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Spectrophotometric Estimation of Donepezil Hydrochloride in pure and dosage forms via ion-pair complex with Erythrosine Dye

Hend M. Mostafa[a], Arwa A. Abdo[a]*, Tarek Wahdan[a], Zeinab M. Anwar[b]*

[a]*Chemistry Department, Faculty of Science, Arish University, Al-Arish 45511, Egypt.*
[b]*Chemistry Department, Faculty of Science, Suez Canal University.*

Abstract

In this study, the goal is to improve and verify a simple, sensitive, accurate spectrophotometric technique for the measurement of drug formulations and pure forms of donepezil HCl (DNP). The procedure is forming an ion-pair complex in an acetate buffer solution between donepezil and the dye erythrosine B. The resulting complex is then measured spectrophotometrically at a wavelength of maximum absorbance (λ max) of 527nm. Beer's law was found to be applicable in the concentration range of 0.2-1.2 mg/ml, with a high correlation coefficient (n=5) of 0.99813. This indicates a strong linear relationship between the concentration of Donepezil HCl and the absorbance values within the specified range. The limit of detection (LOD) for the drug was determined to be 0.95056 mg/ml, representing the lowest concentration at which the method can reliably detect the presence of Donepezil HCl. Additionally, the limit of quantification (LOQ) was established at 3.1685 mg/ml, indicating the lowest concentration at which accurate quantification can be achieved. The proposed method was subjected to validation for accuracy and precision, demonstrating its reliability and reproducibility. The developed spectrophotometric method has been successfully applied to the analysis of Donepezil in its pure form as well as in pharmaceutical dosage form

Keywords: Donepezil, Erythrosine B dye; Spectrophotometry; ion pair; pharmaceutical forms.

1. Introduction

Donepezil Donepezil is a medication used in the treatment of dementia in individuals with Alzheimer's disease [1], a progressive neurological disorder that is characterized by a gradual decrease in memory, cognitive function, communication abilities, and daily life skills [2]. As a cholinesterase inhibitor, donepezil works by increasing levels of acetylcholine in the brain, temporarily aiding in the improvement or stabilization of cognitive symptoms associated with Alzheimer's disease [3,4].

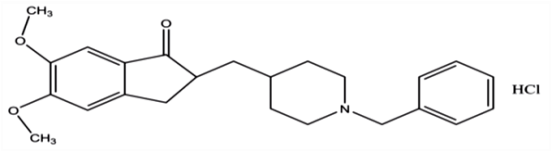

Figure 1: Structure of donepezil hydrochloride.

Donepezil Donepezil belongs to the class of medications known as cholinesterase inhibitors [5], acting to enhance mental function, including memory, attention, communication skills, and the ability to perform daily activities. Donepezil is commonly prescribed for individuals with Alzheimer's disease to potentially improve cognitive abilities or slow the progression of cognitive decline [6]. In terms of analytical methods for its estimation, various chromatographic techniques, such as HPLC [7], HPTLC [8], and LC-MS [9], have been employed.

However, it's worth noting that these methods can be time-consuming and expensive, reflecting the complexity and precision required for the analysis of this medication. Also, spectrophotometric methods need time and expensive chemicals [10,11,12]. Few spectrophotometric methods have been used for the estimation of Donepezil in dosage forms [13-14]. So, the aim of the present work is to improve a simple, economical, rapid, and accurate ion pair spectrophotometric method for estimating Donepezil

*Corresponding author e-mail: arwa.abdo@sci.aru.edu.eg (Arwa A. Abdo)
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HCl in its dosage forms.

2. Experimental

2.1Materials and methods

2.1.1.Instruments

T90+ UV-visible double beam spectrophotometer is in use for spectral studies. pH measuring is made with (AD12) digital pH meter.

3.1.2.Materials

The pure drug Donepezil HCl (M.wt =415.96 g/mol), Erythrosine B dye (M.wt = 879.86 g/mol) are used, glacial acetic acid, sodium acetate, potassium dihydrogen phosphate, sodium hydroxide, and phosphoric acid are used for the preparation of buffer solutions. All used chemicals were purchased from Sigma-Aldrich company. All chemicals were used as supplied without any further purification.

2.1.3Pharmaceutical formulations

Donazil (5mg)(Eva Pharma), ledmetzil (5mg)(B.P.Pharma) and Donhimer (5mg) (Multi Apex Pharma) (MAP)tablets.

Standard and Buffer solutions

Stock standard solution of DNP drug: donepezil (10 mg) was dissolved in l00 ml double-distilled water.

Stock standard solution of Erythrosine B (1×10^{-4} M) accurately weighed quantity of dye 8.8 mg was dissolved in double-distilled water and completed to 100 ml in a volumetric flask.

Acetate buffer (pH=4.5): a weighed 13.5 mg of sodium acetate was dissolved in 150 ml of double-distilled water then 7 ml glacial acetic acid was added and the volume was made up to 300 ml with double-distilled water.

Phosphate buffer (pH=3): a weighed 3.4 g of potassium dihydrogen phosphate was dissolved in double-distilled water 900 ml, adjusting the pH 3.0 with phosphoric acid, and diluted to 1000.0 ml with double-distilled water.

Phosphate buffer (pH=7.5): add 250 ml of 0.2 M potassium dihydrogen phosphate to 393.5 ml of 0.1M sodium hydroxide, then dilute to 1000 ml with double-distilled water.

2.2. General procedures

Construction of Construction of calibration curves. Aliquots of (0.2-1.2 ml) of the standard DNP solutions (0.1 mg /ml) were transferred to 10 ml measuring volumetric flask, 2 ml of buffer solution with pH4.5 was added, then 2.0 ml of 1×10-4 M dye solution was added, then the total volume of each solution was completed to 10 ml by double-distilled water. The absorbance measurements were graphed versus DNP concentrations to generate a standard calibration curve. The experimental procedures were meticulously designed and optimized to ensure the complete formation of an ion-pair complex with increased sensitivity and maximal absorbance. Rigorous adjustments were made, including variations in the pH of the buffer, volume of the dye, and buffer volume, all systematically studied to pinpoint optimal conditions. Preliminary experiments were conducted to fine-tune the reaction parameters, ultimately aiming for the most efficient and reliable process. The central focus of the experiment was on achieving maximal absorbance, indicative of enhanced detection capabilities, while the systematic study of parameters aimed to establish conditions favourable for the formation of the ion-pair complex. Every measurement was taken at room temperature

2.2.1Applications to Pharmaceutical formulation.

Nine tablets were crushed, finely powdered, weighed out and the average weight of one tablet was determined for each drug to assure that there is no weight loss. An accurate weight equivalent to 5.0 mg DNP (the concentration of DNP per one tablet) was dissolved in double-distilled water and the volume was completed to 10 ml. The solution was then filtered through Whatman filter paper then the filtrate was transferred to a 10 ml volumetric flask. 2 ml of 1×10^{-4} M erythrosine b dye was added and 2 ml of acetate buffer solution pH 4.5 was added and the volume of the solution was completed to 10 ml with double-distilled water. The obtained colored solution was measured at 527 nm against a blank solution of the reagent.

3. Results and discussion

The UV-Vis absorbance spectra of the ion-pair complexes, that were formed between DNP and Erythrosine b dye were recorded at the range 450-600 nm. It was found that the ion pair complex has a maximum absorption at λ max 527nm as shown in Figure 2. The effect of adding different concentrations of the drug to the dye is depicted in Figure 3. The subsequent addition of the drug is accompanied by a decrease in the absorbance of the dye.

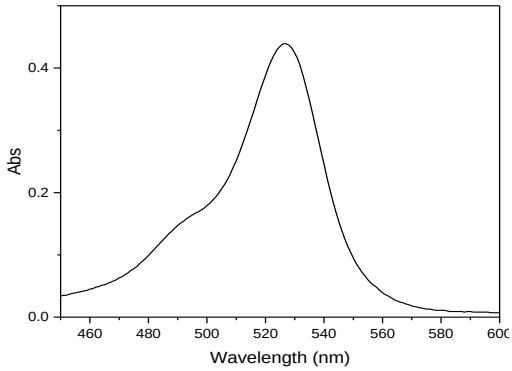


Figure 2: UV-Vis spectra of the ion-pair complex of (6.0 µg/ml) of DNP and 1×10^{-4} M EB dye

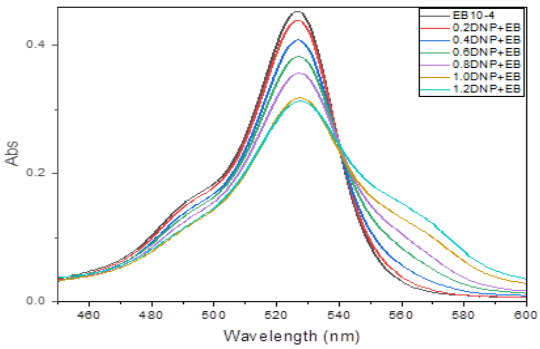
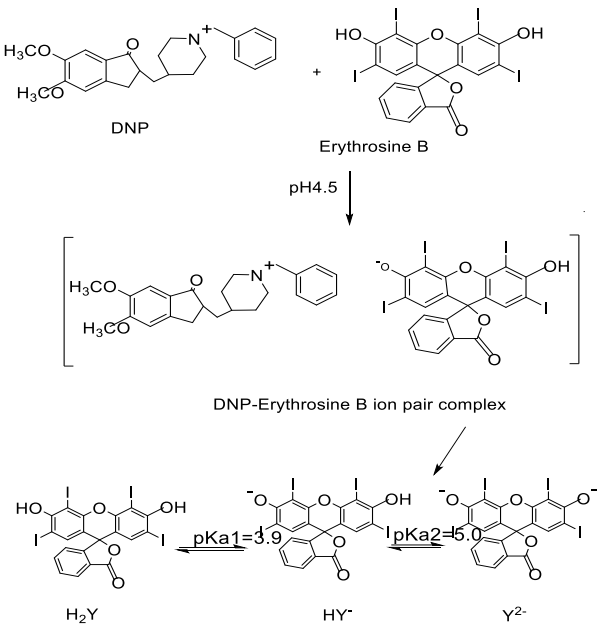


Figure 3: Absorption spectra of donepezil at different concentrations with EB dye in acetate buffer (pH 4.5)

Scheme 1, shows the ion pair mechanism which is proposed to be formed between the drug and the dye at pH 4.5, where the dye is in its monoanionic form HY^-.



Scheme 1: The proposed reaction pathway between Donepezil and Erythrosine B at pH 4.5 indicates the dissociation of EB at different pH values

3.1. Optimum conditions for complex formation.

The procedures were rigorously optimized to achieve complete reaction formation, increased sensitivity, and maximal absorbance. Preliminary experiments were conducted to determine the reaction conditions of the ion-pair complex. The parameters, including the pH of the buffer, volume of the dye, and volume of the buffer, were systematically studied to ensure optimal conditions for the formation of the ion-pair complex.

3.1.1. Effect of buffer pH and species on ion-pair Formation

By extracting the colored formed complexes in the presence of different buffers, the impact of pH on the drug-reagent complex formation was investigated. The results of the investigation showed that, in acetate buffer, pH 4.5 produced the greatest absorbance values and the maximum color intensity, as shown in Figure 4. This consistent observation was maintained across all concentrations of Donepezil drug, as illustrated in the corresponding Figure 5. The pH sensitivity of the complex formation suggests the importance of maintaining a specific pH condition, particularly at pH 4.5, for optimal color development and absorbance in the analytical method.

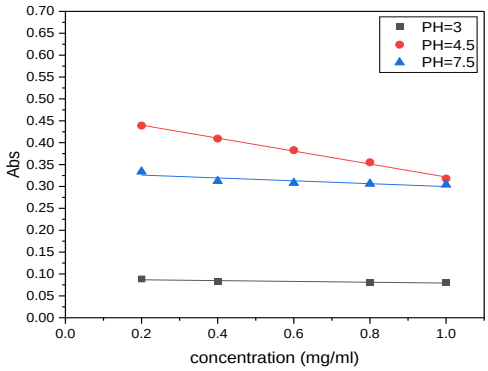


Figure 4: Effect of pH on the absorbance of the ion-pair complex at different concentrations of Donepezil drug.

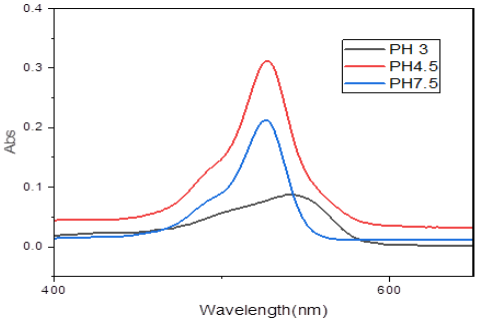


Figure 5: Effect of pH on the absorbance of ion-pair complex Donepezil drug.

3.1.2. Effect of buffer volume on ion-pair Formation

By evaluating the absorbance of the colored complexes in the presence of varied quantities of buffer solutions, the impact of buffer volume on the drug reagent complex formation was investigated. It was noticed that the maximum color intensity was observed in the presence of 2 ml of acetate buffer pH 4.5 as shown in Figure 6,7.

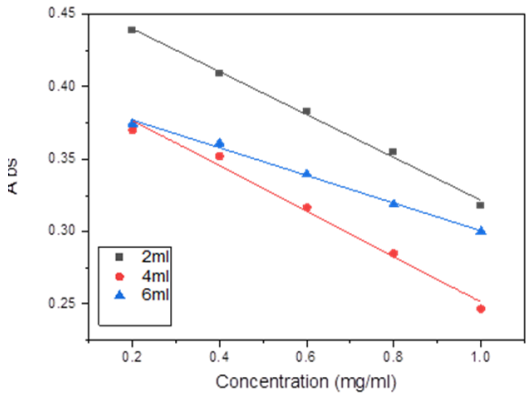


Figure 6: Effect of buffer volume on the absorbance of ion-pair complex at different concentrations of Donepezil drug

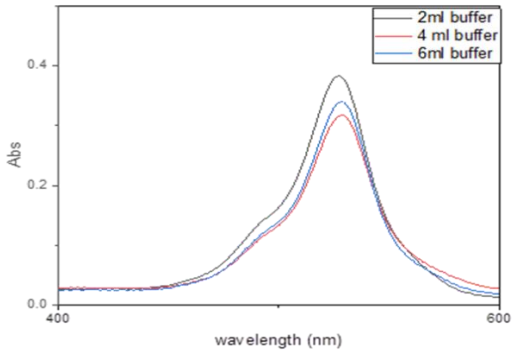


Figure 7: Effect of buffer volume on the absorbance of ion-pair complex Donepezil drug

3.1.3. Effect of dye concentration.

By measuring the absorbance of solutions including a fixed concentration of DNP and the corresponding dye (1×10^{-4} M) (1,2,3 ml), the impact of the reagent was examined. The maximum color intensity of the complex was observed with 2 ml of dye (1×10^{-4} M) solution figure (9). Figure (8) illustrates similar results for all the studied concentrations of Donepezil HCl drug.

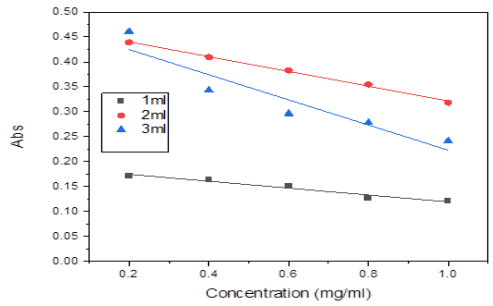


Figure 8: Effect of volume of Erythrosine dye on the absorbance of ion-pair complex at different concentrations of Donepezil drug.

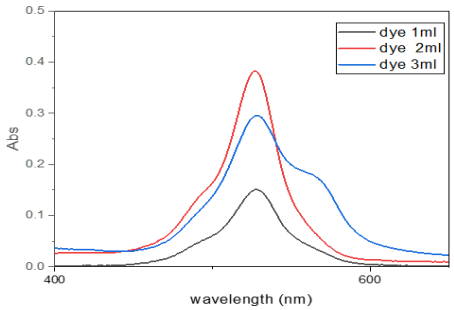


Figure 9: Effect of volume of Erythrosine B dye on the absorbance of ion-pair complex Donepezil drug

3.1.4. The impact of reagent volume on the drug-dye reaction

Job's approach [15] of continuously varying equimolar solutions (1×10-4M) was utilized to ascertain the molar ratio between the donepezil and the dye Erythrosine B. A series of solutions with a constant total volume of 10 ml, containing both the drug and Erythrosine B dye, was prepared. The absorbance of these measured solutions at the optimal wavelength, revealing a molar ratio of 1:1 (drug: dye) in the ion pair complexes. This outcome suggests the formation of ion-pair through electrostatic attraction, wherein positively charged DNP ions interact with Erythrosine B dye negatively charged, confirming a 1:1 molar ratio in the complex formation.

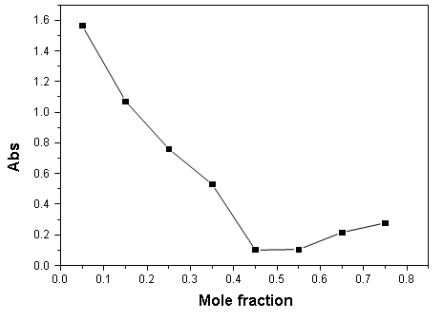


Figure 10 (a)

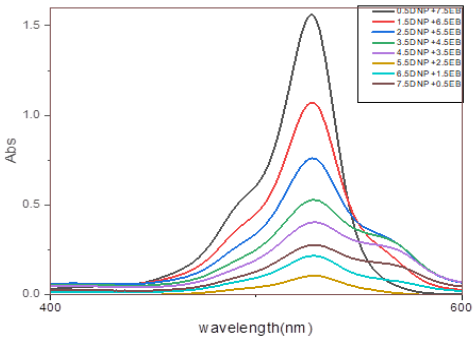


Figure 10 (b)

Figure 10 a,b: Job's approach of continuous variation plots for the reaction of DNP with EB dye

3.2. Method validation

3.2.1. Linearity

Using reagents, standard calibration curves were created by graphing absorbance versus the concentration of DNP in the experimental conditions for determining (DNP). The equation A=aC+b, where A is the absorbance and C is the concentration in µg/ml, was followed by the calibration curves.

The linearity of the proposed method was verified by analyzing six solutions covering the range of the used concentrations is 0.2 to 1.2 mg/ml as shown in figure 3. The calibration curves, constructed by plotting the concentration of the analyte versus absorbance, exhibited good linearity over the specified range. The validation of linearity was confirmed by a high correlation coefficient value (0.99813) and a small Y-intercept of the regression equations. This confirmed a robust linear correlation between the drug's concentration and absorbance. Statistical data analysis was performed by calculating parameters such as linearity range, correlation coefficient, slope, as well as optical and analytical characteristics, including maximum absorption, Beer's law limit, molar absorptivity, and residual standard deviation, presented in Table (1). Figure 11: illustrates the calibration plot for the different concentrations of the donepezil drug interaction with the dye at pH 4.5, where the validation parameters are collected in Table 1.

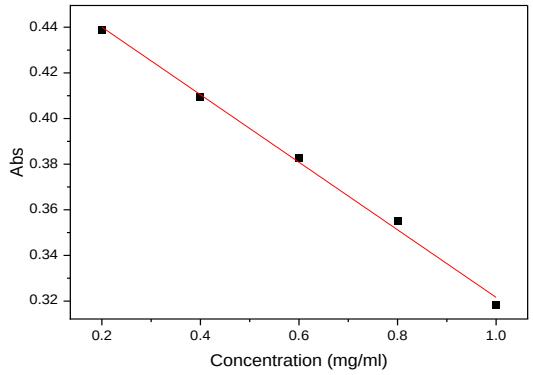


Figure 11: Calibration curve of donepezil in acetate buffer (pH4.5).

Table 1: Optical characterization and analytical data in the determination of Donepezil using Erythrosine B dye.

Parameters	Value
Wavelength λ max(nm)	527
Bear's law limit (mg\ml)	0.2-1.2
Molar absorptivity Ɛ (l\mol-1 cm-1)	6.1583×104
Regression equation	
Intercept (a)	0.46967
Slope (b)	0.14805
Correlation coefficient (r)	0.99813
LOD (mg\ml)	0.95056
LOQ (mg\ml)	3.1685
pH	4.5
Dye concentration (M)	10^{-4}
Volume of dye (ml)	2

A=a+bC where C is the concentration in µg /ml, A is the absorbance; LOD limit of detection; LOQ limit of quantification.

4.2.2. Sensitivity

The following formulas were used to determine the suggested method's limits of detection (LOD) and quantitation (LOQ) [16].

LOD = 3s/k LOQ=10s/k

The slope of the calibration graph is the sensitivity, denoted by k, and s is the standard deviation of the response of the blank or the standard deviation of the regression line intercepts.

3.3. Analysis of pharmaceutical preparation.

The suggested method was applied successfully to determine Donepezil hydrochloride in dosage forms (Donazil, Lidemtzil and Donhimer) Six replicate determinations were made to assess the precision of the used method, and the results showed good agreement with label claims, indicating the accuracy. Data is given in table (2).

Table 2: Recovery of estimation of Donepezil drug in different dosage forms

Sample	Added (µg /ml)	Found (µg/ml)	Recovery %
Donazil (5 mg)	6.0	5.9556	99.26
Lidemtzil (5mg)	2.0	2.082	104.1
Donhimer (5mg)	1.2	1.0876	90.64

4. Conclusion

The paper outlines a novel method for quantifying DNP in pure dosage forms using extracting ion–pair complex formation reaction with acid dyes. The proposed methods are characterized as simple, rapid, and cost-effective, offering increased sensitivity for DNP determination compared to existing spectrophotometric methods. The limit of detection and quantification (LOD) and (LOQ) were to be 0.95056, and 3.1685 mg/ml respectively which represent the lowest concentration at which the method can reliably detect the presence of Donepezil HCl as well the accurate quantification can be achieved. Statistical parameters and recovery data support the methods' accuracy, sensitivity, and precision.

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