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Applying Kinetic Spectrophotometric Method and Artificial Neural Network Model for Determination of Metronidazole with *Albizia Lebbeck Leaves*-capped AgNPs Sensor in Blood and Urine Samples

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Abstract

The environmental pollution caused by drug antibiotic waste presents a foremost concern in the ecosystem, as high levels of these antibiotic drugs after consumption when released into the ecosystem, biological samples are accumulated and are producing overall contamination. Consequently, the need for selective, sensitive, fast, easy-to-handle, and low-cost early monitoring detection systems is growing. In this study, we used a prepared *Albizia Lebbeck Leaves*-capped AgNPs sensor to illustrate examples of friendly biosensors with their real application fields for the sensitive detection of the metronidazole drug in various matrices such as human fluids by kinetic spectrophotometric method. The calibration curve was linear in the range of (0.02 to 10.0 µg L^{-1}). The standard deviation of less than (3%), and detection limits (3S/m) of the method (0.02 µg L^{-1} in time 8 min, 367 nm) were obtained for sensor level response *Albizia Lebbeck Leaves*-capped AgNPs with (95%) confidence evaluated. The artificial neural network model was used as a tool very low for determining mean square error (MSE 0.061) for metronidazole drug by *Albizia Lebbeck Leaves*-capped AgNPs sensor. The observed outcomes confirmed the suitability of recovery and a very low detection limit for measuring the metronidazole drug. The method introduced to measure metronidazole drugs in real samples such as urine and blood was used and can be used for other drugs environmental pollution and hospital samples.

Keywords: Metronidazole, Determination, Kinetic Spectrophotometric, Sensor, Neural Network Model.

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Introduction

Drug delivery systems have been created for improving the therapeutic properties of the drugs and are often in form of a drug-containing capsule. Such systems release the drugs at a specific amount in a specific site, therefore they affect drugs' pharmacokinetics and distribution. Nanoparticles have been widely applied in drug delivery [1]. Determining the amount of drug used in the biological samples is very important to follow the amount of its effect on the body system. Accordingly, different methods with high sensitivity, selectivity, and efficiency, as well as appropriate analysis for the determination, extraction, and measurement are presented of drugs in real samples [2]. One of the biggest problems in the decomposition of biological samples is the existence of different species and their effect on the decomposition process of the drug. For this reason, many drug measurement methods are based on separation methods such as gas chromatography and high-performance liquid chromatography which are very time-consuming methods with difficult working conditions [3].

Metronidazole (1-(2-hydroxyethyl)-2 -methyl-5-nitroimidazole) shown in (Figure 1), is an antibiotic belonging to the nitroimidazole class which is prescribed to treat parasitic and bacterial infections in human beings and is commonly effective [4]. It is used in the aquaculture industry as a growth-promoting feed additive. High doses and long-term treatment with (MNZ) may be associated with the development of neutropenia, increased risk of peripheral neuropathy, ataxia, and seizure [5]. For that reason, precise and reliable calculation of rare (MNZ) in biological samples is crucial for guaranteeing consumers' health.

A diverse quantitative analytical method including electrophoresis [6], capillary gas chromatography [7], liquid chromatography [8], liquid chromatography coupled with mass spectrometry [9,10], and flow injection [11], have been reported among the most used methods in determining Metronidazole medicaments. Accordingly, developing a simple, quick, elective, and delicate method like spectrophotometric measurement in determining metronidazole was highlighted. In this method sensing metronidazole was done with high sensitivity and excellent selectivity for the discerning and accurate reorganization of species (1-inorganic 2- organic and 3-biomolecules) in different intricate matrices, and attention has been drawn to noble metal nanoparticles-based UV–visible spectrometric methods [12-14].

Due to the profitable application of metal nanoparticles, technologies have taken advantage of nano-scale materials in a variety of fields from chemistry to medicine [15,16]. The forms, sizes, and structures of metallic nanomaterials which are extensively linked to their chemical, physical, and optical characteristics, set the ground for successful use in technologies. In this respect, the exceptional physical, chemical, and biological properties of AgNPs have been widely used. This

exceptionality arises from the size, form, composition, crystallinity, and structure of AgNPs in comparison with their bulk form [17]. AgNPs can made be by various methods such as the application of stabilizing and reducing chemicals of hydrate and sodium borohydride, formaldehyde, polyethylene glycol, glucose, electrochemical heating, and photochemical reduction prepared [18]. Therefore, measures have been taken to investigate their exclusive properties and employ them in practical applications like anti-bacterial and anti-cancer therapeutics [19], diagnostics and optoelectronics, water disinfection [20], and other clinical/pharmaceutical applications [21]. Furthermore, AgNPs have been widely used in antimicrobial applications due to their antimicrobial properties. The exclusive properties of AgNPs have applications in the fields of biosensing, nanomedicine, pharmacy, and biomedical engineering varying sizes and shapes have been utilized in a broad range of applications and medical equipment, such as electronic devices, paints, coatings, soaps, detergents [22].

This study was to evaluate the use of an artificial neural network (ANN) model to predict the determination (MNZ) drug in solution by *Albizia Lebbeck Leaves*-capped AgNPs. ANN is a numerical estimation technique, which is used to find nonlinear relationships between input and output variables. In this research, to measure (MNZ) drug, various effective factors such as (pH, (MNZ) drug concentration, *Albizia Lebbeck Leaves*-capped AgNPs concentration, time reaction, etc.) on the response of the method and obtaining the optimal test values and obtaining the linear range, detection and accuracy of the method presented in the measurement of (MNZ) drug as well as comparing the performance of the method with routine clinical techniques and checking the accuracy of the method and the identification and measurement of (MNZ) drug by a kinetic Spectrophotometric method in real samples (blood serum) were checked. The chemical *Albizia Lebbeck Leaves*-capped AgNPs sensor made it possible as an excellent sensor with reproducibility, good recovery, and a very low detection limit for measuring (MNZ) drug. The neural network model to measure (MNZ) drugs in real samples such as urine and blood was used and can be used for other drugs environmental pollution and hospital samples.

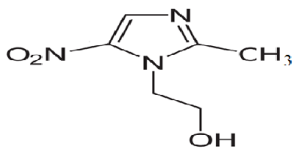

Figure 1. The structures of Metronidazole (MNZ).

Experimental

Materials

All chemicals including Silver nitrate ($AgNO_3$) (98%), Sodium borohydride ($NaBH_4$) (99%), were purchased from Merck Company while metronidazole medication (98.0%) was purchased from (Company Razi, Iran). Again, Universal buffer solutions were prepared from 1 ml of boric acid /acetic acid / phosphoric acid (1.0 M). The final pH was adjusted by the addition of 0.2M sodium hydroxide, was bought from Merck Company (Merck, Darmstadt, Germany). DD H_2O (double distilled water) was used in the preparation of the solutions.

Instrumentation

UV–visible spectra and drugs concentrations were determined and their measurements were done using a Maya Pro 180 spectrophotometer (Shimadzu Company, Japan). The registration of FT-IR or Fourier transform infrared spectra were done on a PerkinElmer (FT-IR spectrum BX, Germany). SEM (Scanning electron microscopy: KYKY-EM 3200, Hitachi Firm, China) under an acceleration voltage of 26kV) was used to study the morphology of samples. For the measurement of pH, the pH/Ion meter (model-728, Metrohm Firm, Switzerland, Swiss) was employed.

Pretreatment of real samples

In a 100 mL beaker, treatment of a 50 mL portion of human fluids (urine or blood) samples was done using 2 mL of concentrated HNO_3 (63%) and an $HClO_4$ (70%) mixture of 2:1 and then covered with a watch glass. for 10 min and then with the help of a 100 mL volumetric flask, desired. 5 mL of the obtained clear solution was picked and the analysis metronidazole drug was found by standard addition method procedure [23,24].

Synthesis of Albizia Lebbeck Leaves-capped AgNPs

In this regard, the following details of the materials are important to consider in their synthesis: surface property, size distribution, apparent morphology, particle composition, and dissolution rate. *Albizia Lebbeck Leaves*-capped AgNPs were prepared by the reduction of $AgNO_3$ with $NaBH_4$ as a modifier according to the method in the literature [25]. Briefly, 10.0 mL of *Albizia Lebbeck Leaves* (0.1 mM) solution was added into the reaction flask that contained 90.0 mL of $AgNO_3$ (0.1 mM) solution under vigorous stirring. After (15 min) was UV–visible spectrum of *Albizia Lebbeck Leaves*-capped AgNPs. The inset picture shows *Albizia Lebbeck Leaves*-capped AgNPs. added into the above solution at room temperature and stirred for 1 h. The dark colloidal solution color was changed to bright yellow, confirming that the formation of *Albizia Lebbeck Leaves*-capped AgNPs. The *Albizia Lebbeck Leaves*-capped AgNPs solution was stored in the dark at ($4.0 \pm 2.0^{\circ}C$) to remain stable for several weeks (Figure 2).

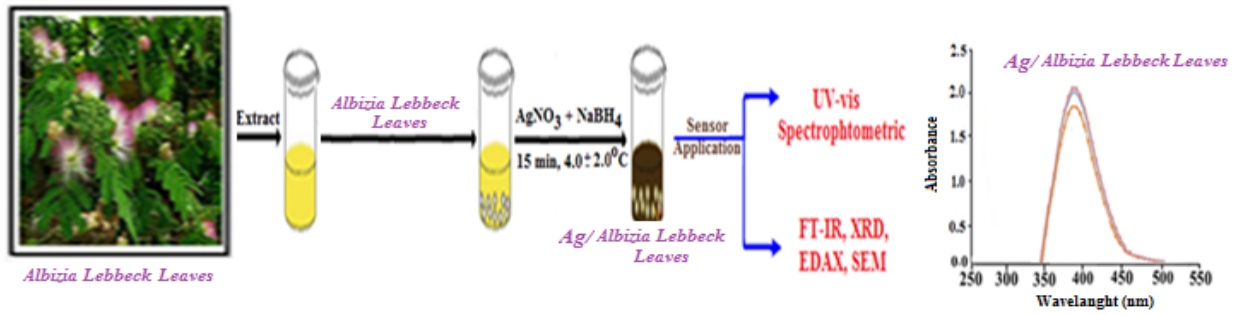

Figure 2. Synthesis of *Albizia Lebbeck Leaves*-capped AgNPs**.**

Artificial neural network

In the applied artificial neural network (ANN), feed-forward back propagation (FFBP) was employed as the training algorithm. The final computed data for FFBP were juxtaposed against the experimentally attained findings. Afterward, by computing and back propagating the errors, they finally were employed for adjusting each neuron weight. A model ANN with four input layers (initial MNZ drug concentration, *Albizia Lebbeck Leaves*-capped AgNPs dosage, pH, and time) was detected at epoch numbers 16, and 22 experimental points were employed to feed the model shown in (Figure 3). The MSE based on the function of error performance showed a minimum value at 5 neurons, from training, test, and data points, respectively, and the data set was derived. In the present study, the Levenberg-Marquardt backpropagation algorithm was used for the network. In the present study, the Levenberg-Marquardt backpropagation algorithm was used for the network [26,27].

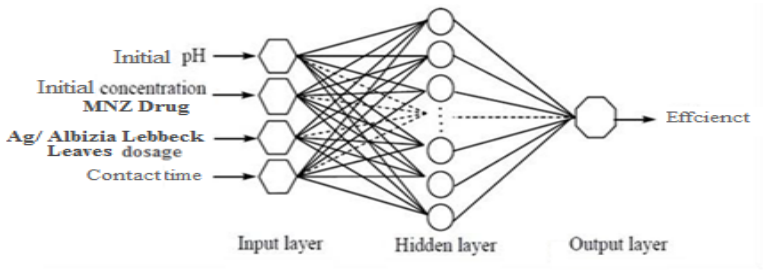

Figure 3. The architecture of neural network system.

This algorithm accelerates attaining convergence and provides the accuracy of the results [27,28]. The tan sigmoid (Eq.1) and linear transfer functions were used for transfer functions in the hidden and output layers. All samples were normalized between 0-1, to achieve fast convergence, and commensurability of the scale of the input, as well as the minimal RMSE values. The normalized values of data were obtained according to Eq. (1):

$$X_{norm} = \frac{X - X_{min}}{X_{max} - X_{min}} \quad (1)$$

Where X is a variable, but X_{min} and X_{max} refer to the minimum value and the maximum value, respectively. Statistical parameters MSE, SSE, AARE, and R^2 are calculated through equations (2-۰), respectively [26-28]:

$$MSE = \frac{\sum(Y_{model,i} - Y_{exp,i})^2}{n} \quad (2)$$

$$SSE = \sum(Y_{model,i} - Y_{exp,i})^2 \quad (3)$$

$$AARE = \frac{1}{n}\sum\left(\left|\frac{Y_{exp,i} - Y_{model,i}}{Y_{exp,i}}\right|\right) \quad (4)$$

$$R^2 = \frac{\sum(Y_{exp,i} - Y_{model,mean})^2 - \sum(Y_{model,i} - Y_{exp,i})^2}{\sum(Y_{exp,i} - Y_{model,mean})^2} \quad (۰)$$

Where mean squared error (MSE) and correlation coefficient (R^2).

Procedure of kinetic spectrophotometric detection

The ensuing steps have been considered for a kinetic spectrophotometric method in the current study at the initial step: the sample solution containing 1 mL of (MNZ) drug (10 µg L^{-1}) was added to a 10 mL volumetric beaker. Then, 1 mL of sodium borohydride as a stabilizer for the sensor (1.5 × 10^{-2} M) was spiked into the flask. In 5 s after the addition of 1 mL of *Albizia Lebbeck Leaves*-capped AgNPs solution (1.5 × 10^{-2} M) into a beaker, the solution was stirred for 30 s. Subsequently, an adequate amount of the solution was added to a 1 cm cell. Finally, using UV-visible spectroscopy, the absorbance was measured at 367 nm in a time interval of 1.0–8.0 min. By adding (MNZ) drug to the solution, the *Albizia Lebbeck Leaves*-capped AgNPs absorbance at the wavelength of 367 nm was observed to decrease. At the same time, the apparent spectral evolution including the formation of a well-defined isosbestic point at around 367 nm was estimated using spectrophotometry. All reaction steps were repeated by increasing (MNZ) drug concentration (0.2 µg L^{-1}) every 30 s. Moreover, the mentioned steps were repeated for a reaction in the absence of (MNZ) drug (Abs b). Eventually, the drug absorbance (Abs a) was calculated as the difference between the absorbance values in the presence and absence of (MNZ) drug The reaction of (MNZ) drug with *Albizia Lebbeck Leaves*-capped AgNPs was detected in the acidic medium at the wavelength of 376 nm. Figs. 4, demonstrate the absorption spectra in an aqueous solution [28]. The obtained results show that the slope of the curve changes at 1:1 [M]/[L], which indicates that the stoichiometry of the complex formed between *Albizia Lebbeck Leaves*-capped AgNPs and (MNZ) drug is 1:1. As can be seen in (Figure 4) [29,30].

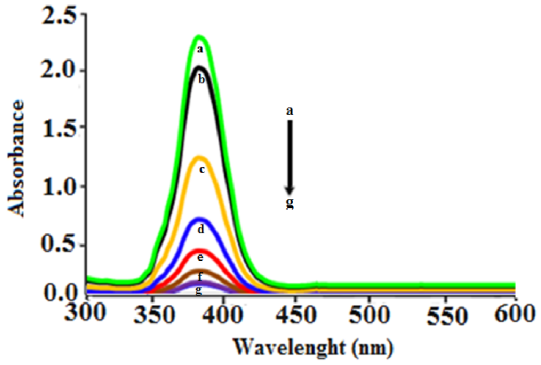

Figure 4. The absorption *Albizia Lebbeck Leaves*-capped AgNPs and (MNZ) drug 30 sec, and increasing concentration of (MNZ) drug solution (0.2 µg L^{-1}).

Results and discussion

Characterization of sensor

In (Figure 5), the FTIR spectrum of activated carbon prepared from *Albizia Lebbeck Leaves*-capped AgNPs nanoparticles is shown. Additionally, the observed absorption signal at 3451 cm^{-1} point to O-H groups' presence because of the alcoholic or phenolic functional groups. Also, the presence of C-H groups is well proven by the signal observed at 3101 cm^{-1}. Correspondingly, the C=O active group's presence is confirmed by the signal observed at 2158 cm^{-1}. The signal at 776.8 cm^{-1} is relevant to the Ag-O group of the *Albizia Lebbeck Leaves*-capped AgNPs nanoparticles. Scrutinizing these functional groups is of great consequence since complexion, or electrostatic attraction between the metal ions and varied surface oxygen-containing functional groups carried by the activated carbons leads to the elimination of heavy metals [31]. Different X-ray emission peaks are *Albizia Lebbeck Leaves*-capped AgNPs shown in (Fig. 6). The signals at 38.5(122), 45.0(111), 52.2(200), 54.4 (231), and 72.7 (220) as shown in the XRD pattern of the *Albizia Lebbeck Leaves*-capped AgNPs [32]. The perfect crystalline nature of the material was proven after functionalizing with *Albizia Lebbeck Leaves*-capped AgNPs however the great intensity of the signal at 45.0 (111) confirmed that there has been a slight amount of material in an amorphous state. The perfect synthesis of *Albizia Lebbeck Leaves*-capped AgNPs is obvious by looking at the XRD pattern. In (Figure 7), the morphological properties of the samples scrutinized by SEM are exhibited. By looking at (Figure 7), the smoothness, homogeneity, and tidiness of *Albizia Lebbeck Leaves*-capped AgNPs are confirmed. Even uniformity size distribution is observable in (Figure 7). After surface modification, the *Albizia Lebbeck Leaves*-capped AgNPs became uneven, larger, and bundled [33].

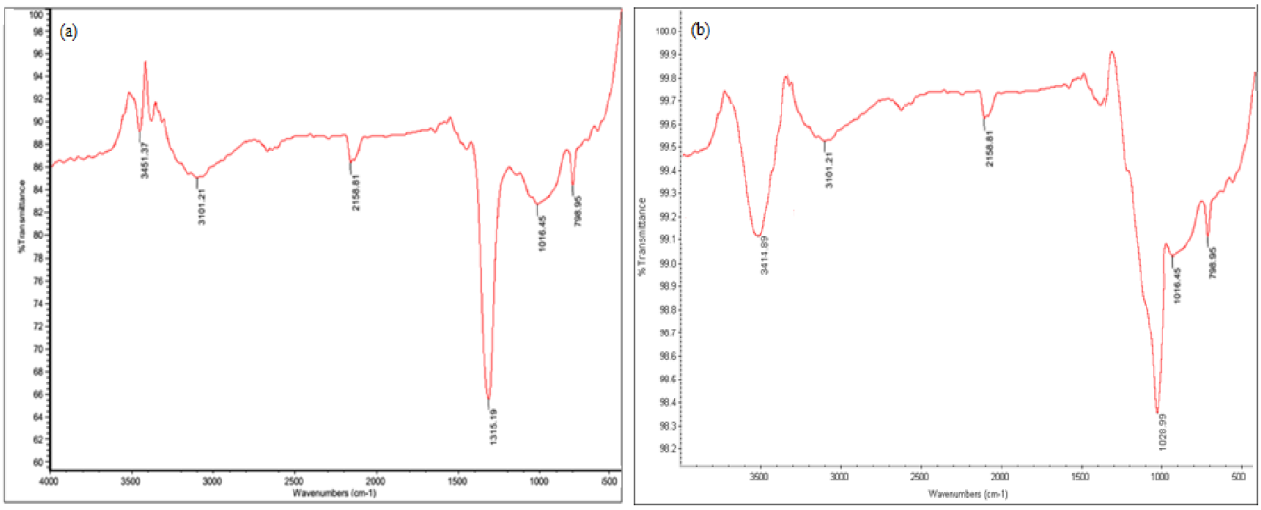

Figure 5. (a) FTIR spectra of the prepared *Albizia Lebbeck Leaves* (b) FTIR spectra of the prepared *Albizia Lebbeck Leaves*-capped AgNPs.

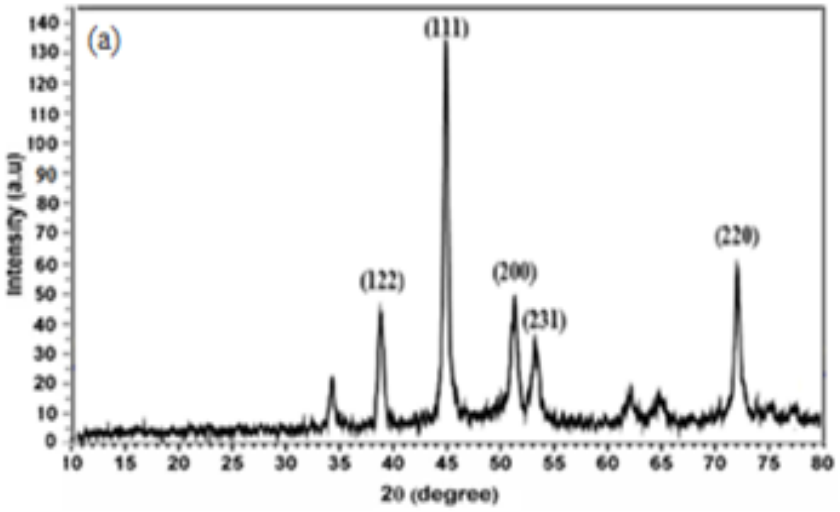

Figure 6. The XRD image of the prepared *Albizia Lebbeck Leaves*-capped AgNPs.

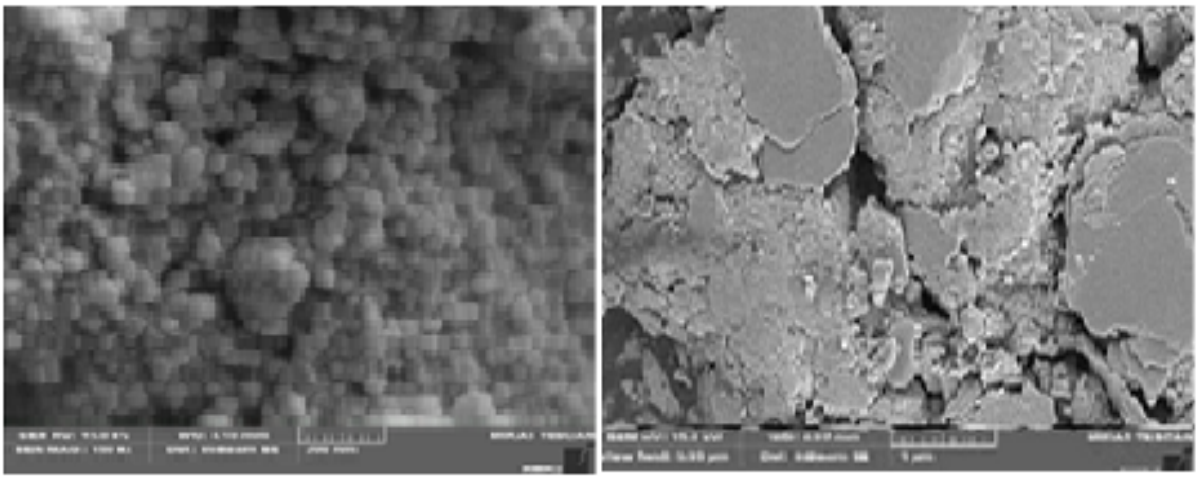

Figure 7. The (SEM) image of the prepared *Albizia Lebbeck Leaves*-capped AgNPs.

Optimization of Sensing Conditions

It would be interesting to know that in the presence of (MNZ) medicaments, there observed a considerable improvement in the effectual colorimetric sensing and absorbance kinetic Spectrophotometric method of the as-prepared *Albizia Lebbeck Leaves*-capped AgNPs. Obtaining an exceptionally sensitive response in detecting (MNZ) medicament rests upon the systematic optimization of pH values, *Albizia Lebbeck Leaves*-capped AgNPs, and incubation time.

In this section, the best type of buffer and its volume for maximum absorption (MNZ) drug with *Albizia Lebbeck Leaves*-capped AgNPs sensor is investigated. For this step, the procedure is as follows: In 10 ml balloons, separately 1 ml of (MNZ) drug (10.0 µg L^{-1}) and a volume of each type of acetic acid / boric acid / phosphoric acid buffer and then 1 ml of 1 ml *Albizia Lebbeck Leaves*-capped AgNPs (2.5×10^{-2} M), 1 ml of utilizing sodium borohydride as a stabilizer for sensor (1.5×10^{-3} M), to the solution inside the balloon and after (8 minutes), the adsorption of the solution was read by UV–Visible spectrophotometry. The results are shown in (Figure 8). Based on the results, 1 ml of acetic acid buffer shows the highest percentage for the determination of (MNZ) drug, so acetate (acetic acid/tri chloric acetate) buffer (1.0 M) to adjust the pH solution as the optimal buffer.

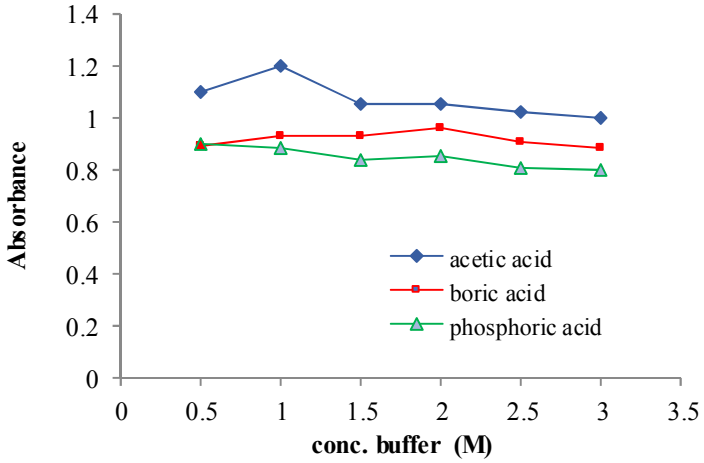

Figure 8. The impact of buffer concentration on the absorbance. (aqueous sample volume, 10 mL: *Albizia Lebbeck Leaves*-capped AgNPs, 2.5×10^{-2} M, sodium borohydride, 1.5×10^{-3} M, (MNZ) drug = 10.0 µg L^{-1}, time 8 min, 367 nm).

The great impact of the pH value of the reaction solution on the interaction between *Albizia Lebbeck Leaves*-capped AgNPs and (MNZ) medicament was undeniable. To examine the influence of *Albizia Lebbeck Leaves*-capped AgNPs on the reaction rate, 1ml (MNZ) medicament (10.0 µg L^{-1}) solution, *Albizia Lebbeck Leaves*-capped AgNPs (2.5×10^{-2} M), and 1 ml sodium borohydride (1.5×10^{-3} M) were mixed in the 10 ml volumetric flask using DD H_2O (Double distilled water).

After measuring the absorbance intensity of the solution, a thorough investigation was carried out on the absorbance pH values in the range of (2-9) for the (MNZ) medicament- *Albizia Lebbeck Leaves*-capped AgNPs complex at (367 nm). As evident in (Figure 9a), absorbance kinetic Spectrophotometric rapidly on changing the pH from 1.0 to 6.0, while it decreased at pH values higher than 6.0. This phenomenon might be because of the weak complexion at lower pH values (pH < 6.0). On the other hand, the reduced response of the proposed *Albizia Lebbeck Leaves*-capped AgNPs sensor for determining (MNZ) at pH > 6.0 could be due to a possible formation of the hydroxide of MNZ drug in solution. Thus, pH 6.0 was selected as a favorable pH for all subsequent experiments [34]. Concurrently, 1 ml (MNZ) medicament (10.0 μg L^{-1}) solution, 1 ml sodium borohydride (1.5×10^{-3} M), and 1ml *Albizia Lebbeck Leaves*-capped AgNPs (0.5×10^{-3} to 4.0×10^{-2} M), were mixed in a volumetric flask 10 ml using DD H_2O (Double distilled water) to find out about the impact of *Albizia Lebbeck Leaves*-capped AgNPs on the reaction rate. Again the absorbance intensity of the solution was assessed. The previously mentioned operation has been replicated for blank solution (the solution in the absence of (MNZ) medicament). The findings are exhibited in (Figure 9b). Consequently (2.5×10^{-2} M) based on those findings was determined as the perfect concentration [29,34].

To look over the efficacy of sodium borohydride concentration, with a help of a volumetric flask 10 ml firstly 1 ml (MNZ) medicament (10.0 μg L^{-1}) solution, 1 ml sodium borohydride with different concentrations (0.05 to 3.0×10^{-3} M), and 1 ml *Albizia Lebbeck Leaves*-capped AgNPs (2.5×10^{-2} M) Again after (8.0 min) sharp, the estimation for sorption of the solution was carried out. The aforementioned steps above were repeated in the absence of (MNZ) medicament (blank solution). The findings are exhibited in (Fig. 9c). The decision on desired concentration for sodium borohydride based on the results was made to be (1.5×10^{-3} M). Also, the impact of reaction time on the absorbance spectrum was investigated. Based on (Fig. 9d), it has become apparent that the absorbance intensity enhanced expeditiously and reached its peak at around 8 min. After 8 min, relative stability was spotted in the absorbance intensity. Thus, 8 min was determined as the perfect reaction time in this experiment [34,35].

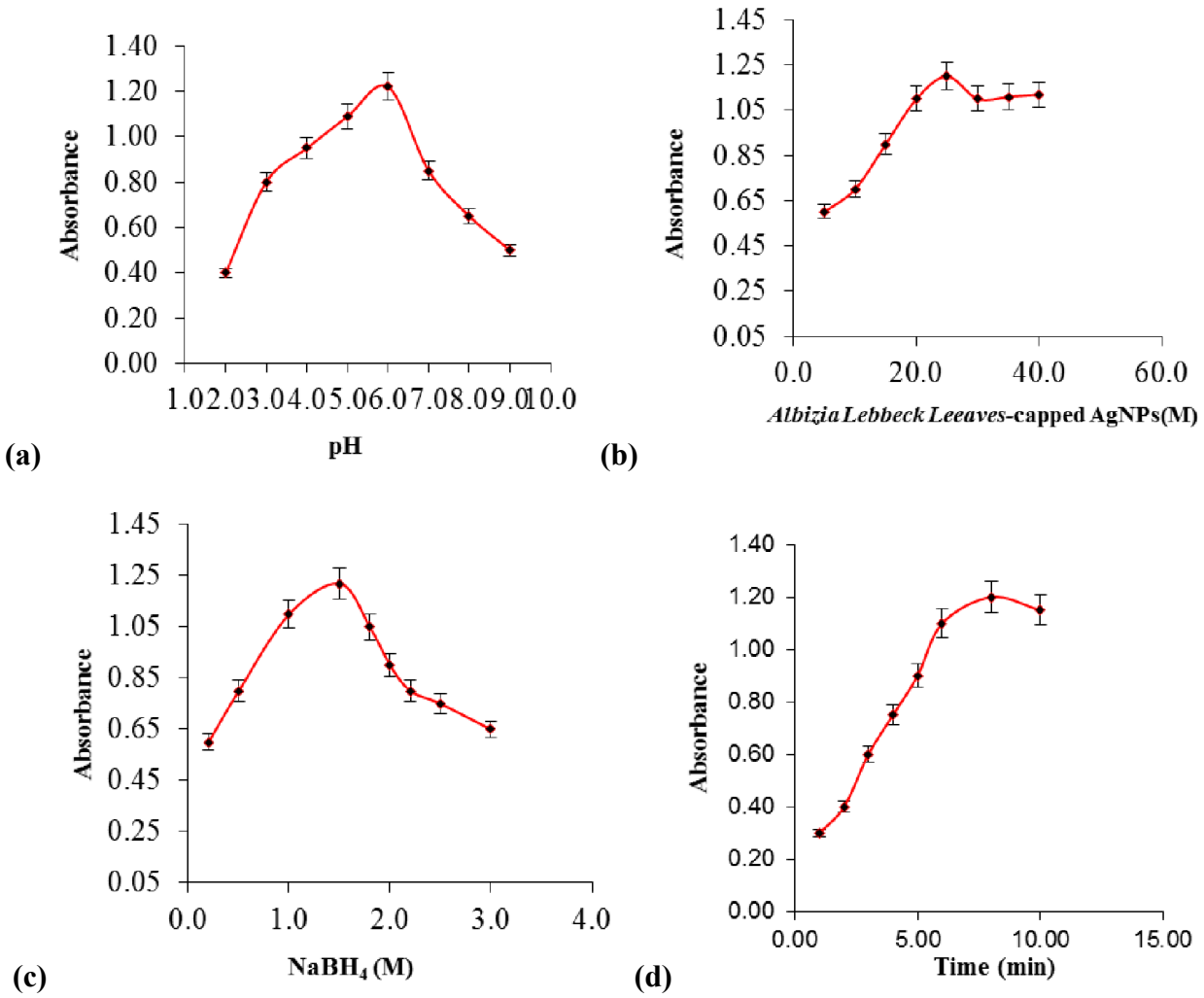

Figure 9. (aqueous sample volume, 10 mL, MNZ drug 10.0 μgL^{-1}, 367 nm): (a) the impact of pH in the absorbance rate, *Albizia Lebbeck Leaves*-capped AgNPs, 2.5×10^{-2} M, sodium borohydride, 1.5×10^{-3} M, time 8 min). (b) The impact of *Albizia Lebbeck Leaves*-capped AgNPs in the absorbance rate. Sodium borohydride, 1.5×10^{-3} M, pH 6, time 8 min). (c) The impact of sodium borohydride in the absorbance rate. *Albizia Lebbeck Leaves*-capped AgNPs, 2.5×10^{-2} M, pH 6, time 8 min). (d) The impact of time in the absorbance rate. *Albizia Lebbeck Leaves*-capped AgNPs, 2.5×10^{-2} M, sodium borohydride, 1.5×10^{-3} M, pH 6).

Analytical figures of merit

After optimizing the factors affecting the measurement of (MNZ) medicament, the calibration curve was plotted under optimized conditions. The absorption intensity of (MNZ) medicament is linearly related to its concentration in the range of (0.02–10.0 μg L^{-1}) shown in (Figure 10), with the following equation: $y = 0.0493x + 0.0605$ ($R^2 = 0.9960$), where x is (MNZ) medicament concentration (μg L^{-1}).

Also, for 7 replicate measurements a (10.0 μg L^{-1}) of (MNZ) medicament solution under optimized conditions, the relative standard deviation (R.S.D) for the response of *Albizia Lebbeck Leaves*-capped AgNPs towards a (10.0 μg L^{-1}) of (MNZ) medicament of less than (3%), and reproducibility

of the response of different starch-capped AgNPs was also studied. The detection limit for the response of *Albizia Lebbeck Leaves*-capped AgNPs was (0.02 μg L^{-1}) [29,36].

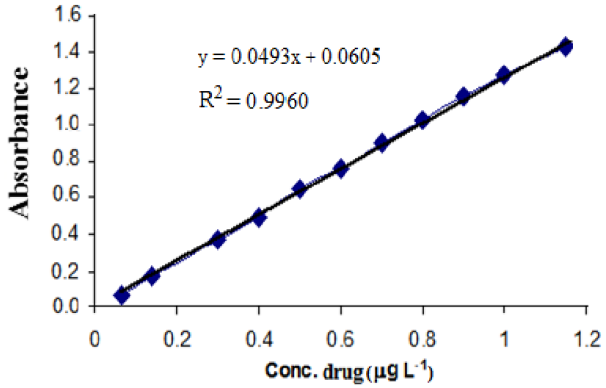

Figure 10. Calibration graph for (MNZ) medicament. (aqueous sample volume, 10 mL: *Albizia Lebbeck Leaves*-capped AgNPs, 2.5×10^{-2} M, sodium borohydride, 1.5×10^{-3} M, 367 nm).

Modeling of Artificial neural network process

In this study, the tan-sigmoid and linear transfer functions were applied for the transfer function in the hidden and output layers. During training, the predicted output was compared with the expected output. To determine the optimal number of neurons in the hidden layer, several neural networks were created with epoch numbers 16 and 22 neurons. The functions of weight (training), net input, and transfer (tansig) governed the performance of the network. The neural network model with 5 neurons in the hidden layer had the lowest MSE (0.061) for the test data and 22 experimental points were employed to feed the model for determining MNZ drug. In the (Figures 11 and 12). During the net training process in this study, the MSE based on the function of error performance showed a minimum value at 5 neurons [27,28]. A model ANN with four input layers (viz., initial (MNZ) drug concentration, Ag/ *Albizia Lebbeck Leaves* dosage sensor, pH, and time) based on the output layers (determining target compounds) was found to be adequately accurate for predicting and estimating the (MNZ) drug determination with MSE and the correlation coefficients (R^2) in (Table 1), for train, test, and validation for (MNZ) drug determining, respectively.

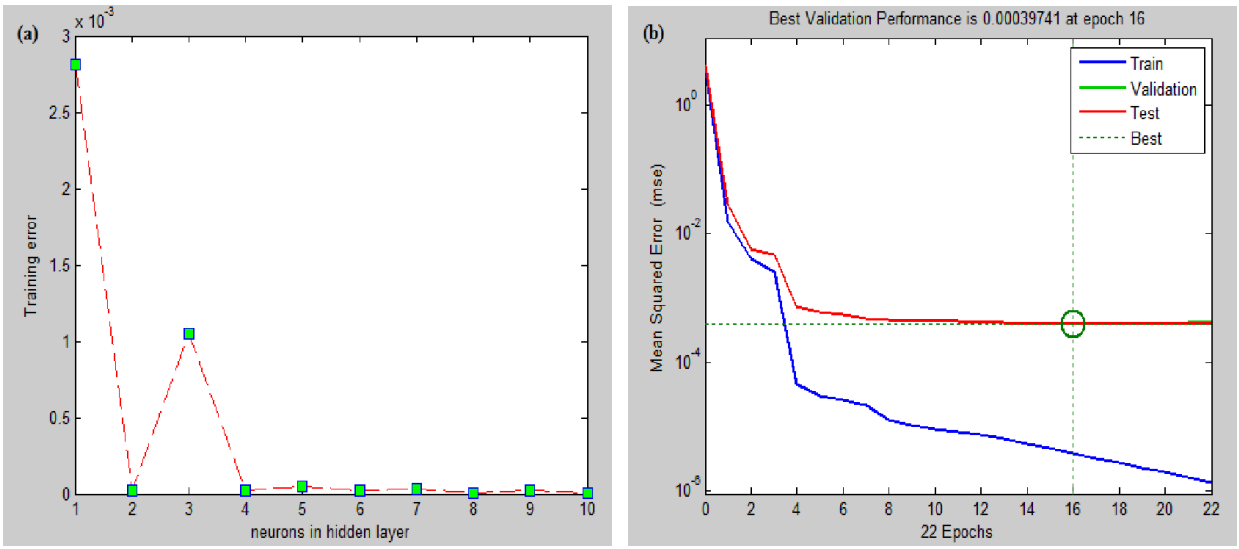

Figure 11. Evolution of training, validation, and test errors as a function of the number of training epochs during ANN for the (MNZ) drug determining by *Albizia Lebbeck Leaves*-capped AgNPs sensor.

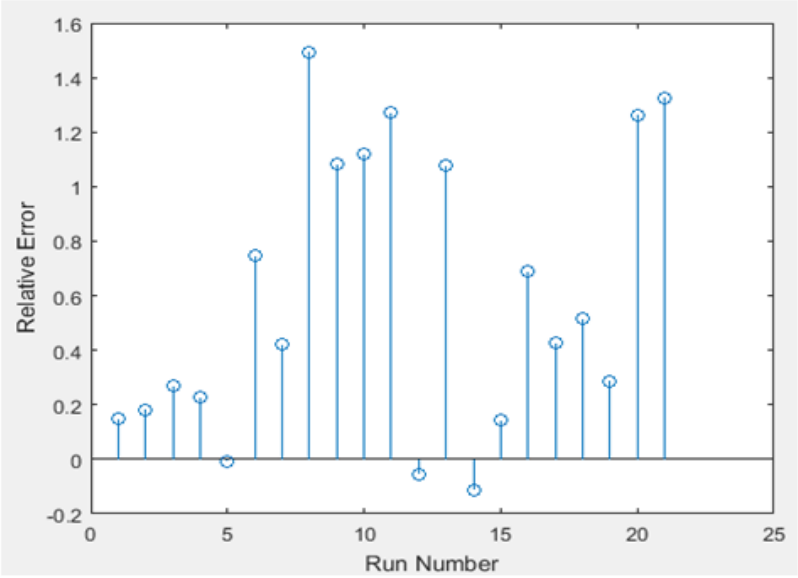

Figure 12. The exhibition of the plot of Relative error histogram for permeability on the (MNZ) drug determined by *Albizia Lebbeck Leaves*-capped AgNPs sensor.

Table 1. Statistical comparison of three models of artificial neural network.

Modeling	SSE	MSE	AARE	SD	R^2
ANN	1.275	0.061	0.036	0.049	0.9998

The summary results relative to the ANN neural network model, the sum of the squares of error, the mean of the sum of the squares of error, the mean of the absolute error, the standard deviation, and the correlation coefficient model in Table 1, are compared. The lowest determination error (MNZ) drug by *Albizia Lebbeck Leaves*-capped AgNPs sensor in demonstrated Process.

Optimum values of parameters

The optimum values of parameters are demonstrated in Table 2. The method can be used as an alternative method for (MNZ) medicament measurement owing to advantages like excellent selectivity and sensitivity, low cost, simplicity, low detection limit, and no need in utilizing an organic harmful solvent.

Table 2. Investigation of method repeatability at conditions.

Parameter	Optimum Value for metronidazole drug
metronidazole drug (M)	(10.0 µg L^{-1})
Albizia Lebbeck Leaves-capped AgNPs (M)	(2.5×10^{-2} M)
concentration $NaBH_4$(M)	(1.5×10^{-3} M)
pH	6.0
Equilibration time (min)	(8.0 min)
Linear range (LDR)	(0.02 - 10.0 µg L^{-1})
The detection limit (LOD)	(0.021 µg L^{-1})
Relative Standard Deviations (RSD)	Less than (3%)
Mean Square Error (MSE)	(0.061)
Advantages	High repeatability, sensitivity, selectivity, accuracy, precision, wide linear range, and no need to use organic solvents

Investigation of competition of (MNZ) medicament with other ions

The study was performed in the presence of drug, vitamins, cation ions, and anion ions in a solution containing (10.0 µg L^{-1}) of MNZ medicament in pH 6.0, and phosphate buffer (0.1 M), and (2.5×10^{-2} M) *Albizia Lebbeck Leaves*-capped AgNPs sensor and (1.5×10^{-3} M), sodium borohydride, time 8 min in 367 nm), and the impact of a different drug, vitamins, ions on the absorbance spectrum was investigated. The results are shown in (Figure 13) [37,38].

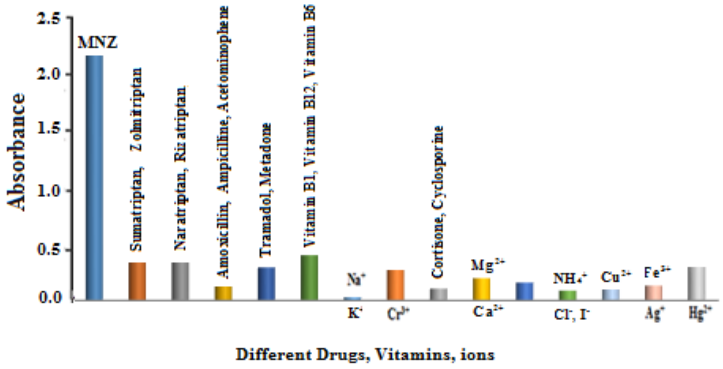

Figure 13. Impacts of the different drug, vitamins, ions on the determination of the (MNZ) drug (aqueous sample volume, 10 mL: phosphate buffer (0.1 M), *Albizia Lebbeck Leaves*-capped AgNPs 2.5×10^{-2} M, sodium borohydride 1.5×10^{-2} M, pH 6, time 8 min in 367 nm).

Application of the real sample

To evaluate the efficiency of the proposed sensor for determining (MNZ) drug in real samples, this *Albizia Lebbeck Leaves*-capped AgNPs sensor was used to measure (MNZ) drug in urine and blood human samples according to the instructions mentioned for (MNZ) drug experiment 3 replicates measuring section [39]. The obtained percentage percentiles in (Table 3), indicate that the prepared sensor has a very good performance for determining the drug (MNZ) drug in urine and blood samples. Therefore, the determination of (MNZ) drug in samples was confirmed utilizing the standard addition method. The level of the (MNZ) drug was estimated to be below the detection limit of the related element. Based on the outcomes of replicating analyses for each sample, it was shown that the medication retrievals were mainly quantitative with a low RSD. The potentiality of the recommended method for the determination of trace quantities of these elements in distinct samples was proven [29,40].

Table 3. Retrieval of trace (MNZ) drug from urine and blood samples after applying the presented procedure (N=3).

Samples	Added ($\mu g\ L^{-1}$)	Founded ($\mu g\ L^{-1}$)	RSD %	Recovery %
Urine	0.00	0.064	3.5	----
	0.10	0.162	2.6	98.0
Blood	0.00	0.084	2.3	----
	0.10	0.185	2.8	101.0

Conclusion

The investigation in this article focused on measuring the amount of trace (MNZ) drug utilizing *Albizia Lebbeck Leaves*-capped AgNPs sensor, in the company of utilizing sodium borohydride as a stabilizer sensor. A successful analytical method for measuring (MNZ) drug was prosperously developed via utilizing a sensitized spectrophotometric with the help of *Albizia Lebbeck Leaves*-capped AgNPs. The method can be used as an alternative method for (MNZ) drug measurement owing to advantages like excellent selectivity and sensitivity, low cost, simplicity, low detection limit, and no need in utilizing organic harmful solvents or extracts.

The neural network model was used for building up an empirical model, and the obtained results revealed that the neural network model was a powerful tool for the determination of mean square error relative (MSE), relative error, the mean of the sum of the squares of error, the mean of the absolute error, the standard deviation, and the correlation coefficient for the model are compared in summary. The lowest determining error (MNZ) drug could be obtained in a short time, which

strongly confirms the greater contribution for the deletion of (MNZ) drug by *Albizia Lebbeck Leaves*-capped AgNPs sensor, and some of the advantages of this biosensor,

- The use of waste *Albizia Lebbeck Leaves*-capped AgNPs sensor as a natural and inexpensive valuable resource and environmentally benign support.
- The use of sodium borohydride as an economic and effective reducing and stabilizing agent.
- *Albizia Lebbeck Leaves*-capped AgNPs sensor provides several advantages such as simple, mild condition, easy workup, and excellent yield in a short time.

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Conflict of interest

The authors declare that they have no conflict of interest related to the publication of this article.

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