a promising combination for bone implant applications [57].

Fig. 8 FTIR spectra of the pure HA as reference, and pure HA, GO/HA (a, d, e) after 21 days of soaking in SBF medium

4 Conclusions

To summarize, composite materials based on hydroxyapatite and graphene oxide were synthesized through in situ precipitation as simple and save-time method. Hybrid materials were loaded with amoxicillin as drug molecule and drug release was assessed by UV–Vis spectroscopy. In addition, the drug release was found to be GO dependent and the kinetics of drug release had a tendency to fit well with Fickian diffusion model. Moreover, the composite materials sustained the release of phosphate and calcium ions in SBF, which synergistically assists the ECM (Extracellular matrix) ossification and promotes new bone regeneration. Furthermore, the GO contentment was found to accelerate the process of biomineralization and promoted new bone formation.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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