One-pot synthesis of spirooxindole-pyrrolizidine compounds using magnetically separable Fe₃O₄-graphene oxide (Fe₃O₄-GO) catalyst

M Hapsari, A H Cahyana and R T Yunarti

Department of Chemistry, Faculty of Mathematics and Natural Sciences (FMIPA), Universitas Indonesia, Depok 16424, Indonesia

Corresponding author’s email: herrykim@ui.ac.id

Abstract. A couple of novel Spirooxindole-pyrrolizidine compounds have been synthesized from chalcone derivatives as α,β-unsaturated carbonyl compounds by the help of heterogeneous magnetically separable Fe₃O₄-GO catalyst. Four steps involved in this research are synthesis of Fe₃O₄-GO catalyst, synthesis of chalcone derivatives from 4-hydroxybenzaldehyde and cinnamaldehyde, synthesis of spirooxindole-pyrrolizidine derivatives by 1,3-dipolar-cycloaddition reaction through the formation of azomethine ylide, and the last is antioxidant activity assay using DPPH method. The result showed that the use of Fe₃O₄-GO catalyst as much as 5 wt% can increase the reaction yield to 86.27 %. From the DPPH assay, it is known that spirooxindole-pyrrolizidine derivatives have an antioxidant activity but spirooxindole-pyrrolizidine derived from 4-hydroxybenzaldehyde is the better one.

Keywords: Spirooxindole-pyrrolizidine, chalcone, Fe₃O₄-GO, 1,3-dipolar-cycloaddition, DPPH

1. Introduction
Spiro is a polycyclic compound which has two or more rings connected by one chiral carbon atom [1]. There are several types of spiro compounds depend on its structure, including spiro-oxindole, spiro-pyrrolidine and spiro-pyrrolizidine, spiro-chromene, spiro-pyrimidine, spiro-xanthene, and spiro-lactone. Among the various types of spiro, spirooxindole attracts the most attention because it promises a good activity [2]. Spirooxindole is the skeleton of many natural alkaloids which possesses biological and pharmacological activities. Besides that, spiropyrrolidine and spiropyrrolizidine also gained much attention because its potential in biological activities such as antimicrobial, antitumor, antiviral, antileukemic, anticonvulsant, and local anesthetic activities [3]. Some further researches also show that the combination of the spirooxindole with pyrrolidine or pyrrolizidine ring has a good activity. Two types of diketopiperazine alkaloids (spirotryprostatin A and B) which were first isolated from Aspergillus fumigatus are examples of spirooxindole-pyrrolidine compounds that have an excellent anticancer activity [4]. They gained much attention for years and finally have successfully synthesized in 1999 [5].

1,3-dipolar cycloaddition reaction is the most widely performed reaction to synthesize spirooxindole-pyrrolidine and pyrrolizidine derivatives. This reaction has been described as the most important method for the formation of five-membered heterocyclic rings in organic chemistry because it has a high level of diastereo-, enantio- and regioselectivity [6].
In recent years, the development of carbon-based heterogeneous catalysts has been carried out because of its environmentally friendly property. Carbon-based catalyst has been known to be used for many organic transformations such as formation of nitriles from primary amines through oxidation reaction, direct C-C bond formation, and synthesis of heterocycles like triazoles and benzimidazoles [7]. The transition metals are slowly replaced by graphene and its functionalized form like graphene oxide (GO) in organic synthesis [8, 9].

From previous research, graphene oxide (GO) played a role in several reactions such as oxidative coupling reaction, Friedel craft alkylation, Diels-Alder reaction, hydration oxidation, and many others [10]. Recently, scientists developed the synthesis of spirooxindole-pyrrolizidine compound using a catalyst GO and aqueous solution medium [10]. This research showed that GO facilitated the 1,3-dipolar cycloaddition reaction through the formation of imine and decarboxylation to become the reactive azomethine ylide which then reacted with dipolarophile compound [10].

The synthesis of spirooxindole-pyrrolizidine using GO catalyst was carried out by reacting isatin, α-amino acids, and alkenes as dipolarophile. By using GO, synthesis of spirooxindole-pyrrolizidine could be done in aqueous medium which was more environmentally friendly. GO was also easy to separate by centrifugation, but it still took a lot of time [10]. Therefore, this study will conduct the synthesis of spirooxindole-pyrrolizidine compounds through a multicomponent reaction between isatin, L-proline, and chalcone derivatives by the help of the Fe₃O₄-GO catalyst. Fe₃O₄ is added just to simplify the separating process of GO from the reaction mixture.

Chalcone derivatives were synthesized first by reacting acetophenone and aromatic aldehyde like 4-hydroxy-benzaldehyde and cinnamaldehyde in alkali medium by Claisen-schmidt condensation reaction. Chalcone is a secondary metabolite compound found in plants. It has three carbons of unsaturated α,β-carbonyl system which connected its two aromatic rings [11, 12]. Chalcone is known to have several bioactivities such as antibacterial, antioxidant, antifungal, anti-inflammatory, and anticancer [13, 14]. Spirooxindole-pyrrolizidine derivatives synthesized by chalcone derivative was characterized and tested for their antioxidant activity analysis.

2. Materials and method

2.1. Materials

The instruments used were Fourier Transform Infrared (FT-IR) IRRestige-21 Shimadzu, UV-Vis Spectrophotometers UV-2450 Shimadzu, X-Ray Diffractometer (XRD), Energy Dispersive Spectroscopy (EDS), Transmission Electron Microscope (TEM), and LC-MS/MS. Materials used to make Fe₃O₄-Graphene Oxide (GO) were graphite powder, NaNO₃, H₂O₂ 30 %, KMnO₄ powder, concentrated H₃SO₄, FeCl₃.6H₂O and FeCl₂.4H₂O. The compound used for the synthesis of chalcone and spiro-oxindole-pyrrolizidine derivatives were acetophenone, 4-hydroxy-benzaldehyde, cinnamaldehyde, NaOH, isatin, L-proline, ethanol, methanol, ethyl acetate, N-hexane, HCl, and distilled water. The material used for the antioxidant test were 1,1-diphenyl-2- picrylhydrazyl (DPPH•).

2.2. Synthesis of graphene oxide (GO)

Graphene Oxide (GO) synthesis was carried out by referring to the Hummers’ method that had been carried out before [15]. In the concentrated H₂SO₄ (34.5 mL), an amount of 1.5 g of graphite and 0.75 g of NaNO₃ were dissolved with continuous stirring for approximately 20 min, followed by cooling the mixture in an ice bath for 2 h. While maintaining the mixture temperature at below 20 °C, KMnO₄ (4.5 g) was added slowly. After that, the reaction mixture was heated and stirred at 35 °C before the distilled water (69 mL) was added to the mixture and stirred for 20 min. Then, the addition of H₂O₂ 30 % droplets (1.5 mL) was carried out until the color of the mixture changed from dark brown to yellow. A total 50 mL of distilled water was added again and the mixture was sonicated for 2 h. The solution was centrifuged, separated, and washed several times with distilled water. The product was dried in an oven at 100 °C for one day to get the graphene oxide (GO).
2.3. Synthesis of Fe₃O₄-GO
Synthesis of Fe₃O₄-GO was carried out by referring to co-precipitation method [16]. An amount of 100 mL of distilled water was poured to 0.1 g of GO followed by sonicating for 1 h. Besides that, an amount of 125 mL of distilled water was poured to 1.1 g FeCl₃·6H₂O and 0.36 g FeCl₂·4H₂O mixture then stirred for 30 min at room temperature. After that, the two mixtures were mixed and stirred at 85 °C for 30 min. This mixture was added by 25 % ammonia solution until pH 10 and the color turned to black as it changed to suspension. After that, the suspension was stirred again for 45 min before it was cooled to room temperature. The solids and liquids were separated by the help of external magnets. Then, this mixture was neutralized using distilled water and it was dried in an oven at 60 °C overnight.

2.4. Synthesis of 4-hydroxychalcone
Synthesis of 4-hydroxychalcone was done by referring to Claisen-Schmidt condensation reaction that previously reported [12, 17]. Into 2.5 g of 4-hydroxy-benzaldehyde (16.43 mmol) in 15 mL ethanol, an amount of 1.98 mL of acetophenone (16.43 mmol) and NaOH 60 % as much as 10 mL were added. The mixture was stirred at 70 °C for 3 hours until the reaction is completed (checked by TLC). After that, ethanol was removed before acidification of the mixture to pH 1 by using 1 M HCl. Anhydrous Na₂SO₄ was added to the organic phase obtained from liquid-liquid extraction (ethyl acetate) to remove the remained water. Furthermore, the solvent was removed and the purification was done by recrystallization. The solids obtained were then analyzed using spectrophotometer UV-Vis, FT-IR and LC-MS/MS.

2.5. Synthesis of 1,5-diphenylpenta-2,4-dienone
Synthesis of 1,5-diphenylpenta-2,4-dienone was done by referring to the procedure that has previously reported [17]. A total of 10 mL ethanol was added to 20 mL NaOH 12.5 % (w/v) with continuous stirring until it cooled down to room temperature. After that, an amount of 15 mmol of acetophenone and 15 mmol of cinnamaldehyde were added to NaOH-ethanol mixture dropwisely with continuous stirring for an hour. The reaction mixture was cooled in an ice bath until it formed a solid. The solution was removed and the solid was washed and recrystallized using ethanol to get the product.

2.6. Synthesis of Spirooxindole-pyrrolizidine derivatives
Synthesis of spirooxindole-pyrrolizidine derivatives was carried out based on the development of the procedure that previously reported [10]. A number of Fe₃O₄-GO catalysts in various mediums were added to a mixture of 1,5-diphenylpenta-2,4-dienone, isatin, and L-proline (1:1:1 mole ratio). The reaction was carried out at 65 °C and monitored using TLC. The catalyst was separated from the external magnetic source, and the solution was evaporated until solids remain. Recrystallization of solids was carried out by washing solids formed using warm ethanol to obtain the derivative of the Spirooxindole-pyrrolizidine-1,5-diphenylpenta-2,4-dienone compounds. Under the same procedure, isatin and L-proline were reacted with 4-hydroxychalcone in 1:1 mole ratio to obtain spirooxindole-pyrrolizidine-4-hydroxychalcone.

2.7. Antioxidant activity assay
The antioxidant activity of the organic compounds were calculated by using 1,1-diphenyl-2-picrylhydrazyl (DPPH•) method [18]. Five different concentrations of synthesized products and curcumin as its standard were made by using methanol as the solvent (200, 100, 50, 25 and 12.5 ppm). Then, 2 mL of the test sample was added to 2 mL of DPPH• 0.1 mM solution. The mixture was incubated in the dark place at room temperature for 30 min. All the mixed solution was measured at maximum wavelength 517 nm using a UV-Vis spectrophotometer. The IC50 of all test sample determined by plotting its concentration and % DPPH scavenged in a linear graphic. The equation used to calculate the free radical scavenging activity is:
where $A$ is the absorption of DPPH solution and $B$ is the absorption of the sample solutions.

3. Results and discussion

3.1. Characterization of graphene oxide (GO)
Preparation of Fe$_3$O$_4$-GO was done in two step processes. First of all, graphite powder was used to synthesize GO by using Hummer’s Method. The synthesized GO was characterized using FT-IR and XRD instruments. There are four peaks appeared at 3578.10 cm$^{-1}$ (O-H), 3101.66 cm$^{-1}$ (C-H sp$^2$), 1753.36 cm$^{-1}$ (C=O Carboxylate), 1258.60 cm$^{-1}$ (C-O-C Epoxide group) in the FTIR spectrum (figure 1) indicating that the graphite powder was successfully functionalized to GO. These FT-IR data are supported by the XRD spectrum (figure 2) of synthesized GO which exhibits a different sharp peak at Bragg angle $2\theta = 10.49^\circ$ compared to graphite ($2\theta = 26.56^\circ$). The different position of Bragg angle between graphite and GO are indicated the formation of GO. Additionally, the crystallite size of GO was calculated by Debye-Scherrer’s equation [19] using XRD data below:

\[
D = \frac{k \lambda}{\beta \cos \theta}
\]

where $D$ = the size of crystallite, $\lambda = 1.5406$ nm (wavelength of the X-ray sources), $k = 0.9$ is the Scherrer constant, $\beta$ = Full Width and Half Maximum/ FWHM (in radian), and $\theta$ is the Bragg diffraction angle or peak position (in radian). By this equation, the crystallite size of GO is estimated about 11.21 nm.

After GO has been successfully synthesized, the second process is reacting GO with FeCl$_3$.6H$_2$O and FeCl$_2$.4H$_2$O using co-precipitation method to get the Fe$_3$O$_4$-GO composite.

3.2. Characterization of Fe$_3$O$_4$-GO
Preparation of Fe$_3$O$_4$-GO was carried out by reacting GO with FeCl$_3$.6H$_2$O and FeCl$_2$.4H$_2$O and the addition of 25 % ammonia solution. The synthesized Fe$_3$O$_4$-GO was characterized by FT-IR, XRD, EDS and TEM instruments. In the FTIR spectrum of Fe$_3$O$_4$-GO (figure 1), although the peaks are not quite clear, it appears that there are four peaks similar to GO and a peak similar to Fe$_3$O$_4$. This spectrum is compared to Fe$_3$O$_4$ and GO and it shows there are four peaks appeared at 3610.89 cm$^{-1}$ (O-H), 3138.31 cm$^{-1}$ (C-H sp$^2$), 1731.18 cm$^{-1}$ (C=O Carboxylate), 1258.60 cm$^{-1}$ (C-O-C Epoxide group) confirming that there is GO in this composite. There is also a peak appeared at 628.82 cm$^{-1}$ (Fe-O bond) confirming that there is Fe$_3$O$_4$. The FTIR spectrum of Fe$_3$O$_4$-GO shows that there are some intensity reduction such as Fe-O vibration peak of the synthesized Fe$_3$O$_4$-GO. The Fe-O vibration peak is not clear and sharp like Fe$_3$O$_4$. The peaks indicated GO compound are also not as sharp as like GO. This intensity reduction is assumed because there is an interaction between Fe$_3$O$_4$ and GO.

XRD pattern or diffractogram of Fe$_3$O$_4$-GO composite is shown in figure 2. This diffraction peaks are shown at Bragg angle $2\theta$ = 30.54$^\circ$, 35.59$^\circ$, 43.39$^\circ$, 57.52$^\circ$ and 63.09$^\circ$ indicated the characteristics of Fe$_3$O$_4$ (JCPDS 19–0629) [19]. In this research, Fe$_3$O$_4$ was also synthesized for the analysis comparison. The XRD spectrum of the synthesized Fe$_3$O$_4$ also showed diffraction peaks at Bragg angle $2\theta$ = 30.17$^\circ$, 35.51$^\circ$, 43.09$^\circ$, 57.14$^\circ$ and 62.57$^\circ$. This diffraction peaks implied that the Fe$_3$O$_4$ core crystal structure was still well-maintained after functionalization. When it examined in more detail, each peak in the XRD spectrum of the synthesized Fe$_3$O$_4$-GO has a lower intensity than Fe$_3$O$_4$. This further supports the assumption that there is an interaction between Fe$_3$O$_4$ and GO.
Figure 1. FTIR spectra of graphite, GO, Fe₃O₄ and Fe₃O₄-GO.

Figure 2. Diffractogram of (a) Graphite, (b) GO, (c) Fe₃O₄ and (d) Fe₃O₄-GO.
Figure 3. EDS spectrum of (a) Fe$_3$O$_4$ and (b) Fe$_3$O$_4$-GO.

Figure 4. Typically TEM image of Fe$_3$O$_4$-GO.

There were no diffraction peaks indicated as GO, due to its low concentration. But the elemental composition of Fe$_3$O$_4$-GO confirmed that there is carbon atom in the synthesized Fe$_3$O$_4$-GO. The elemental composition is determined by energy dispersive spectrum (EDS) analysis. In the synthesized Fe$_3$O$_4$-GO there are 04.49 wt% of C atom, 31.76 wt% of O atom and 63.74 wt% of Fe atom, while in the Fe$_3$O$_4$ there are 00.65 wt% of C atom, 18.03 wt% of O atom and 63.74 wt% of Fe atom. This spectrum is shown in figure 3. Typically, TEM image of Fe$_3$O$_4$-GO composite in the figure 4 shows that most particles Fe$_3$O$_4$-GO have spherical shape and the particle size is 174 nm.

3.3. Application of Fe$_3$O$_4$-GO catalyst in the synthesis of spirooxindole-pyrrolizidine derivatives

In this research, synthesis of Spirooxindole-pyrrolizidine derivatives was done by reacting the compounds isatin, L-proline, and two different types of unsaturated $\alpha,\beta$-carbonyl compound such as 4-hydroxychalcone and 1,5-diphenylpenta-2,4-dienone. Both of unsaturated $\alpha,\beta$-carbonyl compounds were synthesized from the reaction of acetophenone and two different types of aromatic aldehydes like 4-hydroxybenzaldehyde and cinnamaldehyde in an alkaline medium. The reaction of unsaturated $\alpha,\beta$-carbonyl compound which represented by 1,5-diphenylpenta-2,4-dienone can be seen in figure 5.

The optimization of reaction condition was done using the model of reaction between isatin, L-proline, and 1,5-diphenylpenta-2,4-dienone. The reaction of spirooxindole-pyrrolizidine derivative
which is represented by spirooxindole-pyrrolizidine-1,5-diphenylpenta-2,4-dienone can be seen in figure 6. All reaction was done using methanol solvent at 65 °C for approximately 3 h. Quantities of the catalyst used are calculated based on the total weight of all precursors used. The yield of all the reaction are shown in table 1.

From the data in table 1, it can be seen that the use of GO in this reaction has improved the reaction yields compared to reaction with no catalyst. However, the use of Fe₃O₄-GO as much as 5 wt% give the highest result of the desired product. In 3 h of reaction, that condition gives a high yield (86.27 %). The data also showed that increasing amount of catalyst, either Fe₃O₄, GO or Fe₃O₄-GO will decrease the reaction yields. This phenomenon maybe due to the reaction equilibrium between the reactant and the catalyst. Because of this, the excess catalyst can plug the active site of other catalyst so it can be less active. The addition of Fe₃O₄ can also improve the yields, but when compared to GO, there is a confusion that still need to be further investigated. This is because when the GO yields are used as a reference, on the use of 5 wt% catalyst, the addition of Fe₃O₄ to GO can increase the reaction yields, but on the use of 10 wt% catalyst, the addition of Fe₃O₄ to GO actually reduces the reaction yields.

Table 1. Optimization of the reaction condition of synthesis spirooxindole-pyrrolizidine derivatives.

| Catalyst   | Quantity (wt%) | Yield (%)* |
|------------|----------------|------------|
| Fe₃O₄-GO  | 10             | 68.51      |
| GO         | 10             | 75.87      |
| Fe₃O₄     | 10             | 42.85      |
| Fe₃O₄-GO  | 5              | 86.27      |
| GO         | 5              | 78.00      |
| Fe₃O₄     | 5              | 72.33      |
| No Catalyst | -             | 40.36      |

*The yield was obtained from the reaction of 1,5-diphenylpenta-2,4-dienone
GO was also taking part in the optimization because according to the previous report [10], functional group of GO such as -OH and -COOH plays an important role in this reaction. GO facilitated the reaction through hydrogen bonding and π-stacking between the catalyst and the substrates [10]. Furthermore, the addition of Fe₃O₄ was aimed to facilitate the separation from the reaction mixture due to its magnetic properties. It is also known that GO is acidic in nature, so it is facilitated the formation of reactive azomethine ylide. This reactive azomethine ylide leads to the formation of 5 membered heterocyclic compound and the reaction is well known as 1,3-dipolar cycloaddition or Huisgen Cycloaddition. The proposed mechanism of Fe₃O₄-GO catalyst is shown in figure 7.

In this research, characterization of Fe₃O₄-GO and GO after application were done by FTIR. From the FTIR spectrum of Fe₃O₄-GO catalyst after application, it can be seen that the vibration peak of the Fe-O bond has increased in intensity. Meanwhile, from the GO catalyst spectrum after application, it can be seen the overall vibration peaks have decreased in intensity. These results prove that the one that plays a role in the reaction is GO. The peak of Fe-O vibrations has increased due to the decrease in GO. FTIR Spectra of GO and Fe₃O₄-GO after application can be seen in figure 8.

3.4. Antioxidant activity assay.
After all the synthesized product are well characterized (showed in table 2), their antioxidant activity was determined. The free radical scavenging activity which is represented by their IC₅₀ value are shown in figure 9. From the result, spirooxindole-pyrrolizidine-4-hydroxychalcone possess the best free radical scavenging activity (IC₅₀ 1053.65 ppm) compared to spirooxindole-pyrrolizidine-1,5-diphenylpenta-2,4-dienone and 1,5-diphenylpenta-2,4-dienone as its precursor. Meanwhile, 1,5-diphenylpenta-2,4-dienone possesses the lowest free radical scavenging activity (IC₅₀ 2738.6 ppm) compared to the others. This is because 4-hydroxychalcone has a hydroxyl group (-OH) while the others don’t have any hydroxyl group. It is known that the antioxidant activity of a compound is related to their hydroxyl groups and conjugation of its double bond or effects of resonance. In this mechanism, a hydrogen atom removed the free radical so the compound becomes a radical [19].

![Figure 7. Plausible Mechanism of Fe₃O₄-GO Catalyst.](image-url)
Figure 8. FTIR Spectra of (a) GO and (b) Fe₃O₄-GO after application.

### Table 2. Spectral data of the synthesized organic compound.

| No. | Compound                                   | Spectral data                                                  | Yield (%) |
|-----|--------------------------------------------|----------------------------------------------------------------|-----------|
| 1.  | Spirooxindole-pyrrolizidine-4-hydroxychalcone | LC-MS/MS [M+H]+ 424.49, FTIR 3517.34 cm⁻¹ (N-H amide), 3295.52 cm⁻¹ (O-H), 3107.45 cm⁻¹ (C-H sp²), 2980.14 cm⁻¹ (C-H sp³), 1732.15 cm⁻¹ (C=O), 1620.27 cm⁻¹ (C=C aromatic), 1474.64 cm⁻¹ (-CH₂ methylene), 1341.54 cm⁻¹ (-CH methine), 762.87 cm⁻¹ (ortho substituted benzene). | 79.23     |
| 2.  | Spirooxindole-pyrrolizidine-1,5-diphenylpenta-2,4-dienone | LC-MS/MS [M+H]+ 434.52, FTIR 3267.55 cm⁻¹ (N-H amide), 3089.13 cm⁻¹ (C-H sp²), 2969.53 cm⁻¹ (C-H sp³), 1727.32 cm⁻¹ (C=O), 1623.16 cm⁻¹ (C=C aromatic), 1468.85 cm⁻¹ (-CH₃ methylene), 1341.54 cm⁻¹ (-CH methine), 748.41 cm⁻¹ (monosubstituted benzene). | 86.27     |

Figure 9. Free radical scavenging activity of spirooxindole-pyrrolizidine derivatives.
4. Conclusion
Synthesis of Spirooxindole-pyrrolizidine derivatives using magnetically separable graphene oxide (Fe₃O₄-GO) catalyst have been reported. The experiment showed that the use of catalyst in 5 wt% can increase the reaction yield (86.27 %). The result of DPPH assay showed that spirooxindole-pyrrolizidine derivatives have an antioxidant activity but spirooxindole-pyrrolizidine derived from 4-hydroxybenzaldehyde is the better one.

References
[1] Borad M A, Bhoi M N, Prajapati N P and Pate H D 2013 Int. J. Rapid Comm. Synth. Org. Chem. 44 897-922
[2] Chen C et al. 2017 Molecules 22 1295
[3] Chande M S et al. 2005 Eur. J. Med. Chem. 40 1143-8
[4] Cui C-B, Kakeya H and Osada H 1996 Tetrahedron 52 12651-66
[5] Edmondson S, Danishefsky S J, Sepp-Lorenzino L and Rosen N 1999 J. Am. Chem. Soc. 121 2147-55
[6] Coldham I and Hufton R 2004 Chem. Rev. 105 2765-2809
[7] Girish Y R, Pandit S, Pandit S and De M 2017 Chem. Asian J. 12 2393-8
[8] Mohammadi O, Golestanazadeh M and Abdouss M 2017 New J. Chem. 41 11471-97
[9] Navalon S, Dhakshinamoorthy A, Alvaro M, Antonietti M and Garcia H 2017 Chem. Soc. Rev. 46 4501-29
[10] Reddy M S, Kumar N S and Chowhan L R 2018 RSC Adv. 8 35587-93
[11] Tran T-D et al. 2012 Bioorg. Med. Chem. Lett. 22 4555-60
[12] Hapsari M, Windarti T, Purbowatingrum, Ngadiwiyana and Ismiyarto 2018 IOP Conf. Ser. Mater. Sci. Eng. 349 012036
[13] Kumar S and Pandey A K 2013 Sci. World J. 2013 162750
[14] Choudhary A N and Juyal V 2011 Int. J. Pharm. Pharm. Sci. 3 125-8
[15] Marcano D C 2010 ACS Nano 4 4806-14
[16] Yang X, Zhou T, Ren B, Shi Z and Hursthouse A 2017 J. Anal. Methods Chem. 2017 3012364
[17] Anjani O O, Ituen R I and Falomo A 2011 Pak. J. Sci. Ind. Res. Ser. A. Phys. Sci. 54 59-67
[18] Borra S K et al. 2013 J. Med. Plants Res. 7 2680-90
[19] Jaleh B, Khalilipour A, Habibi S, Niyazi M and Nasrollahzadeh M 2017 J. Mater. Sci. Mater. Electron. 28 4974-83