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Kinetic spectrophotometric determination of nizatidine and ranitidine in pharmaceutical preparations

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Abstract

A new simple and sensitive kinetic spectrophotometric method is described for analysis of nizatidine (**I**) and ranitidine (**II**). The method involves the reaction of the drugs with alkaline potassium permanganate, whereby a green color peaking at 610 nm is produced. The reaction is monitored spectrophotometrically by measuring the rate of change of absorbance of the resulting manganate species at 610 nm. Calibration graphs are linear over the concentration range 0.8–4.0 μg/ml and the precision (% RSD 1.80, 1.53 for **I** and **II**, respectively) is quite acceptable. The method is satisfactorily applied for direct analysis of pharmaceutical preparations containing **I** and **II**. A proposal of the reaction pathway is postulated.

Keywords: Nizatidine; Ranitidine; Kinetic spectrophotometric method

1. Introduction

Nizatidine (**I**) and ranitidine (**II**) are specific H_2-receptor antagonists. They are more potent than cimetidine in inhibition of gastric acid secretion induced by various stimuli and they lack cimetidine's anti-androgenic and hepatic microsomal inhibiting effects [1].

Nizatidine (**I**) has been determined in pharmaceutical preparations using spectrophotometry [2,3], potentiometric titration [4], coulometry [5], HPLC [6,7] and polarographic [8] methods. In biological fluids, it was determined using HPLC [9–12] and polarographic methods [13]. The analytical methods reported for the determination of ranitidine in pharmaceutical preparations included spectrophotometry [14,15], NIR spectroscopy [16,17] quantitative NMR [18] X-ray using neutral network [19] polarography [20,21], radioimmunoassay [22] and flow injection [23,24] methods, Capillary electrophoresis [25], HPTLC [26,27] and HPLC [28] methods were also reported for determination of ranitidine in pharmaceuticals or in biological samples [29,30]. The protolytic constants of nizatidine and ranitidine were studied using a spectrophotometric method [31]. An extended bibliography of nizatidine can be found in the comprehensive analytical profile [32].

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Kinetic methods are becoming of great interest in chemical analysis. Various kinetic methods have been applied to the determination of many inorganic and organic species [33].

This work represents the first attempt at assaying nizatidine (**I**) and ranitidine (**II**) in pharmaceutical preparations by use of kinetic methods. The method is based on oxidizing the drugs with alkaline potassium permanganate [34]. The reaction is followed up spectrophotometrically and the rate of change of absorbance at 610 nm is measured. The fixed time method is adopted after full investigation and understanding of the kinetics of the reaction. The proposed method is simple, accurate and sensitive. In addition, it is not susceptible to interference from common tablet excipients.

2. Experimental

2.1. Materials and reagents

Nizatidine was kindly provided by Eli Lilly (Indianapolis, IN). Ranitidine HCl was obtained from Glaxo–Wellcome and used as received. Potassium permanganate, Riedel-de Häen, 1.2% w/v aqueous solution. Sodium hydroxide, BDH, UK, 0.4 M aqueous solution.

2.1.1. Standard drug solutions

Stock solutions of **I** and **II** containing 0.1 mg/ml were prepared in water. The solutions were stable for at least 3 days when kept in the refrigerator (at about 4 °C).

2.2. Apparatus

UV-Visible Spectrophotometer, Unicam He λ 10 S α, IPC with 1 cm quartz cells.

2.3. General recommended procedure

2.3.1. For nizatidine

Transfer 3 ml of 1.2% w/v potassium permanganate and 2 ml of 0.4 M sodium hydroxide solution, into 25 ml volumetric flasks. Add 0.2–1.0 ml aliquots of stock solution of (**I**) containing suitable amounts of the drug (Table 1). Complete to volume with water and mix well. Immerse the flasks in a thermostated water bath at 65 °C for a fixed time of 20 min. Cool the flasks on a water bath maintained at 20 °C. Measure the absorbance of solutions at 610 nm against an appropriate blank. Construct the calibration graph by plotting the final concentration of the drug against the absorbance values, measured at a fixed time of 20 min. Alternatively, derive the corresponding regression equation.

Table 1
Analytical data for the kinetic determination of nizatidine (**I**) and ranitidine (**II**)

Parameter	Nizatidine	Ranitidine
Volume of $KMnO_4$ (ml)	3	2
Temperature (°C)	65	25
Reaction time (min.)	20	8
Concentration range [M]	2.4136×10^{-6}–1.2068×10^{-5}	2.544×10^{-6}–1.272×10^{-5}
Molar absorptivity (ε)	40 976	19772
Regression equation	$A=-0.0191+40\,976.13\ C$	$A=0.0189+19\,772.013\ C$
Correlation coefficient (r)	0.9995	0.9998
$S_{y/x}$	$5.748\ E^{-3}$	$1.866\ E^{-3}$
S_a	$5.748\ E^{-3}$	$1.866\ E^{-3}$
S_b	$2.27\ E^{-3}$	$7.37\ E^{-3}$
RSD%	0.76	0.96
SAE	0.34	0.43

2.3.2. For ranitidine

Transfer 2 ml of 1.2% w/v potassium permanganate and 2 ml 0.4 M sodium hydroxide solutions into 25 ml volumetric flasks. Add 0.2–1.0 ml aliquots of stock solution of (**II**) containing suitable amounts of the drug (Table 1). Complete to volume with water and mix well. Immerse the flasks in a thermostated water bath maintained at 25 °C for a fixed time of 8 min. Measure the absorbance of solutions at 610 nm against an appropriate blank. Construct the calibration graph by plotting the final concentration of the drug against the absorbance values measured at a fixed time of 8 min. Alternatively, derive the corresponding regression equation.

2.3.3. Procedure for pharmaceutical preparations

Transfer an accurate weight of the mixed contents of 10 capsules of (**I**) or the finely powdered and mixed 20 tablets of (**II**) equivalent to 10 mg of **I** or **II** into a small flask, sonicate for 10 min with 50 ml of chloroform for (**I**) or acetone for (**II**). Filter, wash the residue and the flask with chloroform for (**I**) or acetone for (**II**) and pass the washings to a round bottom flask. Evaporate the organic solvent to dryness under vacuum. Dissolve the residue in water by sonication for 10 min, transfer quantitatively the resulting solution into 100 ml volumetric flask and dilute to volume with water. Transfer aliquots of the resulting solution to 25 ml volumetric flasks, containing the appropriate volume of 1.2% w/v $KMnO_4$, 2 ml of 0.4 M sodium hydroxide solution and proceed as under general recommended procedure for nizatidine (**I**) or ranitidine (**II**).

3. Discussion

3.1. Kinetics and optimization of the reaction conditions

Nizatidine (**I**) and ranitidine (**II**) were found to react with alkaline potassium permanganate producing a green color peaking at 610 nm (Fig. 1). At this wavelength, the various experimental parameters affecting the development and stability of the reaction product were optimized by changing each variable in turn, while keeping all others constant. The effect of potassium permanganate concentration on the reaction was studied over the range 1.52×10^{-3}–15.2×10^{-3} M. The maximum absorbance was obtained at concentrations of 9.12×10^{-3} M and 6.08×10^{-3} M for (**I**) and (**II**), respectively. Higher concentrations of potassium permanganate yielded lower absorbance values, probably due to decomposition of the product (Fig. 2). Complete reaction be-

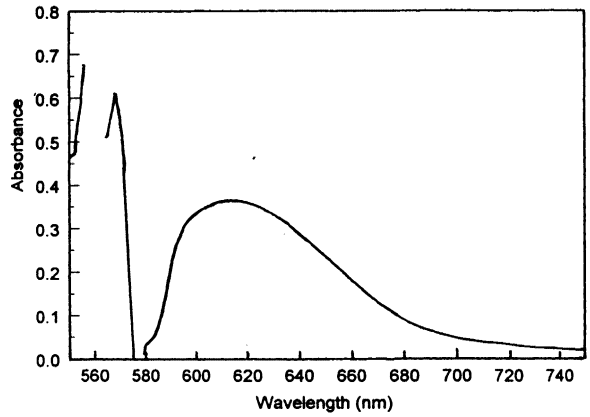


Fig. 1. Absorption spectrum of the reaction product of nizatidine (9.65×10^{-6} M) with alkaline potassium permanganate.

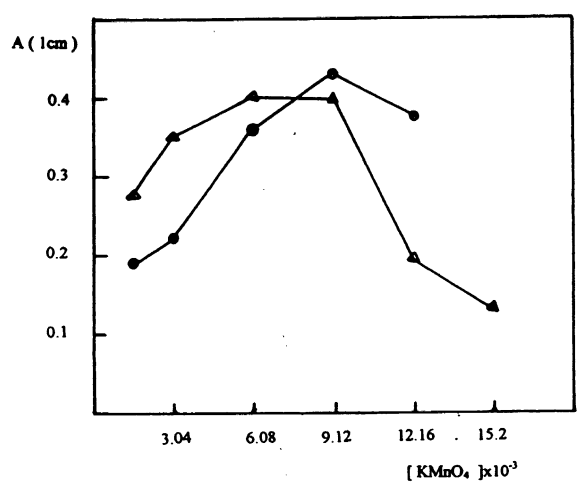


Fig. 2. Effect of potassium permanganate concentration on the reaction product of 1.21×10^{-5} M nizatidine (●—●) or 1.27×10^{-5} M ranitidine (▲—▲) measured at room temperature after 20 min.

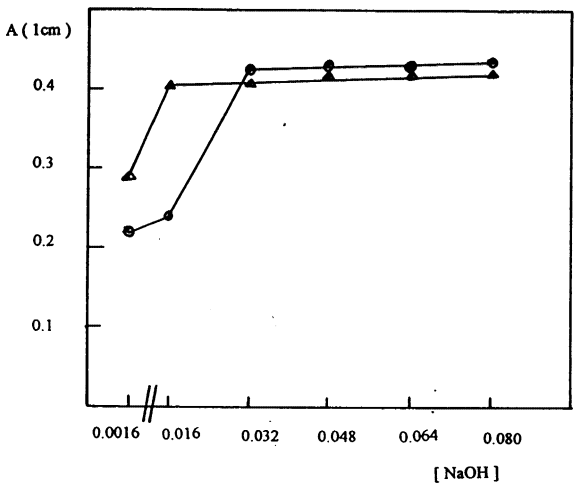
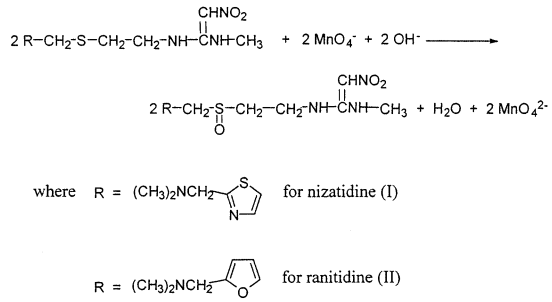


Fig. 3. Effect of sodium hydroxide concentration on the reaction product of 1.21×10^{-5} M nizatidine (●—●) or 1.27×10^{-5} M ranitidine (▲—▲) measured at room temperature after 20 min.

tween (**I**) or (**II**) and potassium permanganate takes place only in alkaline medium. The influence of the medium alkalinity was investigated between 1.6×10^{-3} M–0.08 M sodium hydroxide. It was observed that the reaction is zero order with respect to hydroxyl ion concentration between 0.016 and 0.08 M sodium hydroxide (Fig. 3). Therefore, 0.032 M sodium hydroxide in the final concentration (2 ml of 0.4 M sodium hydroxide) was chosen for all subsequent experiments. The effect of temperature was studied in the range of 25–80 °C. The rate of reaction of (**I**) with potassium permanganate increased with increasing temperature up to 65 °C; at higher temperatures, lower absorbance values were obtained. Therefore, 65 °C was selected as the optimum temperature (Fig. 4). Meanwhile, upon heating (**II**) with potassium permanganate the absorbance values increased between 25 and 60 °C. At higher temperatures, lower absorbance values were obtained (Fig. 4). However, 25 °C was selected as the optimum temperature due to the low reproducibility of absorbance values obtained at higher temperatures. The reaction stoichiometry was studied adopting the molar ratio method [35] and was found to be 1:1. Based on the presence of the thioether linkage and its liability to oxidation to the corresponding sulphoxide, the following pathway is proposed as the reaction mechanism:



Based on the obtained values of slopes of the absorbance versus time curves for solutions containing different amounts of nizatidine (**I**) or ranitidine (**II**) (Figs. 5 and 6), the reaction is pseudo-first order with respect to nizatidine (**I**) or ranitidine (**II**). Under the described experimental conditions, the reagent showed pseudo-zero order dependence, so, the following kinetic equation is proposed:

$$V = k[C]$$

where V is the rate of the oxidation reaction and k is the conditional rate constant and $[C]$ is the molar concentration of nizatidine (**I**) or rani-

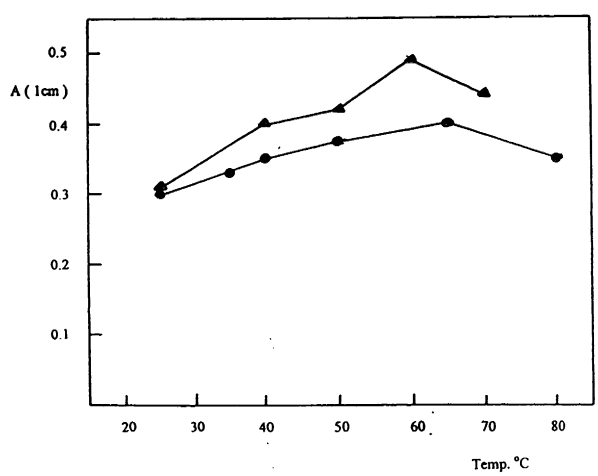


Fig. 4. Effect of temperature on the reaction between 1.21×10^{-5} M nizatidine (●—●) or 1.27×10^{-5} M ranitidine (▲—▲) with alkaline potassium permanganate.

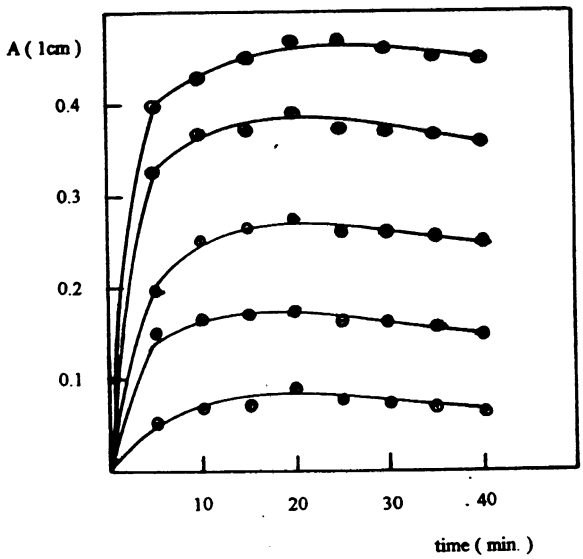


Fig. 5. Absorbance versus time graphs for the reaction between nizatidine and potassium permanganate at 65 °C showing the dependence of the reaction on nizatidine concentration. Nizatidine concentration 2.41×10^{-6}–1.21×10^{-5} M.

tidine (**II**). Taking logarithms of rates and concentrations, the above equation becomes:

$$\text{Log } V = \log \frac{\Delta A}{\Delta t} = \log k = n \log [C]$$

where A is the absorbance and t is the time in seconds. Regression of log (V) versus log [C] gave the regression equations:

$$\log (V) = 1.55 + 1.009 \log C,$$

$r = 0.9990$ for nizatidine (**I**).

$$\log (V) = 0.9266 + 0.8545 \log C,$$

$r = 0.9990$ for ranitidine (**II**).

Hence $K = 35.48\ s^{-1}$ for nizatidine (**I**) and 8.45 s^{-1} for ranitidine (**II**) and the reaction is pseudo-first order ($n \approx 1$) with respect to nizatidine (**I**) or ranitidine (**II**).

3.2. Linearity

The kinetic curves obtained at different concentrations of nizatidine (**I**) or ranitidine (**II**), under the optimized conditions, were processed by the fixed-time method [36]. Calibration graphs of absorbance versus initial concentrations of (**I**) or (**II**) were established at different fixed-time intervals. It was found that the slopes increase with time and the most acceptable values of the correlation coefficient (r) and the intercept were obtained at a fixed time of 20 min for (**I**) and 8 min for (**II**) which were, therefore, chosen as the most suitable time intervals for measurement. The calibration graphs were linear over the concentration range of 2.4136×10^{-6}–1.2068×10^{-5} M (0.8–4.0 μg/ml) for (**I**) and 2.544×10^{-6}–1.272×10^{-5} M (0.8–4.0 μg/ml) for (**II**). Regression analysis indicates linear relationships with negligible intercepts. Table 1 presents the analytical parameters, molar absorptivity and the results of the statistical analysis of the experimental data: regression equations calculated from calibration graphs along with standard deviation of the slope (S_b) and intercept (S_a) on the ordinate and the standard deviation of residuals ($S_{y/x}$). The high values of the correlation coefficients of regression equations indicate good linearity and conformity to Beer's law.

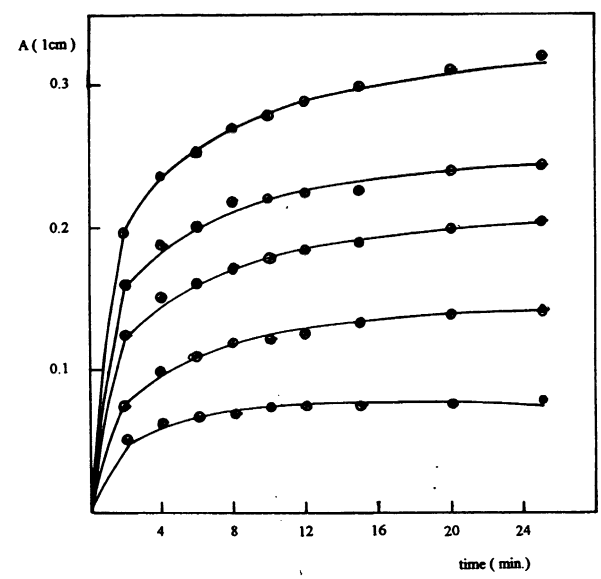


Fig. 6. Absorbance versus time graphs for the reaction between ranitidine and potassium permanganate at 25 °C showing the dependence of the reaction on ranitidine concentration. Ranitidine concentration 2.54×10^{-6}–1.27×10^{-5} M.

The detection limits [37] were 0.22 and 0.13 μg/ml (6.64×10^{-7} and 4.13×10^{-7} M) for (**I**) and (**II**), respectively. While the quantification limits were 0.73 and 0.42 μg/ml for **I** and **II**, respectively.

3.3. Accuracy and precision

Five replicate determinations at different concentration levels were carried out to test the precision and accuracy of the proposed method. The relative standard deviation (RSD) and standard analytical error (SAE) are shown in Table 1. The figures obtained point out to the good accuracy and repeatability of the method. The ruggedness of the method was studied by the within-day and between-day precision. Analysis of a 4.0 μg/ml sample solutions of **I** and **II** were performed 5 times a day for 4 consecutive days. The within-day coefficients of variation obtained were 1.80 and 1.53% for **I** and **II**, respectively, and the between-day coefficients of variation were 1.63 and 1.38% for **I** and **II**, respectively. The robustness of the method was demonstrated by the versatility of the experimental factors that affect the absorbance values.

3.4. Applications

The proposed method has been successfully applied to determine nizatidine (**I**) in capsules and ranitidine (**II**) in tablets. The concentrations of the drugs were calculated using the corresponding regression equations at fixed time of 20 min for (**I**) and 8 min for **II**. The results obtained are presented in Table 2. Statistical analysis of the results obtained by both the proposed method and reference spectrophotometric methods [3,14] revealed no significant difference in the performance of the two methods regarding accuracy and precision as revealed by *t*-test and *F*-test, respectively (Table 2).

Table 2
Application of the proposed kinetic method to the determination of nizatidine (**I**) and ranitidine (**II**) in their pharmaceutical preparations

Preparation	% Recovery ± SD[a]	
	Proposed method	Reference method
Axid capsules (nizatidine, 150 mg/capsule)[b]	98.47 ± 0.62	98.94 (1.50)
	$t = 0.65$, $F = 5.85$	
Axid capsules (nizatidine, 300 mg/capsule)[c]	98.29 ± 0.45	98.18 (0.53)
	$t = 0.35$, $F = 5.85$	
Zantac tablets (ranitidine, 150 mg/tablet)[d]	101.75 ± 0.63	101.80 (0.83)
	$t = 0.11$, $F = 1.74$	

[a] Mean of five separate determinations. Tabulated values of *t* and *F* are 2.31 and 6.39 at 95% confidence level.
[b] Product of Lilly, Indianapolis, IN, Batch No. 3144.
[c] Product of Lilly, Indianapolis, IN, Batch No. 1 MG-07N.
[d] Product of Glaxo–Wellcome, London, Batch No. K6199F.

3.5. Interferences

Interference due to common excipients and some related compounds was studied using glucose, sucrose, ascorbic acid, salbutamol, cimetidine and famotidine. The apparent concentration of (**I**) and (**II**) in these samples were determined and the tolerance limit (concentration of interfering substance causing less than 3% relative error) were calculated (Table 3).

Table 3
Effect of various foreign species on the determination of 4.0 μg/ml of nizatidine (**I**) or ranitidine (**II**) at the optimum conditions

Species	Tolerance limit (μg/ml)	
	Nizatidine	Ranitidine
Glucose	0.07	0.14
Sucrose	0.08	0.32
Ascorbic acid	0.09	0.21
Nizatidine	–	0.07
Ranitidine	0.25	–
Cemitidine	0.68	0.72
Famotidine	0.80	0.60
Salbutamol	0.07	0.07

4. Conclusion

The above results show the suitability of the proposed method for the kinetic determination of nizatidine (**I**) and ranitidine (**II**). The high reproducibility and sensitivity make the method applicable for routine analysis of (**I**) and (**II**).

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