

Spectrophotometric Determination of Dopamine Using Curcumin Nanoparticles in the Presence of Copper(II) Ions

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Abstract

A simple, sensitive, and cost-effective spectrophotometric method for the determination of dopamine was developed using curcumin nanoparticles (curcumin NPs) in the presence of copper(II) ions. Curcumin, a natural polyphenolic compound, exhibits notable optical properties that were exploited to form a colored complex with Cu (II). The addition of dopamine increased the absorbance intensity of this complex through specific interactions between dopamine molecules and the curcumin-Cu-Cu(II) system. The experimental parameters were optimized to include a pH of 8.5, an ammonia buffer volume of 1.5 mL, a Cu(II) concentration of 20 mg·L⁻¹, a reaction time of 40 minutes, and a curcumin NPs volume of 0.7 mL in a final volume of 10 mL. Under these conditions, the calibration graph for dopamine was linear in the range of 0.5–20 mg·L⁻¹, with a high correlation coefficient ($r = 0.9994$). The regression equation was $\Delta A = 0.0062 C + 0.0743$, where ΔA is the absorbance difference in the presence and absence of dopamine, and C is the dopamine concentration in mg·L⁻¹. This method demonstrates good linearity, sensitivity, and reproducibility, offering a promising approach for dopamine detection in clinical and pharmaceutical applications.

Keywords: Calibration curve; Colorimetric analysis; Copper (II); Curcumin nanoparticles; Dopamine determination; Spectrophotometry.

1. Introduction

Nanomaterial synthesis and morphology are critical factors that influence the physicochemical properties of nanostructures. Modifying their structure can significantly enhance or alter properties such as optical behavior [1]. Considerable research has been devoted to developing synthesis techniques and characterizing

nanostructures [2]. Despite advancements, challenges remain in dealing with active metals such as silver (Ag) and copper (Cu), which are prone to environmental degradation and often require surfactants during colloidal synthesis to protect them [3]. Nanotechnology, first introduced by Richard P. Feynman in 1959 [4], has since revolutionized material science by enabling the manipulation of matter at the nanoscale. Nanoparticles (NPs), defined as materials with at least one dimension below 100 nm [5], may exist in one-dimensional (1D), two-dimensional (2D), or three-dimensional (3D) structures [6]. Their unique size-dependent properties, particularly their optical behaviour, have opened new opportunities for applications such as bioimaging and sensing. For example, gold (Au), silver (Ag), palladium (Pd), and platinum (Pt) nanoparticles exhibit distinct colors depending on their size and morphology [7].

Contributions of this work:

The present study introduces a novel, sensitive, and cost-effective spectrophotometric method for the determination of dopamine using curcumin nanoparticles in the presence of copper(II). Unlike existing techniques, the proposed method provides high linearity, reproducibility, and simplicity, making it suitable for clinical and pharmaceutical applications.

As shown in Figure 1(d), the color of the solution changes due to variations in nanoshell thickness, aspect ratio, and gold concentration. Alterations in any of these factors affect the nanoparticles' optical absorption characteristics, leading to observable differences in colour [8].

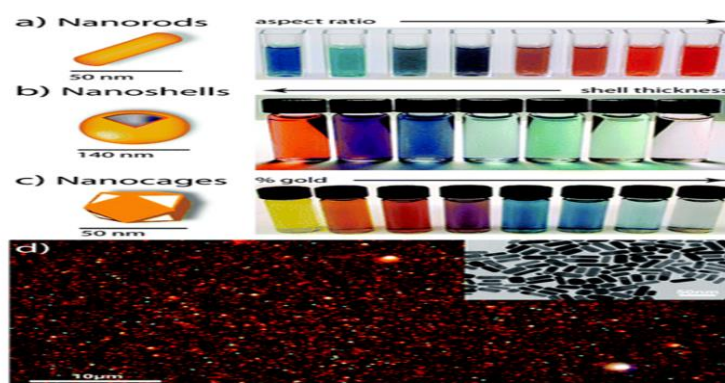


Figure 1 (a,b,c,d): The color of Au NPs influenced by their size and shape.

2. Solution preparations

2.1. Preparation of curcumin NPs

The curcumin NPs were fabricated by modification of the technique previously described method [9] by Pourreza and Golmohammadi [10]. A typical fabrication procedure was depicted as follows: they were first prepared separately in organic and aqueous phases. To prepare the organic phase, 125 mg of curcumin was dissolved in 25 mL of dichloromethane solvent. To prepare the aqueous phase, Triton X-100 (5% V/V), 10 mL of Triton X-100 5% (v/v) was added to 90 mL of boiling water. The aqueous phase was transferred to a 40 Hz ultrasonic bath for 15 minutes, then transfer to a 250 ml round bottom flask. Afterwards, 2 mL of organic phase was added dropwise to the aqueous phase (at a rate of approximately 10 drops per minute) under ultrasonic conditions for another 20 minutes. Following that, the contents were stirred at 1500 rpm by a magnetic stirrer at room temperature for 20 min until a yellow color was obtained. Eventually, this solution was placed in a rotary evaporator in order to ensure the complete removal of

dichloromethane. This solution was then stored in a brown bottle. Finally, the resulting clear yellow solution was kept in a dark bottle.

2.2. Preparation of dopamine solution (1000 mg mL⁻¹)

This solution was prepared by dissolving 0.1000 g of dopamine in water and diluting to 100 mL in a volumetric flask. All solutions with lower concentrations were obtained by diluting this stock solution.

2.3. Preparation of copper (II) solution

A stock solution of 1000 mg L⁻¹ of Cu(II) was prepared by dissolving 0.3929 g of CuSO₄·5H₂O (Merck) in distilled water and diluting to 100 mL in a volumetric flask. All solutions with lower concentrations were prepared by daily dilution of this solution.

2.4. Preparation of HCl solution (1 mol L⁻¹)

To prepare this solution, 8.3 mL of hydrochloric acid (37% purity and 1.19 g mL⁻¹ density) was transferred to a 100 mL volumetric flask was filled to the marked line with distilled water. All solutions with lower concentrations were obtained by diluting this solution.

2.5. Triton X-100 solution (5% V/V)

To prepare this solution, 5 mL of non-ionic surfactant Triton X-100 was transferred to a 100 mL volumetric flask and made up to the mark with distilled water.

2.6. Preparation of ammonia buffer

The ammonia buffer was prepared using HCl and NH₃. A 0.1 M solution of ammonia was prepared by transferring 0.4 mL of the NH₃ solution (d=0.91, 25%). This solution was poured into a beaker, and HCl (0.1 M) was gradually added to obtain a pH 8.5 buffer solution using a pH meter.

2.7. Recommended procedure

The following colourimetric analysis was performed by following these steps to measure dopamine. Different volumes of dopamine solution (100 mg L⁻¹), 2 mL of copper solution (II) solution (20 mg L⁻¹), and 1.5 mL ammonia buffer (pH = 8.5) were added to a 10 mL volumetric flask and made up to volume with distilled water. After 40 minutes, the absorbance of the solution was measured at 475 nm. The blank solution was prepared with the same method without adding any dopamine solution. Differences in the absorption intensity of curcumin NPs-copper in the presence and absence of dopamine were used as an analytical marker (ΔA) to measure dopamine.

3. Results and discussions

The UV-Vis absorption spectra of the curcumin NPs alone and in the presence of copper (II) and in the presence of both copper (II) and dopamine are illustrated in Figure 2 (A, B, C). There is a significant decrease in the absorbance of curcumin NPs alone and curcumin NPs-copper solutions. The interaction between curcumin and metal ions such as copper, iron, and other transition metals has been recognized and implied in many studies [11,12]. In previous studies, a 1:1 and 1:2 Cu(II)-curcumin complex has been synthesized and characterized [13]. Under the conditions in this procedure, only a 1:2 complex Cu(CUR)₂²⁺ is probably formed since 1:1 complex is formed in the presence of acetate as an auxiliary ligand [13]. However, when dopamine is present, it also forms a complex with copper ions, and curcumin is released, and the absorbance is increased again. This change in the absorbance of (ΔA) in the presence and absence of dopamine is used as an analytical signal to measure dopamine by the colorimetric method.

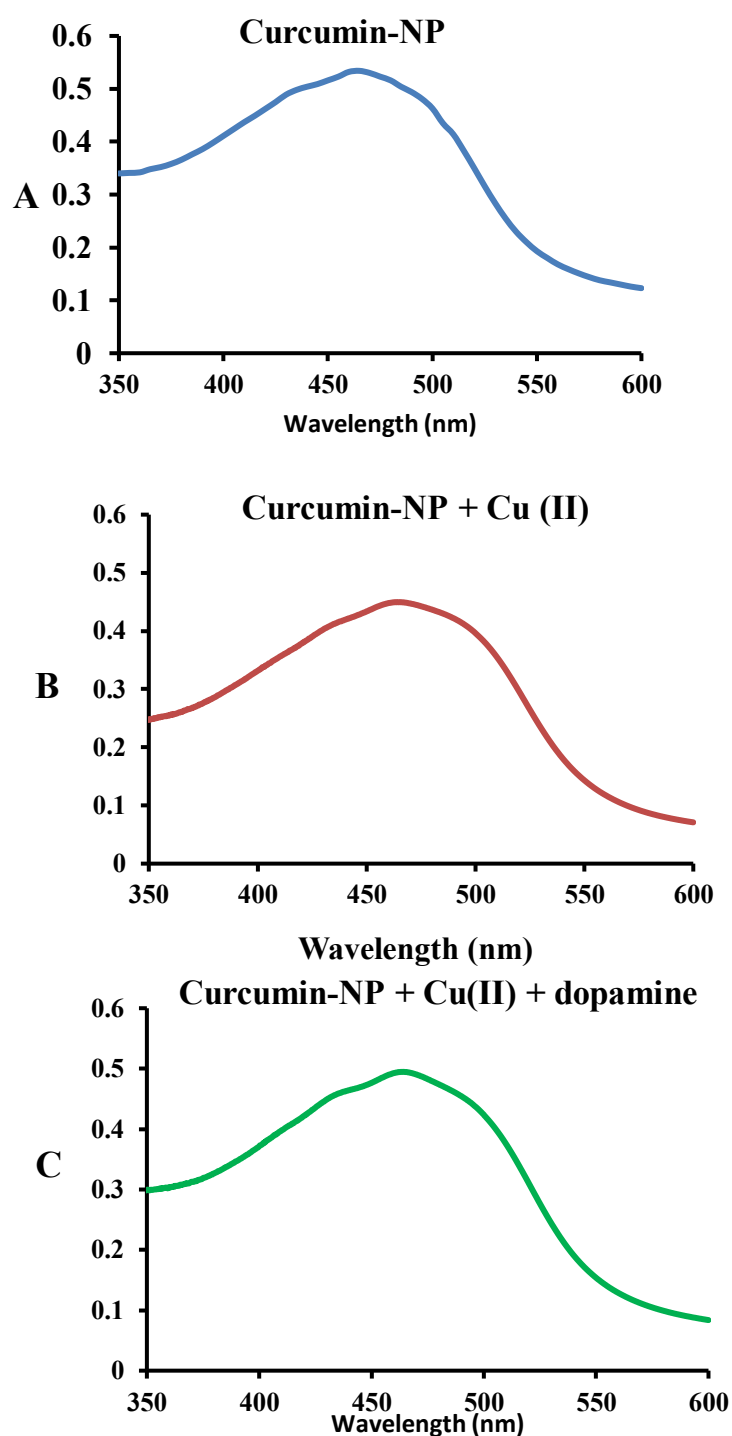


Figure 2: UV–Vis absorption spectra of the curcumin NPs alone(A) and in the presence of copper (II))B)and in the presence of both copper (II) and dopamine(C).

TEM images of curcumin NPs alone and in the presence of copper (II) and in the presence of both copper (II) and dopamine are presented in Fig.3 (A, B, C).

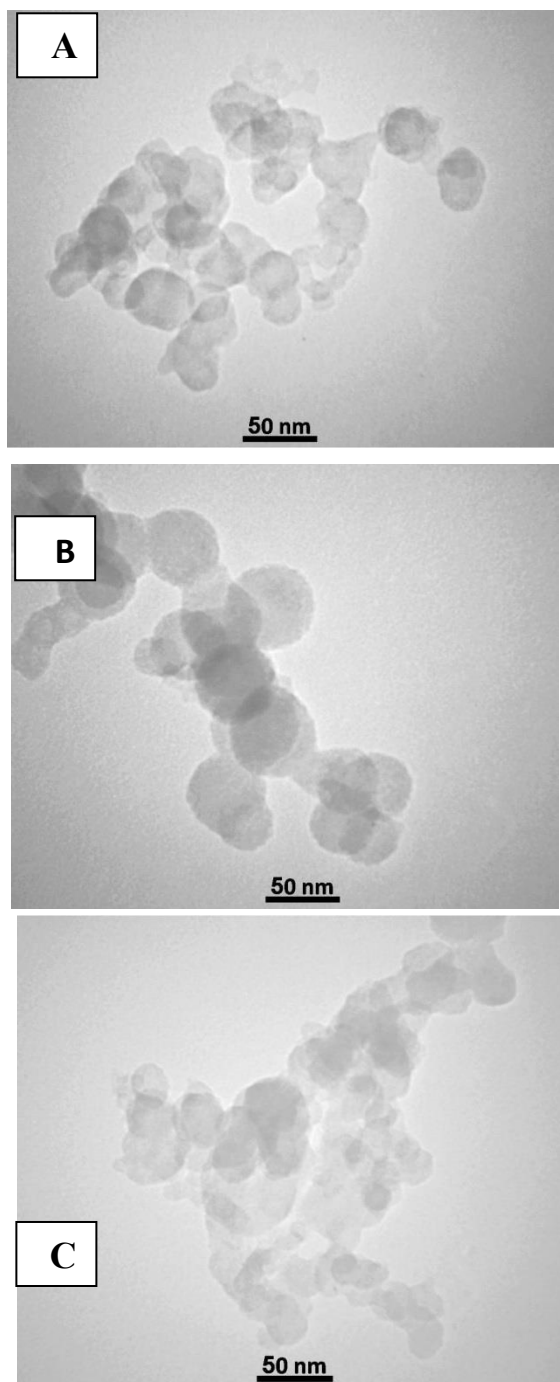


Fig.3. TEM images of (A) curcumin NPs, (B) in the presence of copper (II), and (C) in the presence of both copper (II) and dopamine

3.1. The investigation of parameters and optimization of variables affecting dopamine measurement

The effects of various parameters, including pH, copper(II) concentration, curcumin nanoparticle volume, and interaction time, were investigated on the dopamine measurement method. To achieve optimal conditions and obtain the maximum adsorption difference between the sample and blank solutions (ΔA), a one-variable-at-a-time method was used. At all stages of the optimisation, the above factors were evaluated at a constant dopamine concentration of 10 mg L⁻¹.

3.1.1. The effect of pH

The pH of the samples was adjusted according to the following method using the solutions of hydrochloric acid (0.1 mol L⁻¹) and sodium hydroxide (0.1 mol L⁻¹). 1 mL of dopamine solution (100 mg L⁻¹), 2 mL of copper (II) solution (20 mg L⁻¹), 1 mL of curcumin NPs, and varying volumes of hydrochloric acid and sodium hydroxide solutions were added to adjust the pH. The resulting solution was diluted to the mark in a 10 mL volumetric flask with distilled water. Initially, the volumetric flask was shaken to make the solution uniform. After 40 minutes, the absorbance of the solution was measured at 475 nm. The blank solution was prepared in the same way, but without dopamine. The results are shown in Table 1 and Figure 4. According to the results, the maximum value of ΔA is obtained when the pH reaches 8.5. At higher pHs, due to copper hydroxide deposition, the formation of the copper-dopamine complex is negligible, and as a result, the amount of ΔA is reduced. The ammonia buffer was selected to stabilize the pH.

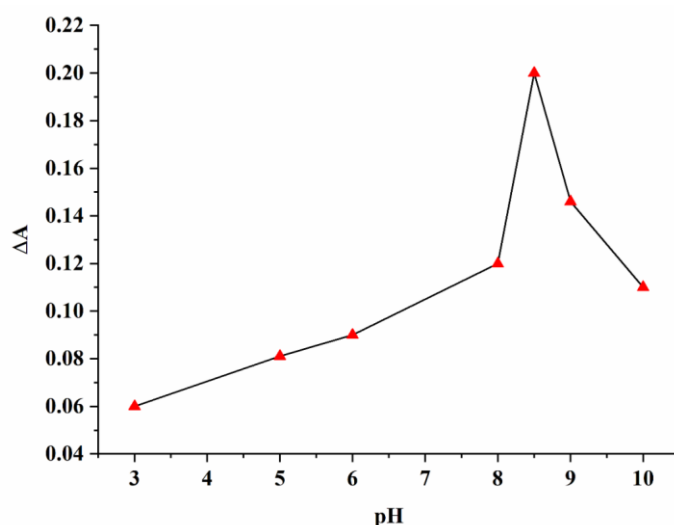


Fig. 4. Influence of pH on the determination of dopamine. Each number is the average of three determinations

Table 1. The results of pH on the determination of dopamine

pH	ΔA
3.0	0.060
5.0	0.081
6.0	0.090
8.0	0.120
8.5	0.200
9.0	0.146
10.0	0.110

3.1.2. The effect of ammonia buffer volume

To investigate the effect of buffer volume, different volumes of ammonia buffer were added separately to a series of 10 mL volumetric flasks. The flasks already contained 1 mL of dopamine (100 mg L⁻¹), 2 mL of copper solution (II) solution (20 mg L⁻¹), and 1 mL of curcumin NPs, which were made up to volume with distilled water. The flask was shaken to make a uniform solution. After 40 minutes, the absorbance of the prepared solutions was measured at the constant wavelength of 475 nm. The blank solution was prepared in the same way, but without dopamine. The results are presented in Table 2 and Fig. 5. The highest absorbance difference (ΔA) was observed with 1.5 ml of ammonia buffer. Therefore, 1.5 mL of ammonia buffer (pH=8.5) was selected as the optimal value for adjusting pH and obtaining the highest absorption difference.

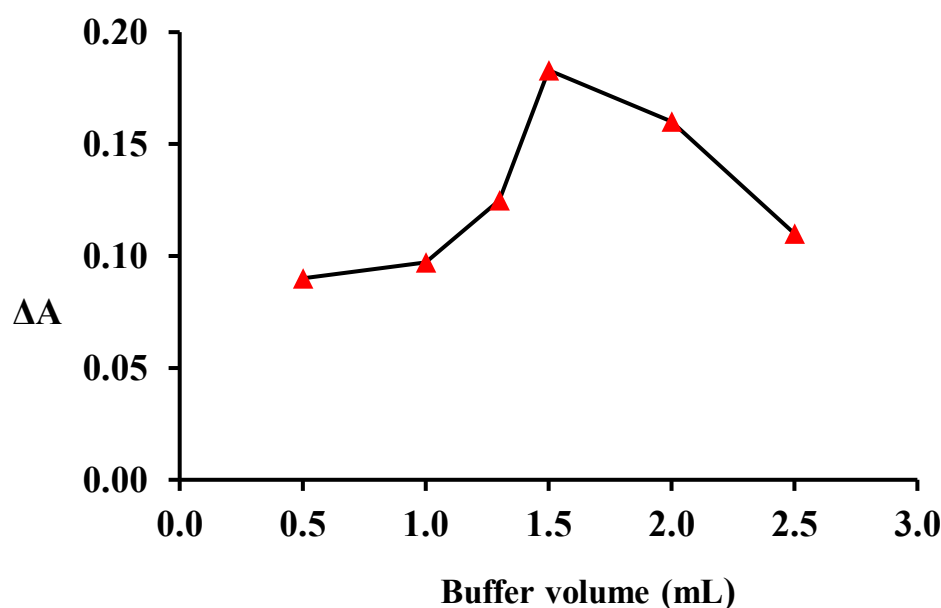


Figure5: Influence of ammonia buffer volume on the determination of dopamine

Table 2. The results of ammonia buffer volume on the determination of dopamine

V_{Buffer} (mL)	ΔA
0.5	0.090
1.0	0.097
1.3	0.125
1.5	0.183
2.0	0.160
2.5	0.110

3.1.3. The effect of copper (II) concentration

To investigate the effect of copper ion concentration on the dopamine measurement, the following process was performed: in a 10 mL volumetric flask, 1 mL of dopamine solution (100 mg L^{-1}), different volumes of copper (II) (20 mg L^{-1}), 1.5 mL of ammonia buffer ($\text{pH} = 8.5$) and 1 mL of curcumin nanoparticle solution was added. The resulting solution was made up to the mark with distilled water. The flask was shaken to make a uniform solution. After 40 minutes, the absorbance of the prepared solutions was measured at the constant wavelength of 475 nm. The blank solution was also prepared in the same way, but without adding any dopamine solution. The results (Table 3 and Figure 6) show that the concentration of copper (II) has a significant effect on the analytical signals (ΔA). The highest amount of ΔA was obtained when adding 2 mL of 20 mg L^{-1} of copper (II) solution which corresponds to 4 mg L^{-1} of copper (II) in the final volume of 10 mL, but the amount of ΔA decreases at higher concentrations. In this regard, 4 mg L^{-1} of copper (II) copper solution (II) was selected as the optimal value.

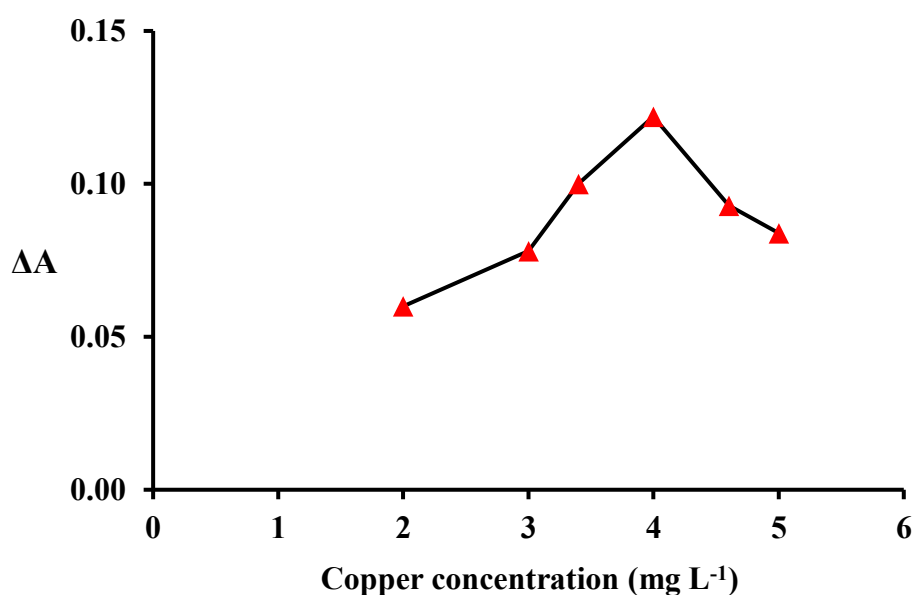


Figure 6: Influence of copper (II) concentration on the determination of dopamine

Table 3. The results of volumes of copper (II) concentration on the determination of dopamine

Copper (II) concentration (mg L^{-1})	ΔA
2.0	0.060
3.0	0.078
3.4	0.100
4.0	0.122
4.6	0.093
5.0	0.084

3.1.4. The effect of volume of curcumin NPs

To investigate the effect of curcumin concentration, the dopamine measurement process was performed according to the working method of section 2.5 and by changing the volume of curcumin NPs in the range of 0.5 mL to 1.5 mL. The blank solution was prepared in the same way, but in the absence of dopamine. According to the results in Table 4 and Figure, the obtained signals in volumes less than 0.5 mL of curcumin nanoparticle solution are negligible. In case of higher volumes (greater than 0.5 mL), the ΔA increases. On the other hand, due to the increase in initial absorption (A_0) for higher volumes (> 1 mL), the absorption decreases. Hence, the volume of 0.7 mL of curcumin NPs solution was chosen as the optimal volume in order to achieve high ΔA .

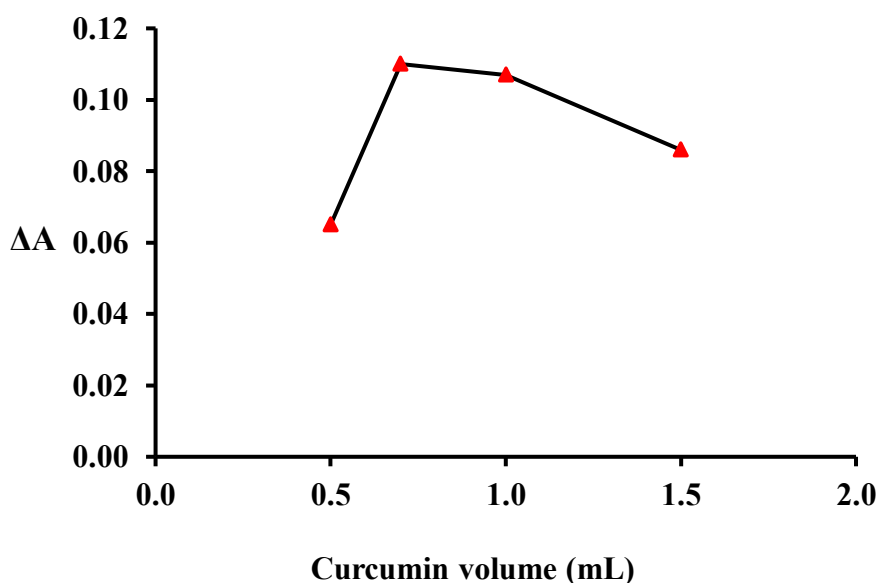


Figure 7: Influence of curcumin volume on the determination of dopamine

Table 4. The results of curcumin volume on determination of dopamine

V_{Curcumin} (mL)	ΔA
0.5	0.065
0.7	0.119
1.0	0.107
1.5	0.086

3.1.5. Influence of time

To investigate the effect of time on the dopamine measurement process, a time range of 10 to 60 minutes was examined. To a series of 10 mL volumetric flasks, 1 mL of dopamine solution (100 mg L⁻¹), 2 mL of copper solution (II) (20 mg L⁻¹), 1.5 mL of ammonia buffer (pH = 8.5), and 0.7 mL of curcumin NPs were added. The resulting solutions were made up to the mark with distilled water. The flask was shaken to make a uniform solution. Afterwards, the absorbance of the prepared solutions was measured at a constant wavelength of 475 nm at different times. The blank solution was prepared in the same way, but without dopamine. The results are shown in Fig.8 and Table 5. According to the results, the absorption of curcumin NPs in the presence of copper (II) decreased over time. This phenomenon is due to the formation of a curcumin nanoparticle-copper (II) complex. In the presence of

dopamine, the absorption value increases due to its inhibitory effect on the formation of the curcumin-copper nanoparticle complex. To find a suitable time, the absorption value was investigated. The absorption value remained approximately constant after 35 minutes, confirming the inhibitory effect of dopamine. As shown in Figure 2.7, the absorption difference remains almost constant beyond 35 minutes. Therefore, 40 minutes was chosen as standing time.

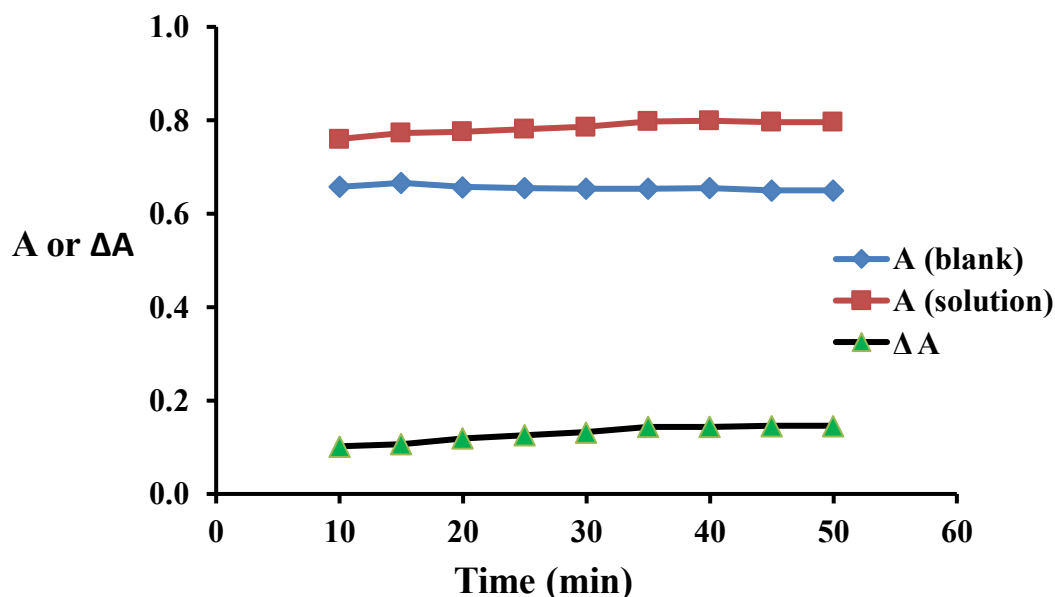


Figure 8: Influence of standing time on the determination of dopamine

Table 5. The results of standing time on the determination of dopamine

Time (min)	A _{blank}	A _{solution}	ΔA
10	0.658	0.76	0.102
15	0.666	0.773	0.107
20	0.657	0.776	0.119
25	0.655	0.781	0.126
30	0.654	0.786	0.132
35	0.654	0.798	0.144
40	0.655	0.799	0.144
45	0.650	0.796	0.146
50	0.650	0.796	0.146

3.2. Optimum condition

Optimal conditions based on experimental results obtained from different effective factors for dopamine measurement by spectrophotometry using curcumin NPs were obtained as follows (All optimal cases are in the final volume of 10 ml):

pH of the reaction medium: 8.5

Ammonia buffer volume: 1.5 mL

Concentration and volume of copper (II): 20 mg L⁻¹, 2 mL

Reaction time: 40 min

Volume of curcumin NPs: 0.7 mL

3.3. The calibration graphs

The calibration graph was plotted using the proposed method for measuring dopamine under optimal conditions. Based on the results obtained in Table 6 and Figure .9 the equation of the proposed method is linear in the concentration range of 0.5-20 mg L⁻¹ with a correlation coefficient R^2 of 0.9994, and the equation of the line is $\Delta A = 0.0062 C + 0.0743$. In this equation, ΔA is the difference in absorption intensity of curcumin-copper NPs in the presence and absence of analyte (dopamine), and C is the concentration of analyte in terms of mg L⁻¹.

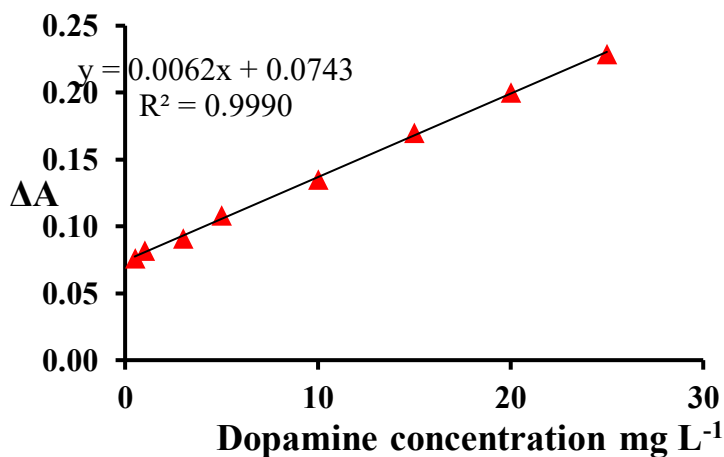


Figure 9: Calibration graph of different dopamine concentrations

Table 6. The results of different concentrations of dopamine

Dopamine concentration (mg L ⁻¹)	ΔA
0.5	0.076
1.0	0.082
3.0	0.091
5.0	0.108
10	0.135
15	0.170
20	0.200
25	0.229

Conclusion

The present study demonstrates a novel and efficient spectrophotometric method for the determination of dopamine using curcumin nanoparticles in the presence of copper (II) ions. The method is based on the enhancement of the absorbance of the curcumin-Cu-Cu(II) complex upon interaction with dopamine, providing a simple and effective means of detection. The reaction conditions were optimized to achieve high sensitivity and precision, with excellent linearity observed across a wide concentration range (0.5–20 mg·L⁻¹) and a strong correlation coefficient ($r = 0.9994$).

This approach offers several advantages, including low cost, simplicity, and the use of environmentally friendly materials. Moreover, the method does not require sophisticated instrumentation or complex procedures, making it suitable for routine analysis in clinical laboratories, pharmaceutical quality control, and research applications.

The successful application of curcumin nanoparticles highlights their potential as a green nanomaterial in analytical chemistry. Future studies may focus on applying this method to real biological samples or further enhancing sensitivity through nanoparticle surface modifications.

Conflicts of Interest

The authors declare no conflicts of interest.

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