Studying the Adsorption Process of Riboflavin on Silver-Deposited Fe$_3$O$_4$ Nanoparticles

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The adsorption characteristics of riboflavin onto silver-deposited iron oxide magnetic nanoparticles (Ag/Fe$_3$O$_4$) have been described. Characterization of the synthesized Ag/Fe$_3$O$_4$ nanoparticles was achieved by FTIR spectra, TEM image and XRD pattern. The influence of several experimental parameters such as nanoparticles dosage, pH of the sample solution, different orientations of the riboflavin molecules toward Ag/Fe$_3$O$_4$ surface, riboflavin concentration, contact time of the reagents, temperature, ionic strength and presence of halide anions were studied. Experimental data indicated that Ag/Fe$_3$O$_4$ nanoparticles adsorb more than 90% of riboflavin under the optimum experimental conditions of the absorbent dosage of 4.0 mg, a pH of 6.0, and a contact time of 2.0 min, when an initial riboflavin concentration of 0.02 mM is used. The results revealed that the presence of halide anions lower the adsorption of riboflavin on the surface of nanoparticles due to dissolution of the silver layer of the nanoparticles. It was found that the adsorption isotherm is best fitted to Dubinin-Radushkevich and Freundlich models and kinetic model followed a pseudo-second-order adsorption rate.

Keywords: Riboflavin, Silver-deposited Fe$_3$O$_4$ nanoparticles, Adsorption, UV-Vis spectroscopy, Magnetic nanoparticles

INTRODUCTION

It has been reported that riboflavin acts as a potential drug to kill tumor tissues in the photodynamic therapy [1]. Other functions, such as detumescence, and decrease of cardiovascular or cerebrovascular illness have been also attributed to riboflavin [2]. Riboflavin is used as a ligand for cancer targeting for a number of reasons. As a small molecule, riboflavin has several functional groups that are amenable for covalent modifications.

There is an over-expression of riboflavin receptors on the cell membrane of breast and prostate cancer cells [3,4]. These receptors allow riboflavin to enter the cell and assist in the formation of the redox cofactors that are necessary for basic functions of the cell. So, riboflavin receptors can be used as a biomarker to direct the drug to the cancer cells only.

Recently, there has been much interest in the fabrication of composite particles, such as core-shell structured materials [5,6]. These core-shell materials produced by controlled coating of core particles with organic or inorganic layers are exploited in the areas of catalysis and separations.

In 2012, Kamran et al. investigated the adsorption of folic acid, riboflavin and ascorbic acid by Fe$_3$O$_4$ Magnetic nanoparticles using [C$_6$MIM] ionic liquid as modifier [7]. The immobilization of zinc(II)-cyclen complexes as binding motifs for the extraction and release of riboflavin from aqueous media on carbon-coated magnetic metal nanoparticles was introduced by Kainz et al. in 2012 [8]. Spectrophotometric behavior of riboflavin adsorbed on silver nanoparticles has been evidenced in 2013 by Voicescu research group [9].

In the present study, the adsorption characteristics of riboflavin on the surfaces of silver-deposited iron oxide magnetic nanoparticles (Ag/Fe$_3$O$_4$) are investigated. Due to riboflavin tendency for interaction with Ag nanoparticles, silver deposited on Fe$_3$O$_4$ would create appropriate
condition for binding riboflavin molecules onto the surface of Fe₃O₄ nanoparticles.

EXPERIMENTAL

Instruments

A Pharmacia Ultraspec 4000 UV-Vis spectrophotometer connected to a personal computer equipped with a 1-cm quartz cell was used for recording the visible spectra and absorbance measurements.

A Zeiss transmission electron microscope (model Emloc) was taken to determine the size of the prepared Fe₃O₄ nanoparticle. The XRD measurements were performed on an XRD Bruker D8 Advance instrument. The FTIR spectra were recorded on a Spectrum RX1, Perkin Elmer spectrometer. A water ultrasonicator (Model CD-4800) was used to disperse the nanoparticles in solution and a Metrohm 780 pH meter was used for monitoring the pH of the solutions. A super magnet Nd-Fe-B (1.4 T, 10 cm × 5 cm × 2 cm) was used. All measurements were performed at ambient temperature.

Reagents

Riboflavin with certified purity of 98% was purchased from Acros. Ethyl amine (70%), sodium hydroxide, hydrochloric acid (37% w/w), FeCl₃·6H₂O (96% w/w), FeSO₄·7H₂O (99.9% w/w) and ethanol (96%) were purchased from Merck. Silver nitrate was bought from Panreac.

Riboflavin was dissolved in water to prepare stock solution (0.266 mM). For treatment experiments, the riboflavin solutions with concentrations in the range of 0.02-0.20 mM were prepared by successive dilution of the stock solution. The pH adjustments were performed with HCl and NaOH solutions (0.1 M).

Synthesis of Fe₃O₄ Nanoparticles

The Fe₃O₄ nanoparticles were prepared by co-precipitating Fe(II) and Fe(III) ions in sodium hydroxide solution (1.5 M) with a constant stirring of 1500 rpm as recommended in the literature [10]. Firstly, 0.50 M FeCl₃, 0.25 M FeSO₄ and 1.5 M NaOH aqueous solutions were prepared. The precipitation of magnetic iron oxide nanoparticles was achieved by dropwise addition of Fe(II) and Fe(III) solutions to NaOH solution under vigorous stirring. To obtain the highest yield for magnetic nanoparticles, through the co-precipitation process, an Fe(II)/Fe(III) molar ratio of 0.5 was used. The precipitate was washed three times with distilled water (50.0 ml water in each time). The nanoparticles were dispersed in distilled water by ultrasonicator for 10 min at room temperature. Then, the Fe₃O₄ nanoparticles were magnetically separated.

Synthesis of Ag-Deposited Fe₃O₄ Nanoparticles

To deposit silver on the surface of the magnetic nanoparticles, 200.0 mg of Fe₃O₄ powder was placed in a polypropylene container into which 10 ml of an ethanolic silvering solution (80.0 mM AgNO₃ in absolute ethanol and 74.5 mM ethylamine) was added followed by incubating for 40 min at 50 °C with vigorous shaking [11]. The polypropylene container was used to avoid nonspecific silvering of the reaction vessel. The Ag-deposited Fe₃O₄ (hereafter, denoted as Ag/Fe₃O₄) nanoparticles were separated magnetically from the reaction mixture and then rinsed with ethanol.

Optimization Procedure

To study the effect of the experimental parameters, the batch technique was conducted. To do so, aliquots of 2.0 ml of the riboflavin solutions in a concentration range of 0.02-0.10 mM were used and the pH effect was studied in the range of 2.0-12.0. The value of pH was adjusted by 0.10 M HCl and/or NaOH solutions. The riboflavin solutions were transferred into individual beakers containing a known dosage of Ag/Fe₃O₄, in the range of 1-10 mg. Each suspension was immediately stirred with a magnetic stirrer for a predefined period of time in the range of 2-10 min. After the mixing time elapsed, the Ag/Fe₃O₄ nanoparticles were magnetically separated and the mother solutions were spectrophotometrically analyzed at 260 nm for the residual of riboflavin. The percent adsorption of riboflavin was determined by using the following equation:

\[
\text{Riboflavin adsorption efficiency (\%)} = \left( \frac{C_0 - C_f}{C_0} \right) \times 100
\]

where \(C_0\) and \(C_f\) (in mM), respectively, represent the initial
Fig. 1. The TEM image of the synthesized Fe$_3$O$_4$ nanoparticle.

Fig. 2. The XRD spectra of the synthesized Fe$_3$O$_4$ (A) and Ag/Fe$_3$O$_4$ (B) nanoparticles.
RESULTS AND DISCUSSION

Characterization of Fe$_3$O$_4$ and Ag/Fe$_3$O$_4$ Nanoparticles

The particle size and shape of Fe$_3$O$_4$ nanoparticles were determined by transmission electron microscopy (TEM). Figure 1 shows the TEM of the synthesized nanoparticles. The nanoparticles showed a narrow size distribution with an average particle diameter of 10 nm.

The XRD pattern of Fe$_3$O$_4$ nanoparticles is shown in Fig. 2A. Six characteristic peaks [12,13] for Fe$_3$O$_4$ ($2\theta = 30.1^\circ$, $35.5^\circ$, $43.3^\circ$, $53.4^\circ$, $57.2^\circ$ and $62.5^\circ$), respectively, corresponding to indices (220), (311), (400), (422), (511) and (440) can be clearly observed in the pattern.

For Ag/Fe$_3$O$_4$ nanoparticles, the XRD pattern is provided in Fig. 2B. Four additional characteristic peaks for Ag are noticeable at 20 values of $38.1^\circ$, $44.3^\circ$, $64.4^\circ$ and $76.8^\circ$, corresponding to the reflections of (111), (200), (220) and (311) crystalline planes of the cubic Ag, respectively [11].

The size of Ag nanoparticles deposited on the Fe$_3$O$_4$ surfaces was determined to be 13.1 nm from the full width at half maximum (FWHM) of the X-ray diffraction peak of (111) shown in Fig. 2B using the Scherrer’s equation and an X-ray wavelength of 1.54 Å [14].

The FTIR spectrum of Fe$_3$O$_4$ nanoparticles are shown in Fig. 3. The broad absorption band at 3440 cm$^{-1}$ indicates the presence of surface hydroxyl groups (O-H stretching). The bands at low wave numbers ($\leq 700$ cm$^{-1}$) are related to the vibrations of the Fe-O bonds. The presence of magnetic nanoparticles in the samples was verified by the appearance of two strong absorption bands around 632 and 585 cm$^{-1}$ [15,16].

The FTIR spectrum of Ag/Fe$_3$O$_4$ nanoparticles is shown in Fig. 3. Compared to Fe$_3$O$_4$, peak shifts of Ag/Fe$_3$O$_4$ nanoparticles are indicative of formation of Ag layer onto Fe$_3$O$_4$ nanoparticles’ surface. Absence of the peak in 3440 cm$^{-1}$ represents the reduction in the number of O-H groups on Ag/Fe$_3$O$_4$ nanoparticle.

Point of Zero Charge Measurement

The pH at the potential of zero charge (pH$_{zpc}$) of Ag/Fe$_3$O$_4$ was measured using the pH drift method [17,18]. The value of pH$_{zpc}$ is necessary for discussing the effect of pH on the adsorption of riboflavin onto the nanoparticles’ surface. To maintain a constant ionic strength, the pH of 0.01 M NaNO$_3$ solution was adjusted in the range of 3.0-11.0 by adding either HCl or NaOH. Nitrogen gas was bubbled through the solution at 25 °C to remove dissolved carbon dioxide until the initial pH stabilized. The Ag/Fe$_3$O$_4$ nanoparticles (15.0 mg) were added to 15.0 ml of the solution. After pH stabilization (typically after 24 h), the final pH was recorded. The pH value at the point of zero charge (pH$_{zpc}$) was determined by plotting the difference of final and initial pHs ($\Delta$ pH) versus the initial pH.

As seen in Fig. 4, the pH$_{zpc}$ value of Ag/Fe$_3$O$_4$ nanoparticles was 9.8 while that of Fe$_3$O$_4$ nanoparticles was 6.5; the latter was already tested in a previous work [19]. Fig. 4. The difference between pH$_{zpc}$ of Ag/Fe$_3$O$_4$ and Fe$_3$O$_4$ nanoparticles could be due to less O-H groups available on the surface of Ag/Fe$_3$O$_4$ as also concluded from the FTIR spectra.

Riboflavin Adsorption Study

In the preliminary tests, Fe$_3$O$_4$ nanoparticles showed no tendency toward adsorption of riboflavin as demonstrated by measuring absorbance at 266 nm (Fig. 5). On the other hand, the adsorption of riboflavin on silver surface has been already reported [20]. So, it was decided to investigate the possible adsorption of riboflavin on Ag/Fe$_3$O$_4$ nanoparticles. In contrast to Fe$_3$O$_4$ nanoparticles, as seen in Fig. 5, Ag/Fe$_3$O$_4$ nanoparticles showed appropriate tendency for riboflavin adsorption. To study this adsorption process, various experimental parameters including nanoparticle dosage, pH of the solution, riboflavin concentration, contact time between reagents, ionic strength, temperature, and effect of foreign ions including halides were taken into consideration. Results are presented and discussed in the following sections.

Fourier Transform Infrared Spectrograms Analysis

Fourier transform infrared spectra of Fe$_3$O$_4$ and Ag/Fe$_3$O$_4$ nanoparticles along with riboflavin as well as riboflavin adsorbed on Ag/Fe$_3$O$_4$ nanoparticle are shown in
Fig. 3. The FTIR spectra of the synthesized Fe₃O₄ (A) and Ag/Fe₃O₄ (B) nanoparticles.

Fig. 4. The ΔpH vs. initial pH for determination of point of zero charge (pHₚzc) of Fe₃O₄ (A) and Ag/Fe₃O₄ (B) nanoparticles.
Fig. 5. The UV-Vis absorption spectra of riboflavin 0.02 mM of its aqueous solution (−), after adsorption process by Fe₃O₄ (−−) and Ag/Fe₃O₄ (−−−) nanoparticles. Experimental conditions for both adsorption processes by Fe₃O₄ and Ag/Fe₃O₄ nanoparticles with a nanoparticle dosage of 4.0 mg and a stirring time of 5.0 min at 1500 rpm.

Fig. 6. The FTIR spectra of riboflavin, Ag/Fe₃O₄ nanoparticle, and riboflavin adsorbed onto Ag/Fe₃O₄ nanoparticles.
Fig. 6. For riboflavin, the FTIR peak in 1650 cm\(^{-1}\) is due to O=CNH [21] while aromatic rings and ketones were observed in about 1500 cm\(^{-1}\) and 1730 cm\(^{-1}\), respectively. For riboflavin adsorbed onto Ag/Fe\(_2\)O\(_4\) nanoparticles, compared with free Ag/Fe\(_2\)O\(_4\) nanoparticle, new peaks were observed in 1542.9, 1650.9 and 1728.1 cm\(^{-1}\) showing that riboflavin has been adsorbed on the surface of nanoparticles.

**Effect of Nanoparticle Dosage**

Adsorbent dosage required for adsorbing a given initial concentration of the adsorbate is called the capacity factor. To this end, different dosages of powdered Ag/Fe\(_2\)O\(_4\) nanoparticles in the range of 2.0-10.0 mg for adsorption of a fixed amount of riboflavin were tested. The initial concentration of riboflavin solution (2.0 ml) and pH were fixed at 0.02 mM and 6.0, respectively, for all batch experiments. The resulting suspensions were immediately stirred for 2.0 min. After elapsing the mixing time, the nanoparticles were magnetically separated and the solutions were analyzed for the residual riboflavin.

Results, shown in Fig. 7, indicate that 76% of riboflavin is adsorbed from their individual aqueous solutions when an initial dosage of 1.0 mg nanoparticles is used. The percent adsorption of riboflavin is increased with increasing Ag/Fe\(_2\)O\(_4\) dosage (up to 4.0 mg) and then reaches over 90% at this dosage. This observation was almost in agreement with this fact that when a higher dosage of the adsorbent (i.e. Ag/Fe\(_2\)O\(_4\)) is used, more adsorption sites would be available for the adsorbate, i.e. riboflavin. However, adsorption capacity of Ag/Fe\(_2\)O\(_4\) leveled off at dosages higher than 4.0 mg. Hence, the optimum dosage of Ag/Fe\(_2\)O\(_4\) powder for adsorption of riboflavin from its solution was found to be 4.0 mg.

**Effect of Contact Time**

The contact time (stirring time) between adsorbate and adsorbent is the most important design parameter affecting the performance of adsorption processes. Short contact time favors the application of the adsorption process. The effect of contact time on the adsorbing ability of Ag/Fe\(_2\)O\(_4\) for riboflavin was investigated while pH and Ag/Fe\(_2\)O\(_4\) dosage were fixed at 6.0 and 4.0 mg, respectively.

The initial riboflavin concentration in test solutions was 0.02 mM and the adsorption efficiency (%adsorption) of riboflavin was monitored as a function of stirring time ranging from 1.0-10.0 min. The results (Fig. 8) indicated that adsorption started immediately upon adding Ag/Fe\(_2\)O\(_4\) nanoparticles to the riboflavin solution. The adsorption efficiency increased from 87% in the first minute of contact time to a value of 90%, when the stirring continued to 2.0 min. Therefore, the optimum contact time was considered to be 2.0 min.

**Effect of Solution pH**

The pH of the solution plays an important role in the adsorption process due to altering the surface charges of the nanoparticles as well as changing the degree of ionization of riboflavin (pK\(_a\) = 10.0). The influence of pH on adsorption of riboflavin onto Ag/Fe\(_2\)O\(_4\) surface was assessed in the pH range of 2.0-12.0 and the corresponding results are shown in Fig. 9. The initial concentration of riboflavin (2.0 ml), adsorbent dosage, and stirring time were set at 0.02 mM, 4.0 mg, and 2.0 min, respectively. Figure 9 shows that the maximum adsorption of riboflavin is achieved in the pH range of 4.0-7.0.

In 2012, Liu et al. [20] carried out the surface enhancement Raman studies on interaction of riboflavin molecules with silver surface. Their study revealed that this interaction performs through C=O and N-H groups of the uracil ring with different orientations depending on the pH of the solution. In the pH range of 3.4-4.5, the riboflavin molecules are assumed to be adsorbed in a predominant tilted orientation of the uracil ring toward the silver surface, Scheme 1A, only through the N-H group of the ring as schematically demonstrated in Scheme 1A.

In the pH range of 4.5-8.3; however, the riboflavin molecules could be adsorbed in a less tilted orientation, almost with a perpendicular orientation, with respect to the uracil ring, toward the silver surface, Scheme 1B. At the pH range of 8.3-11.6, a flat orientation of the uracil ring, Scheme 1C, has been reported [20].

Considering the above facts, the low percent adsorption of riboflavin onto Ag/Fe\(_2\)O\(_4\) surface, observed at pH < 4.0 and pH > 7.0, is indicative of the low contribution of tilted and flat orientations of riboflavin molecules toward the surface of nanoparticles in an efficient adsorption
processing. Consequently, the perpendicular orientation should provide higher percent adsorption in the pH range of 4.0-7.0, as shown in Fig. 9.

Effect of Riboflavin/Nanoparticles Weight Ratio
Different concentrations of riboflavin were examined to find its optimum amounts that could be adsorbed onto a known amount (4.0 mg) of Ag/Fe$_3$O$_4$ nanoparticle surfaces under optimum experimental conditions. The results (Fig. 10) showed that adsorption percentage of riboflavin decreases from 94% for initial riboflavin/nanoparticles weight ratio of 0.004 to a 83.5% when riboflavin/nanoparticles weight ratio of 0.018 is examined. This study indicated that the nanoparticles’ active sites are saturated by riboflavin molecules when the riboflavin/nanoparticles weight ratio increases, consequently leads to the less adsorption of riboflavin. To follow optimization procedure, 4.0 mg of Ag/Fe$_3$O$_4$ and 2.0 ml of 0.02 mM riboflavin were used for the rest of the work.

Effect of Ionic Strength of Solution
Since the presence of any ion could affect the hydrophobic and electrostatic interaction between riboflavin and Ag/Fe$_3$O$_4$ nanoparticles, the effect of solution ionic strength on adsorption of riboflavin was investigated under optimum experimental conditions in batch technique. Individual beakers containing 2.0 ml of 0.02 mM riboflavin solution and 0.0 to 1000.0 mM NaNO$_3$ were prepared while the solution pH and Ag/Fe$_3$O$_4$ dosage were fixed at 6.0 and 4.0 mg, respectively, and the stirring time was 2.0 min. After the mixing time elapsed, the Ag/Fe$_3$O$_4$ nanoparticles were magnetically separated and each solution was analyzed for the residual riboflavin. The results showed that NaNO$_3$ has a significant effect on the adsorption of riboflavin; however, at concentrations higher than 50.0 mM NaNO$_3$, the adsorption percentage of riboflavin is almost independent of NaNO$_3$ concentration.

At NaNO$_3$ concentrations below 20.0 mM, no aggregation of Ag/Fe$_3$O$_4$ occurred while aggregation rate
Fig. 7. Effect of initial dosage of Ag/Fe$_3$O$_4$ nanoparticles on adsorption of riboflavin from 2.0 ml of 0.02 mM of its aqueous solution. Experimental conditions: pH 6.0, stirring time of 2.0 min. The error bars correspond to standard deviations of replicated analysis (n = 3).

Fig. 8. Effect of stirring time on adsorption of riboflavin from 2.0 ml of 0.02 mM of its aqueous solution. Experimental conditions: Ag/Fe$_3$O$_4$ dosages of 4.0 mg and pH of 6.0. The error bars correspond to standard deviations of replicated analysis (n = 3).

Fig. 9. Effect of initial pH on adsorption of riboflavin from 2.0 ml of 0.02 mM of its aqueous solution. Experimental conditions: Ag/Fe$_3$O$_4$ dosage of 4.0 mg and stirring time of 2.0 min and 1500 rpm. The error bars correspond to standard deviations of replicated analysis (n = 3).
increased at higher concentrations. At approximately 50.0 mM NaNO₃, the aggregation rate was achieved to a maximum value and further increase in NaNO₃ concentration had no appreciable effect on the aggregation process [22]. So, adsorption and sedimentation by aggregation took place simultaneously at concentrations more than 20.0 mM that results in a general decrease in adsorption percentage of riboflavin.

**Effect of Temperature**

The effect of temperature on the adsorption of riboflavin on Ag/Fe₃O₄ nanoparticles was investigated at an initial riboflavin concentration of 0.02 mM. The solution pH and Ag/Fe₃O₄ dosage were fixed at 6.0 and 4.0 mg, respectively, and a stirring time of 2.0 min was applied. The results indicated that solution temperature has no significant effect on the adsorption of riboflavin in the range of 288-318 K. The interaction of riboflavin with Ag/Fe₃O₄ nanoparticles is concluded to be of the van der Waals forces types [23]. This was also concluded from the adsorption isotherms results discussed in Section 3.12.

**Effect of Halide Anions**

To investigate whether halide anions have any effect on adsorption of riboflavin onto Ag/Fe₃O₄ nanoparticles, and also to prove that Ag layer has the main role in adsorption of riboflavin, different potassium halide solutions in the concentrations range of 100.0-1000.0 mM were individually tested with 0.02 mM riboflavin solution. The solution pH and Ag/Fe₃O₄ dosage were fixed at 6.0 and 4.0 mg, respectively, and a stirring time of 2.0 minutes was applied.

By increasing the KCl concentration, percent adsorption of riboflavin significantly decreased from 90% to 46.8%. In the case of KBr and KI, it decreased to 15.3% and 0.0%, respectively. The interactions of halides (1000.0 mM) with nanoparticles were in the order of Cl⁻ < Br⁻ < I⁻; This order of interaction follows the same order of reaction completeness between silver layer and anions upon the order of $K_{sp}$ values of their silver salts; i.e. $K_{sp}$ AgI < $K_{sp}$ AgBr < $K_{sp}$ AgCl.

Although silver is one of the stable metals, the following mechanism was proposed by Li [24] for dissolution of silver in the presence of KX salt.

\[
4Ag + O_2 \leftrightarrow 2Ag_2O(s) \\
Ag_2O(s) + H_2O \leftrightarrow 2Ag^+ + 2OH^- \\
Ag^+ + X^- \leftrightarrow AgX(s)
\]

Moreover; Henglein et al. reported that in a dilute aqueous suspension of silver nanoparticles, in the presence of atmospheric oxygen but in the absence of an electrolyte, dissolution of the produced Ag₂O is hindered due to the adsorption and accumulation of Ag⁺ at the particle surface [25].

**Adsorption Isotherm Modeling**

The adsorption isotherms of riboflavin onto the Ag/Fe₃O₄ surface were performed by adding various amounts of Ag/Fe₃O₄, in the range of 4.0-14.0 mg, to a series of beakers containing 2.0 ml of 0.20 mM riboflavin at pH = 6.0. The solutions were stirred for 2.0 min at 25 °C to attain the equilibrium condition. The aqueous solutions were analyzed for the residual riboflavin after applying the magnetic field for settlement of the nanoparticles. The amount of the riboflavin adsorbed onto Ag/Fe₃O₄ was calculated based on the following mass balanced equation:

\[
q_e = \frac{V(C_0 - C_e)}{m}
\]

where $q_e$ (in mg g⁻¹) is the adsorption capacity (mg riboflavin adsorbed onto gram amount of adsorbent), V (in l) is the volume of the riboflavin solution; $C_0$ and $C_e$ (in mg l⁻¹) are the initial and equilibrium riboflavin concentrations, respectively; and m (in g) is the mass of dried adsorbent.

The equilibrium adsorption data of riboflavin onto Ag/Fe₃O₄ nanoparticles surface were analyzed using Langmuir, Dubinin-Radushkevich and Freundlich models. The models fitted to equilibrium adsorption results of riboflavin were assessed by the values of the correlation coefficients ($R^2$) of the linear regression plots. Table 1 shows the summary of the constants and the corresponding
Fig. 10. Effect of weight ratio of riboflavin/nanoparticle on adsorption of riboflavin. Experimental conditions: Ag/Fe$_3$O$_4$ dosage of 4.0 mg, pH 6.0, stirring time of 2.0 min. The error bars correspond to the standard deviations of replicated analysis (n = 3).

Table 1. Adsorption Isotherms Parameters of Riboflavin onto Ag/Fe$_3$O$_4$ Nanoparticles

<table>
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<th>Dubinin-Radushkevich model</th>
<th>Freundlich model</th>
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<tr>
<td></td>
<td>$q_D$</td>
<td>$B_D$</td>
</tr>
<tr>
<td>Riboflavin</td>
<td>1.5</td>
<td>0.28</td>
</tr>
</tbody>
</table>

$q_D$ (in mg g$^{-1}$) is the Dubinin-Radushkevich isotherm constant, $B_D$ (in mol$^2$ kJ$^{-2}$) is related to the free energy of sorption per mole of the sorbate as it migrates to the surface of the adsorbent from infinite distance in the solution, $E$ (in kJ mol$^{-1}$) is apparent energy of adsorption, $K_F$ (in mg g$^{-1}$) and $n$ (in g l$^{-1}$) are the constants from the Freundlich equation representing the capacity of the adsorbent for the adsorbate and the reaction order, respectively.

Table 2. Adsorption Kinetic Constants of Riboflavin onto Ag/Fe$_3$O$_4$ Nanoparticles

<table>
<thead>
<tr>
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<th>Pseudo-first-order model</th>
<th>Pseudo-second-order model</th>
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<tbody>
<tr>
<td></td>
<td>$K_1$</td>
<td>$q_e$</td>
</tr>
<tr>
<td>Riboflavin</td>
<td>0.300</td>
<td>1.11</td>
</tr>
</tbody>
</table>

$q_e$ (in mg g$^{-1}$) is the amount of adsorbate adsorbed at equilibrium, $k_1$ (in min$^{-1}$) is the pseudo-first-order rate constant and $k_2$ (in g mg$^{-1}$ min$^{-1}$) is the pseudo-second order rate constant.
correlation coefficients of those models that best fitted; i.e. Dubinin-Radushkevich ($R^2 = 0.9721$) and Freundlich ($R^2 = 0.9571$). It should be noted that the Langmuir isotherm model showed an $R^2$ value of 0.9309. The value of adsorption energy ($E = 1.34 \text{ kJ mol}^{-1}$) indicates that the adsorption process is physical in nature [26].

**Adsorption Kinetic Modeling**

To study the adsorption kinetics of riboflavin onto Ag/Fe$_3$O$_4$ nanoparticles, Ag/Fe$_3$O$_4$ (4.0 mg) was incubated with 2.0 ml of the sample solutions (pH 6.0), containing 0.10 mM riboflavin, and the suspension was immediately stirred (1500 rpm) for different periods of time. Adsorption kinetic data were obtained by measuring the concentration of riboflavin in the sample solution at different time after removing nanoparticles.

To describe the adsorption behavior and the corresponding rate, the data obtained from adsorption kinetic experiments were evaluated using pseudo-first- and pseudo-second-order reaction rate models. The plots of the experimental results of riboflavin fitted to the selected adsorption models were constructed for both models. Table 2 gives a summary of the models and their corresponding constants along with the correlation coefficients for the linear regression plots of riboflavin.

As shown in Table 2, higher values of $R^2$ was obtained for pseudo-second-order adsorption rate model indicating that the adsorption rate can be more appropriately described using this model. This indicates that the rate of occupation of adsorption sites is proportional to the square of the number of unoccupied sites [27].

**CONCLUSIONS**

In this study, Ag/Fe$_3$O$_4$ nanoparticles were produced and examined as adsorbent for riboflavin. Results indicated that the synthesized nanoparticles could effectively adsorb riboflavin in a short period of 2.0 min. The optimum dosage of nanoparticles and pH of the sample solution were obtained to be 4.0 mg and 6.0, respectively, while a stirring time of 2.0 min in 1500 rpm was applied. Isotherm modeling revealed that Dubinin-Radushkevich and Freundlich models could better describe the adsorption of riboflavin onto Ag/Fe$_3$O$_4$ nanoparticles and the process is physical in nature. Kinetic data were appropriately fitted to the pseudo-second-order adsorption rate.

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