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ORIGINAL ARTICLE

Kinetic spectrophotometric determination of hyoscine butylbromide in pure form and in pharmaceutical formulations

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Abstract A simple and sensitive kinetic method was described for the determination of hyoscine butylbromide in pharmaceutical preparations. The method is based upon a kinetic investigation of the oxidation reaction of the drug with alkaline potassium permanganate at room temperature for a fixed time of 15 min. The absorbance of the colored manganate ion was measured at 610 nm. The absorbance–concentration plot was rectilinear over the range of 1.0–10 μg mL^{-1} (r = 0.9999) and detection limit of 0.092 μg mL^{-1}. The concentration of hyoscine butylbromide was calculated using the corresponding calibration equation for the fixed-time method. The determination of hyoscine butylbromide by the fixed-concentration and rate constant methods is also feasible with the calibration equations obtained but the fixed-time method has been found to be more applicable. The different experimental parameters affecting the development and stability of the colors were carefully studied and optimized. The proposed method was applied to the determination of hyoscine butylbromide in pharmaceutical formulations. The results obtained were in good agreement with those obtained using the official British Pharmacopeial method (2004).

1. Introduction

Hyoscine butylbromide (HBB) (1S,3s, 5R, 7S,8r)-6,7-epoxy-3-[(S)-(3-hydroxy-2-phenylpropiony)oxy]-8-butyl-8-methyl-8-azoniabicyclol [3.2.1] octane bromide is used as an antispasmodic in treating peptic ulcer, gastritis and various disorders of the gastrointestinal tract which are characterized by spam. It has also found employment for the relief of spasmodic conditions of the bile duct and urinary tract and for the treatment of dysmenorrhoea (Crossland, 1980) (Fig. 1).

Hyoscine butylbromide is a white, crystalline powder or colorless crystals, efflorescent, freely soluble in water, soluble in alcohol. It melts at about 197 °C with decomposition,

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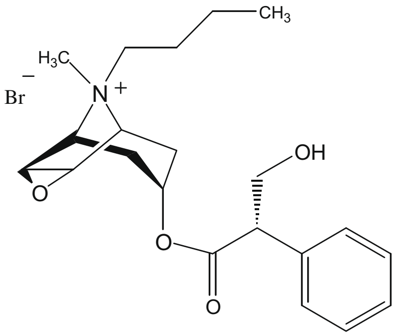

Figure 1 Chemical structure of hyoscine butylbromide (HBB).

determined after drying *in vacuo* for 24 h and then at 100–105 °C for 2 h and it should be protected from light (Neil Maryadele, 2001; British pharmacopoeia, 2004).

Due to the vital importance of HBB determination in pharmaceutical preparations and in biological fluids, many analytical techniques have been reported in the literature. HBB has been determined in pharmaceutical preparations using titrimetric (British pharmacopoeia, 2004; Falco and Vianna, 1979), spectrophotometric (Taha and Gomaa, 1975; Taha et al., 1974; Taha and Ruecker, 1997; Abdelkader et al., 1980; Issa et al., 2005; Thomos et al., 1994; Davidson and Hassan, 1984; Mahrous et al., 1992; Sharaf El-Din, 1994; Erk and Onur, 1996; Erk, 1996; Abramovich et al., 1993; Issopoulos and Pavlouzervou, 1994; Toral et al., 2005), high performance liquid chromatographic (Karali et al., 1998; Huller et al., 1993; Nakagawa et al., 2000; Wang and Zhu, 2000; Mandal et al., 1991; Pohjola and Harpf, 1994; Issopoulos and Pavlouzervou, 1994; Lau and Mok, 1997; Parissipoulou and Panderi, 1999), capillary electrophoresis (Cherkaoui et al., 1999; Chang et al., 2000) and electrochemical methods (Abadia et al., 1982; Ionescu et al., 1983; Cheng and Gao, 1983; Buschmann, 1987; Li et al., 1988; Ganjali et al., 2003, 2004), among other methods.

The literatures are still poor in analytical procedures based on kinetics especially for pharmaceutical or biological fluids. Furthermore, some specific advantages in the application of kinetic methods can be expected (Espinosa-Mansilla et al., 1998):

1. Selectivity due to the measurement of the evolution of the absorbance with the time of reaction instead of the measure of a concrete absorbance value.
2. Possibility of no interference of the colored and: or turbidity background of the samples.
3. Possibility of no interference of other active compounds present in the commercial product if they are resisting the chemical reaction conditions established for the proposed kinetic method.

In the present work, kinetically based method was proposed for the determination of HBB by measuring the absorbance at 610 nm after oxidation reaction with alkaline $KMnO_4$. Although, the poor selectivity of the proposed oxidation method with $KMnO_4$, it is simpler than the time consuming HPLC methods and is more sensitive than the other spectrophotometric methods.

The aim of the present work was to study the reaction between HBB and potassium permanganate kinetically in an attempt to evaluate the drug in dosage forms. The results obtained were promising. The proposed method was simple and did not need sophisticated instrument or special skill.

2. Experimental

2.1. Apparatus

All absorption spectra were made using Kontron 930 (UV–Visible) spectrophotometer (German) with a scanning speed of 200 nm/min and a band width of 2.0 nm, equipped with 10 mm matched quartz cells.

2.2. Materials and reagents

All chemicals and materials were of analytical grade and all solutions were freshly prepared in bidistilled water.

Pure grade hyoscine butylbromide (HBB) was kindly supplied by Amriya Pharmaceutical Industries, Alexandria, Egypt. Its purity was found to be 100.28 ± 0.77 ($n = 5$) according to BP method (British pharmacopoeia, 2004).

The following commercial formulations were subjected to the analytical procedures: Nu-Spasm tablets (Egyptian International Pharmaceutical Industries Company (EIPICO), Tenth of Ramadan City, Egypt) labeled to contain 10 mg HBB/tablet. Farcorelaxin ampoules (Pharco Pharmaceuticals Company, Al Amriya, Alexandria, Egypt) labeled to contain 20 mg HBB/mL. Spasmocin ampoules (Memphis Company for Pharmaceutics and Chemistry, Cairo, Egypt) labeled to contain 20 mg HBB/mL.

A stock solution (100 $\mu g\ mL^{-1}$) was prepared by dissolving 10 mg of HBB in 100 mL of distilled water and further diluted with the same solvent as appropriate.

Potassium permanganate (Merck, Darmstadt, Germany): 5×10^{-3} M aqueous solutions, freshly prepared and standardized. Sodium hydroxide (BDH, UK): 0.5 M aqueous solution.

2.3. Recommended general procedures

Transfer aliquots (0.1–1.0 mL) of 100 $\mu g\ mL^{-1}$ standard solution accurately measured, into series of 10 mL volumetric flasks; add 1.0 mL of 0.5 M NaOH followed by 2.0 mL of 5×10^{-3} M $KMnO_4$ to each flask and shake the mixture well. Allow the reaction mixture to stand for 15 min. Make up to the volume with bidistilled water. The HBB concentration was determined by measuring the rate of manganate formation at 610 nm as the tangent of the kinetic curve during the first 3.0 min of reaction and using the appropriate graphs. Log reaction rate versus log concentration of HBB was plotted to get order of the reaction. To get the standard calibration graph, the above procedure was carried out and the reaction mixture was allowed to stand for 15 min, where the absorbance of the reacting solution was measured at 610 nm against a blank solution prepared simultaneously. Plot the values of the absorbance against the final concentration in $\mu g\ mL^{-1}$ to get the calibration graph. Alternatively, derive the regression equation.

2.4. Procedures for pharmaceutical formulations

2.4.1. For tablets

Thoroughly powder and mix the contents of 10 tablets of the investigated drug and accurately weigh, an amount of the powder equivalent to 10 mg of HBB. Shake well with warm water for 5.0 min and complete to 100 mL with bidistilled water in a measuring flask. Filter if necessary and further dilute with water as required. The general procedure described above was used for the determination of the drug concentration using the corresponding regression equation.

2.4.2. For ampoules

The content of five ampoules was mixed and diluted to 100 mL with bidistilled water. An accurately measured volume equivalent to 10 mg of HBB was further diluted with water in a 100 mL volumetric flask to obtain a test solution of proper concentration. The general procedure described above was used for the determination of the drug concentration using the corresponding regression equation.

3. Results and discussion

3.1. Kinetic and optimization of the reaction conditions

The reaction between HBB and $KMnO_4$ in alkaline medium yields a green color due to the production of manganate ion, which absorbs at 610 nm (Fig. 2). As the intensity of the color increases with time, this was used as a useful method for the determination of HBB in pharmaceutical formulations.

The spectrophotometric properties of the colored product as well as the different experimental parameters affecting the color development and its stability were carefully studied and optimized. Such factors were changed individually while the others were kept constant. These factors include, effect of temperature, concentration of the reagents ($KMnO_4$ and NaOH), different solvents, and time.

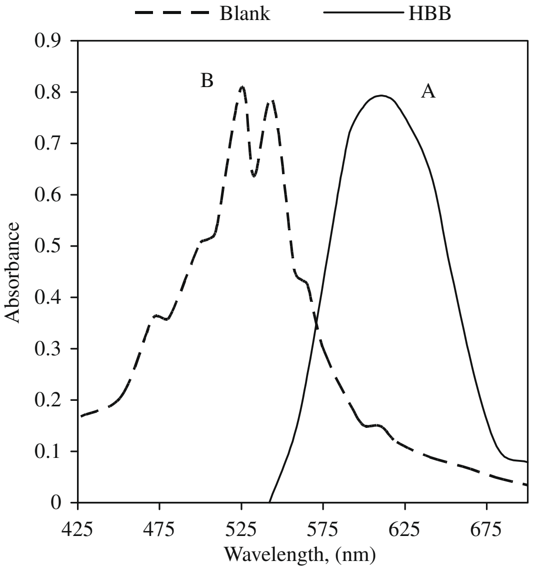

Figure 2 Absorption spectra of the reaction product of HBB after reaction with $KMnO_4$/NaOH system. (A) Reaction product 8.0 μg mL^{-1} against reagent blank (1×10^{-4} M $KMnO_4$) (B).

3.2. Effect of temperature

At room temperature, the reaction rate increased substantially as the color development increased. Therefore, room temperature was selected as the optimum temperature. Heating the solution was found to increase the rate of the reaction but MnO_2 was precipitated.

3.3. Effect of $KMnO_4$

The reaction rate and maximum absorbance increased with time, and with increasing $KMnO_4$ concentration. It was found that 2.0 ± 0.1 mL of 5×10^{-3} M $KMnO_4$ was adequate for the maximum absorbance (Fig. 3).

3.4. Effect of NaOH

The influence of NaOH concentration on the reaction rate was also studied using 0.2–3.0 mL of 0.5 M NaOH. It was found that increasing the volume of 0.5 M NaOH would increase the absorbance of the reaction product up to 1.0 mL, after which further increase in the volume of 0.5 M NaOH resulted in a very slight decrease in the absorbance of the reaction product, thus, 1.0 mL of 0.5 M NaOH was found to be the most suitable concentration for maximum absorbance (Fig. 4).

3.5. Effect of solvent

The effect of diluting solvent was also studied. Different solvents such as bidistilled water, ethanol, acetonitrile and dimethyl sulfoxide were used. It was found that bidistilled water was the best solvent as it gave the highest absorbance reading.

The rate of reaction was also found to be dependent on HBB concentrations. The rates were followed at room temperature with various concentrations of HBB in the range of 1.0–10 μg mL^{-1} keeping $KMnO_4$ and NaOH concentrations constant.

The reaction rate was found to obey the following equation:

$$\text{rate} = K' \times [\text{HBB}]^n, \tag{1}$$

where K' is the pseudo-order rate constant and n is the order of the reaction.

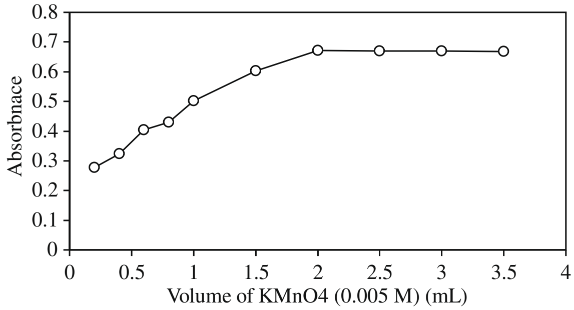

Figure 3 Effect of 5×10^{-3} M $KMnO_4$ volume on the absorbance of HBB (8.0 μg mL^{-1}) at 610 nm.

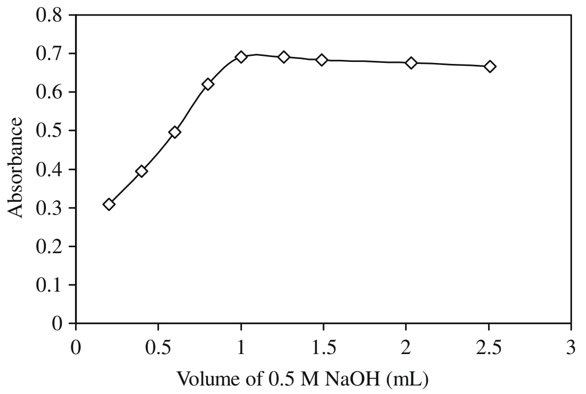

Figure 4 Effect of 0.5 M NaOH volume on the absorbance of HBB (8.0 μg mL^{-1}) at 610 nm.

Table 1 Logarithms of rates for different concentrations of HBB at room temperature at 610 nm.

$\log \Delta A/\Delta t$	log [HBB] M
−4.151	−5.644
−3.824	−5.343
−3.538	−5.042
−3.337	−4.86
−3.229	−4.741
−3.125	−4.644

$\log \Delta A/\Delta t = 0.9807 \log C - 1.5799$, $R^2 = 0.9995$.

The rate of the reaction may be estimated by the variable-time method measurement (Weisberger et al., 1953) as $\Delta A/\Delta t$, where A is the absorbance and t is the time in seconds. Taking logarithms of rates and concentrations (Table 1), Eq. (1) is transformed into:

$$\log(\text{rate}) = \log \Delta A/\Delta t = \log k' + n \log[\text{HBB}]. \tag{2}$$

At 610 nm; regression of log (rate) versus log (HBB) gave the regression equation:

$$\log \text{rate} = \log \Delta A/\Delta t = -1.5799 + 0.9807 \log C \quad (r = 0.9995).$$

Hence $K' = 2.63 \times 10^{-2}\,s^{-1}$ and the reaction is first order ($n = 0.9807$).

3.6. Evaluation of the kinetic methods

The quantitation of HBB under the optimized experimental conditions outlined above would result in a pseudo-first order with respect to their concentrations where $KMnO_4$ concentration was at least 20 times of the concentration of HBB and NaOH concentration was at least 250 times the initial concentration of HBB.

Several experiments were then carried out to obtain HBB concentration from the rate data according to Eq. (1). Initial rate, rate constant, fixed-concentration and fixed-time methods (Yatsimirskii, 1966; Laitinen and Harris, 1975) were tried and the most suitable analytical method was selected taking into account the applicability, the sensitivity, the intercept and the correlation coefficient (r).

Table 2 Values of K' calculated from slopes of log A versus t graphs at 610 nm.

$K' \times 10^{-4}$	[HBB]
−5.34	4.542×10^{-6}
−4.52	9.08×10^{-6}
−3.81	1.36×10^{-5}
−2.76	1.82×10^{-5}
−1.89	2.271×10^{-5}

3.7. Rate-constant method

Graphs of log absorbance versus time for HBB concentration in the range of 4.452×10^{-6} to 2.271×10^{-5} M were plotted and all appeared to be rectilinear. Pseudo-first order rate constants (K') corresponding to different HBB concentrations (C) were calculated from the slopes multiplied by −2.303 and are presented in Table 2. Regression of C versus K' gave the equation:

$$K' = -5.9051 \times 10^{-4} + 19.049C \quad (r = 0.9991).$$

3.8. Fixed-concentration method

Reaction rates were recorded for different HBB concentrations in the range of 9.08×10^{-6} to 2.271×10^{-5} M. A preselected value of the absorbance (0.4) was fixed and the time was measured in seconds and the reciprocal of time ($1/t$) varies in the range 7.937×10^{-4} to $2.87 \times 10^{-3}\,S^{-1}$. The reciprocal of time ($1/t$) versus the initial concentration of HBB was plotted and the following equation of the calibration graph was obtained:

$$1/t = -6.004 \times 10^{-4} + 150.87C \quad (r = 0.9985).$$

3.9. Fixed-time method

Reaction rates were determined for different concentrations of HBB. At a preselected fixed-time, which was accurately determined, the absorbance was measured. Calibration graphs of absorbance versus initial concentration of HBB were estab-

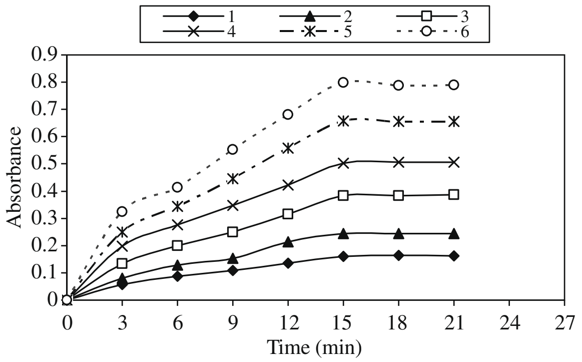

Figure 5 Absorbance versus time graphs for the reaction between HBB and MnO_4^-/OH^- system showing the dependence of the reaction on HBB concentrations. Concentrations of HBB are: (1) 2.271×10^{-6}; (2) 4.542×10^{-6}; (3) 9.08×10^{-6}; (4) 1.36×10^{-5}; (5) 1.82×10^{-5}; (6) 2.271×10^{-5} M.

Table 3 Regression equation for HBB at different fixed time over the range 2.271×10^{-6} to 2.271×10^{-5} M at room temperature at 610 nm.

Time (min)	Regression equation	Correlation coefficient (*r*)
3	$A = 0.0247 + 0.0283C$	0.9988
6	$A = 0.0567 + 0.0358C$	0.9992
9	$A = 0.0556 + 0.0491C$	0.9995
12	$A = 0.0714 + 0.0608C$	0.9995
15	$A = -0.0015 + 0.0795C$	0.9999

A is the absorbance at 610 nm and C is the concentration in μg mL^{-1}.

lished at fixed times of 3, 6, 9, 12, 15 min (Fig. 5) with the regression equations assembled in Table 3.

It is clear that the slope increases with time and the most acceptable values of the correlation coefficient (*r*) and the intercept were obtained for a fixed time of 15 min, which was therefore chosen as the most suitable time interval for measurement.

After optimizing the reaction conditions, the fixed-time method was applied to the determination of HBB in pure form over the concentration range 1.0–10 μg mL^{-1} and molar absorptivity 3.472×10^4 l mol C m^{-1}. Analysis of the data gave the following regression equations:

$$A = -0.0015 + 0.0795C \quad (r = 0.9999),$$

where A is the absorbance at 610 nm.

The LOD was found to be 0.092 μg mL^{-1} (2.09×10^{-7} M) and LOQ was found to be 0.3036 μg mL^{-1} (6.90×10^{-7} M).

Statistical analysis (Miller and Miller, 1993) of the results obtained by the proposed and official BP method (depending on HPLC with UV detection at 210 nm) (British pharmacopoeia, 2004), using Student's *t*-test (0.635) and variance ratio *F*-test (1.786) at $P = 0.05$ (the tabulated values of *t* (2.571) and *F* (6.256) revealed no significant difference between the

Table 4 Application of the proposed method to the determination of HBB in dosage forms.

Dosage form	Taken (μg/mL)	Found (μg/mL)	Recovery[a] (%)	
			Proposed method	Official method
Nu-Spasm tablets (10 mg HBB/tablet)	2	1.998	99.92	
	6	6.004	100.07	
	10	9.976	99.76	
Mean ± SD			99.92 ± 0.155	99.70 ± 0.16
Variance			0.024	0.0256
RSD			0.155	0.16
t[b]			2.30	
F[b]			1.067	
Spasmocin ampoules (20 mg HBB/mL)	2	2.022	101.11	
	6	5.992	99.86	
	10	9.974	99.74	
Mean ± SD			100.24 ± 0.759	99.89 ± 0.61
Variance			0.438	0.372
RSD			0.7569	0.611
t[b]			0.847	
Farcorelaxin ampoules (20 mg HBB/mL)	2	2.005	100.25	
	6	5.994	99.90	
	10	9.982	99.82	
Mean ± SD			99.99 ± 0.229	99.80 ± 0.15
Variance			0.0523	0.0225
RSD			0.2287	0.15
t[b]			1.65	
F[b]			2.324	

[a] The results are the average of six separate determinations.

[b] The theoretical *t*- and *F*-values (2.365) and (6.59), respectively for five degree of freedom at 95% confidence level.

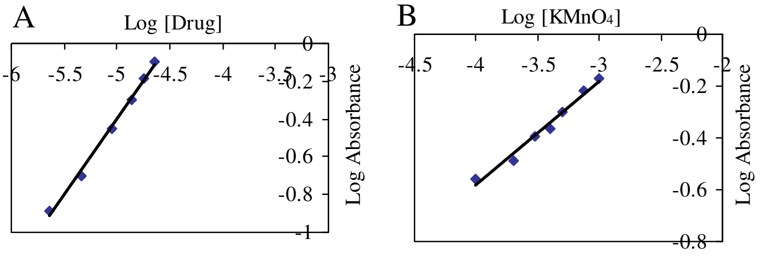

Figure 6 Limiting logarithmic plots for the molar ratio: (A) log A vs. log [Drug]; (B) log A vs. log [$KMnO_4$].

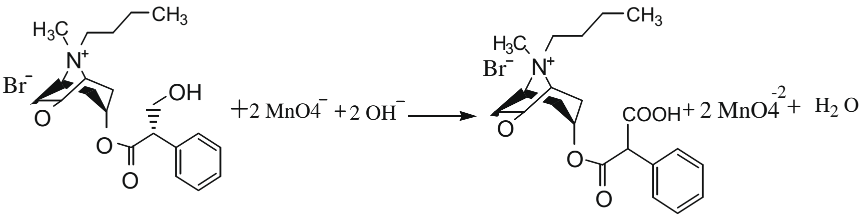

Figure 7 The proposed mechanism of reaction between HBB and $KMnO_4$/NaOH.

performance of the two methods regarding the accuracy and precision).

The precision of the method was evaluated by analyzing standard solutions of HBB. The results for pure sample were in accordance with those obtained by the BP method (British pharmacopoeia, 2004). The method was also applied to tablets or ampoules containing HBB (Table 4). The results agreed with those obtained by the BP method (British pharmacopoeia, 2004).

Excipients such as talc, starch, gelatin, magnesium stearate and lactose did not interfere with the assay.

3.10. Mechanism of the reaction

The stoichiometry of the reaction was studied adopting Bent and French method (1941). The absorbance of the reaction product was alternatively measured in the presence of excess of either $KMnO_4$ or HBB. A plot of log absorbance versus log [$KMnO_4$] and log [HBB] gave straight lines; the values of the slopes are 0.7962 and 0.3996, respectively (Fig. 6). Hence, it is concluded that, the molar reactivity of the reaction is 0.7962/0.3996, i.e. the reaction proceeds in the ratio of 2:1. Based on the obtained molar reactivity, the proposed mechanism of reaction between HBB and $KMnO_4$ in alkaline medium is shown in Fig. 7.

4. Conclusion

Different kinetic methods were established to determine HBB, where, the reaction rate, rate constant and fixed-time methods were applied over the concentration range 5.4×10^{-6} to 5.4×10^{-5} M HBB. The fixed-concentration method was applied over the range 2.70×10^{-5} to 5.4×10^{-5} M. Applying fixed-time method, it was clear that the slope increased with time and the most acceptable values of correlation coefficient (r) and intercept were obtained for a fixed time of 15 min which was therefore chosen as the most suitable time interval for measurements.

References

Abadia, V., Grovetto, G., Thomas, J., 1982. Cienc. Ind. Farm 1, 121.

Abdelkader, M.A., Taha, A.M., Abdelfattah, S., 1980. Pharmazie 35, 30.

Abramovich, B.F., Horvath, K.S., Gaal, F.F., 1993. Analyst 118, 899.

Bent, H.E., French, C.L., 1941. J. Am. Chem. Soc. 63, 568.

British pharmacopoeia, 2004. Her Majesty Sationary Office, London, p. 805.

Buschmann, N., 1987. Fresen Z. Anal. Chem. 326, 123.

Chang, Y.S., Ku, Y.R., Wen, K.C., Ho, L.K., 2000. J. Liq. Chromatogr. Relat. Technol. 23, 2009.

Cheng, G., Gao, H., 1983. Nanjing Yaoxueyuan Xuebao 3, 1.

Cherkaoui, S., Mateus, L., Christen, P., Veuthey, J.L., 1999. J. Pharm. Biomed. Anal. 21, 165.

Crossland, J. (Ed.), 1980. Lewis's Pharmacology, fifth ed. Churchill Livingstone Inc., New York, p. 224.

Davidson, A.G., Hassan, S.M., 1984. J. Pharm. Sci. 73, 413.

Erk, N., 1996. Sci. Pharm. 64, 173.

Erk, N., Onur, F., 1996. Anal. Lett. 29, 369.

Espinosa-Mansilla, A., Acedo Valenzuela, M.I., Salinas, F., Canada, F., 1998. Anal. Chim. Acta. 376, 365.

Falco, R.F., Vianna, M.J., 1979. Rev. Port. Farm. 29, 201.

Ganjali, M.R., Tahami, M., Poursaberi, T., Pazoukian, P., Javanbakht, M., Shamsipur, M., Baezat, M.R., 2003. Anal. Lett. 36, 347.

Ganjali, M.R., Norouzi, P., Golmohammadi, M., Rezapour, M., Salavati-Niasari, M., 2004. J. Electroanal. 16, 910.

Huller, G., Barthel, W., Klatt, H., Haustein, K.O., 1993. Pharmazie 48, 548.

Ionescu, M.S., Negoiu, D., Cosoferet, V.V., 1983. Anal. Lett. 16, 553.

Issa, Y.M., Youssef, A.F., Awady, M.A., 2005. Sci. Pharm. 73, 217.

Issopoulos, P.B., Pavlouzervou, E., 1994. Farmaco 49, 205.

Karali, N., Ozkirimli, S., Gursoy, A., 1998. Farmaco 53, 62.

Laitinen, H.A., Harris, W.E., 1975. Chemical Analysis, second ed. McGraw-Hill, New York.

Lau, O.W., Mok, C.S., 1997. J. Chromatogr. A 766, 270.

Li, B., Zhang, Z., You, X., Lu, T., Yin, G., 1988. Analyst 113, 57.

Mahrous, M.S., Daabees, H.G., Beltagy, Y.A., 1992. Spectrosc. Lett. 25, 389.

Mandal, S., Naqvi, A.A., Thakur, R.S., 1991. J. Chromatogr. A 547, 468.

Miller, J.C., Miller, J.N., 1993. In: Statistics for Analytical Chemistry, third ed. Ellis Horwood, Chichester, UK, p. 53 (Chapter 3).

Nakagawa, Y., Shimazu, T., Ishii, Y., Ishibashi, M., Hashimoto, Y., 2000. J. Mass. Spectrom. Soc. Jpn. 48, 42.

Neil Maryadele, J.O., 2001. Merk Index, 13th ed. Merk, Darmstadt, pp. 609.

Parissi-poulou, M., Panderi, I., 1999. J. Liq. Chromatogr. Relat. Technol. 22, 1055.

Pohjola, J., Harpf, M., 1994. J. Chromatogr. A 686, 350.

Sharaf El-Din, H.S., 1994. J. Pharm. Sci. 3, 119.

Taha, A.M., Gomaa, C.S., 1975. J. Pharm. Sci. 64, 1398.

Taha, A.M., Ruecker, G., 1997. Arch. Pharm. 310, 485.

Taha, A.M., Ahmad, A.K.S., Gomaa, C.S., El-Fatatry, H.M., 1974. J. Pharm. Sci. 63, 1853.

Thomos, K.M., Dabholkar, D.A., Jain, C.L., 1994. Indian Drugs 31, 391.

Toral, M.I., Munoz, M.A., Orellana, S.L., 2005. J. AOAC Int. 88, 1173.

Wang, T., Zhu, R., 2000. Yaowu Fenxi Zazhi 20, 392.

Weisberger, A., Friess, S.L., Lewis, E.S., 1953. In: Techniques of Organic Chemistry, Part I, vol. III. Interscience, New York.

Yatsimirskii, K.B., 1966. Kinetic Methods of Analysis. Pergamon Press, Oxford.