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Kinetic spectrophotometric methods for the quantitation of triprolidine in bulk and in drug formulations

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Abstract

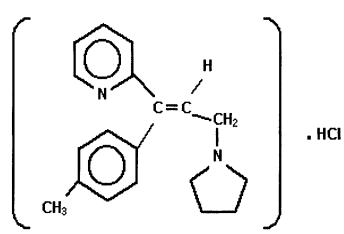
A kinetic method for the accurate and sensitive determination of triprolidine has been described. The method is based on the alkaline oxidation of triprolidine with $KMnO_4$. At a fixed time of 20 min, the formed manganate ion is spectrophotometrically measured at 612 nm. The concentration of triprolidine is calculated using the calibration equation for the fixed time method. Beer's law was obeyed from 6 to 40 μg ml^{-1} and the R.S.D. ($n = 10$) was 0.97%. Recovery was 99.80%. The method is suitable for quantitative determination of triprolidine in the presence of co-formulated drugs, since pseudoephedrine hydrochloride, which is frequently co-formulated with triprolidine did not interfere with this assay. The intra- and inter-day R.S.D. values indicated the ruggedness of the method. The method has been applied successfully to commercial tablet dosage form. The results obtained agreed with those obtained by the BP method. The determination of triprolidine by the fixed-concentration and rate constant methods is feasible with the calibration equations obtained, but the fixed time method proves to be more applicable.

Keywords: Kinetic determination; Triprolidine; Potassium permanganate; Actifed; Pharmaceutical analysis

1. Introduction

Triprolidine, chemically known as (*E*)-2-[3-(pyrrolidine-1-lyl)-1-*p*-tolylprop-1-enylpyridine is used as an antihistamine drug (Histamine H_1-receptor antagonist) [1].

Several methods involving high performance liquid chromatography (HPLC) and high performance thin layer chromatography [2–8] were reported for determination of triprolidine, all of which require lengthy treatment and extraction



Structural formula of triprolidine

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procedures. UV ratio spectroderivative and spectrophotometric methods have been proposed [9–12]. Colorimetric methods were reported depending on measurement of the color produced after reaction with 3-methylbenzothiazolin-2-one hydrazone [13]. In addition, voltammetric [14], and polarographic [15] procedures have been employed for the assay of triprolidine.

The British Pharmacopoeia [16] adopts a non-aqueous titration method, as does the United States Pharmacopoeia [17]. However, for the analysis of tablet formulations, the BP used the spectrophotometric procedure and the USP adopted a liquid chromatographic method.

In the present work, a kinetically based method is proposed for the determination of triprolidine by measuring the absorbance at 612 nm after oxidation reaction with alkaline $KMnO_4$. The aim of the present work was the development of a simple and sensitive analytical method for the assay of triprolidine in pharmaceutical tablets.

The proposed method is more sensitive than the non-aqueous titrimetric methods adopted by the BP or USP (for the drug in bulk) and is simpler than the time consuming HPLC method (for the drug in tablets). Furthermore, the BP spectrophotometric method for the assay of triprolidine in tablets using several extraction steps is tedious. This represents a serious disadvantage for these methods.

The proposed method is also not susceptible to interferences from common tablet excipients such as starch, talc powder, avisil, gelatin, and magnesium stearate.

2. Experimental

2.1. Apparatus

The study was conducted using a Pye Unicam PU 8800 UV/VIS Spectrophotometer (Philips) and 1.00-cm quartz cells.

2.2. Materials

All materials used were of analytical reagent grade. Reference triprolidine was kindly provided by the Wellcome Foundation Ltd., London and was used as received. Actifed tablets (batch number A8669A) were purchased from a local market (contains 2.5 mg triprolidine hydrochloride and 60 mg pseudoephedrine hydrochloride per tablet).

2.3. Reagents and solutions

Triprolidine standard solution, 0.5 mg ml^{-1} (1.5×10^{-3} M) was prepared in distilled water. The solution was stable for at least 5 days when kept in a refrigerator (at 4°C). A working solution (3×10^{-4} M) was prepared by dilution of 5 ml of the standard solution to 25 ml with distilled water. Potassium permanganate solution, 12 mg ml^{-1} was prepared in distilled water. Sodium hydroxide solution, 0.5 M was prepared and kept as a stock solution.

2.4. Sample solutions of triprolidine in formulations

From the average weight of 20 crushed tablets into fine powder, an accurately weighed quantity of the mixed powder containing an equivalent to 50.0 mg of triprolidine was dissolved in 5 ml of water. The solution was filtered through a Whatman No. 41 filter paper and then diluted to volume with distilled water in a 10-ml calibrated flask. An aliquot of this solution was diluted with distilled water as required and analyzed according to the mentioned procedure.

2.5. Procedure

An aliquot (0.4 ml) of $KMnO_4$ solution and 1.0 ml of 0.5 M NaOH solution were placed in 10-ml calibrated flasks. Accurate volumes of working solution (3×10^{-4} M) of triprolidine, over the concentration range 3×10^{-6}–6×10^{-5} M, were added and the solutions were diluted to volume with distilled water. At a fixed time of 20 min, the absorbance was measured directly at 612 nm against an appropriate blank. The triprolidine concentration was then computed from the corresponding equation of the calibration graph for the fixed-time method.

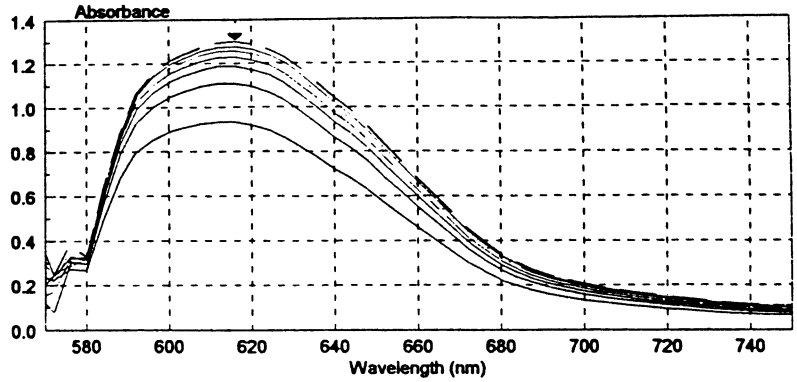


Fig. 1. Repetitive scan for the reaction of triprolidine (3×10^{-4} M) with $KMnO_4$ (3.04×10^{-3} M) in NaOH (0.05 M) medium, at room temperature with different time values (after 5, 10, 15, 18, 20, 25 and 30 min).

3. Results and discussion

3.1. Kinetics and optimization of the reaction conditions

The reaction between triprolidine and $KMnO_4$ in alkaline solution yields a green color as a result of manganate species, which absorbs at 612 nm. As the intensity of color increases with time (Fig. 1), it was deemed useful to elaborate a kinetically based method for the determination of triprolidine. In order to come to this conclusion, the reaction was investigated under various conditions of reagent concentration and alkalinity.

At room temperature, the reaction increased substantially with time, as revealed by the intensification of the developed color and subsequent increases in the slope of the calibration graph (Table 1), indicating higher analytical sensitivity. The reaction rate was found to increase with increasing temperature and a subsequent increase in the slope of the calibration graph. This indicates higher analytical sensitivity, but it results in poor linearity, and perhaps unwanted chemical changes might occur. Therefore, room temperature was selected as the optimum temperature.

The reaction rate and maximum absorbance increased with increasing $KMnO_4$ concentration and ultimately the adoption of 480 μg ml^{-1} $KMnO_4$ in the final solution proved to be adequate for maximum concentration of triprolidine used in the calibration curve. The influence of NaOH concentration on the reaction rate was studied between 0.005 and 0.2 M; it was found that increasing NaOH concentration increased the reaction rate with maximum absorbance being reached in a shorter time. It was also observed that there was no significant difference in the absorbencies of reactant solutions at NaOH concentrations above 0.05 M, while decreasing NaOH concentration resulted in lower absorbance values. Therefore, 0.05 M NaOH was chosen as the most suitable concentration.

Table 1
Calibration equations at different fixed times for triprolidine concentrations in the range 3×10^{-6}–6×10^{-5} M keeping NaOH (0.05 M) and $KMnO_4$ (3.04×10^{-3} M) at room temperature

Time (min)	Calibration equation	Correlation coefficient
5	$A = 8.486 \times 10^{-3} + 2.990 \times 10^{3} C$	0.9956
10	$A = 0.017 + 3.224 \times 10^{3} C$	0.9959
15	$A = 0.0246 + 3.314 \times 10^{3} C$	0.9966
20	$A = 0.027 + 3.376 \times 10^{3} C$	0.9999
25	$A = 0.0293 + 3.502 \times 10^{3} C$	0.9968

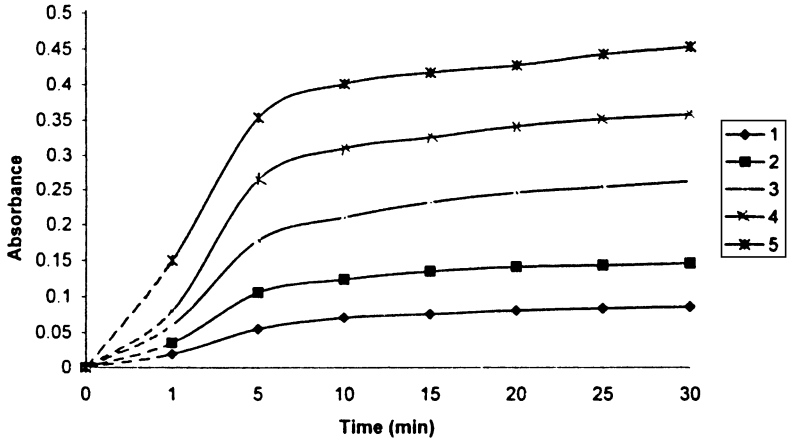


Fig. 2. Absorbance versus time graphs for the reaction of triprolidine and alkaline potassium permanganate showing the dependence of the reaction on triprolidine concentration. Concentration of triprolidine: 1, 1.8×10^{-5}, 2, 3.0×10^{-5}, 3, 6.0×10^{-5}, 4, 9.0×10^{-5}, and 5, 1.2×10^{-4} M.

The rate of the reaction was also found to be [triprolidine]-dependent. The rates were followed at room temperature with various concentrations of triprolidine in the range of 6–40 μg ml^{-1}, keeping $KMnO_4$ and NaOH constant at high concentrations as above.

The graphs shown in Fig. 2 were obtained, from which it is clear that the rate increases as the triprolidine concentration increases, indicating that the reaction rate obeys the following equation:

$$\text{Rate} = k' \text{ [triprolidine]}^n \quad (1)$$

where k' is the pseudo-order constant of the reaction.

The limiting logarithmic method [18] was used for the determination of the molar ratio between $KMnO_4$ and triprolidine in the reaction. This method depends on measuring the optical densities of solutions of $KMnO_4$ and triprolidine in which the concentrations of the two species are varied in turn at a constant total ionic strength.

The ratio may be found by plotting the logarithms of the absorbance (A) of the two sets of solutions versus composition, one with constant $KMnO_4$ concentration and variable triprolidine concentration, the other with constant triprolidine and variable $KMnO_4$ concentration. The slope of the curve in case 1 yields the number of moles of triprolidine while that in case 2 give the number of moles of $KMnO_4$ and so the composition of the compound produced can be evaluated (Fig. 3). The molar ratio was found to be 1:2 for triprolidine/$KMnO_4$.

Apparently, the reaction proceeds in two steps. The first step is fast and the second is the rate-determining step. Scheme 1 represents a proposed mechanism for the reaction between triprolidine and $KMnO_4$ in NaOH solution at room temperature [19]. This process is thought to proceed via cyclic permanganic esters, and finally to lead to the formation of 1,2-diol and other oxidation products [19].

From Fig. 2, the rate may be estimated by the variable-time method measurements [20,21], as $\Delta A/\Delta t$, where A is the absorbance and t is the time in seconds.

Taking logarithms of rates and concentrations (Table 2), Eq. (1) is transformed into

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

Regression of log [triprolidine] versus log (rate) by the least squares method yielded the calibration equation

$$\log \text{ (rate)} = 0.242 + 0.96 \log C \quad (3)$$

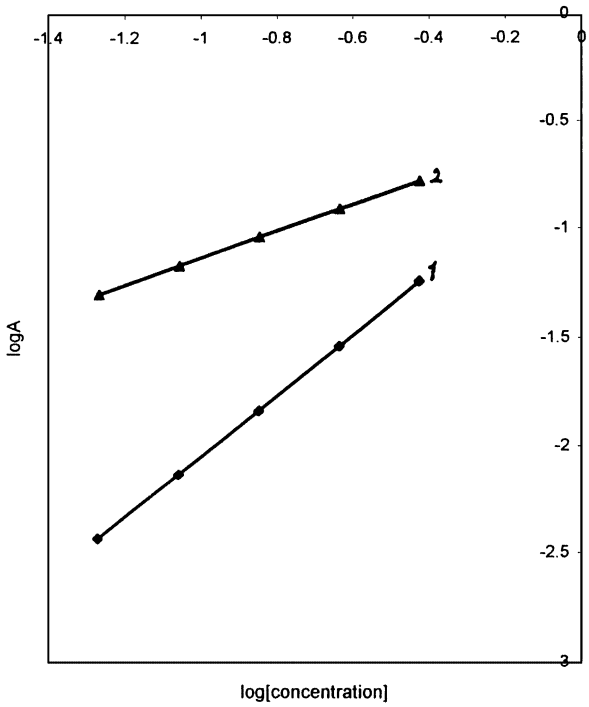


Fig. 3. Determination of the molar ratio between $KMnO_4$/triprolidine by limiting logarithmic method. (1) Set of solutions with constant triprolidine concentration and variable $KMnO_4$ concentration. (2) Set of solutions with constant $KMnO_4$ concentration and variable triprolidine concentration.

with correlation coefficient $r = 0.9998$. Hence $k' = 1.747\ s^{-1}$ and the reaction is first order ($n \cong 1$) with respect to triprolidine.

Table 2
Logarithms of the rates for different concentrations of triprolidine at constant concentration of $KMnO_4$ (3.04×10^{-3} M) and 0.05 M NaOH at room temperature

Log (rate), Log $\Delta A/\Delta t$	Log [triprolidine/M]
−4.307	−4.745
−4.187	−4.620
−4.097	−4.523
−4.014	−4.444
−3.815	−4.222
−3.658	−4.046
−3.524	−3.921

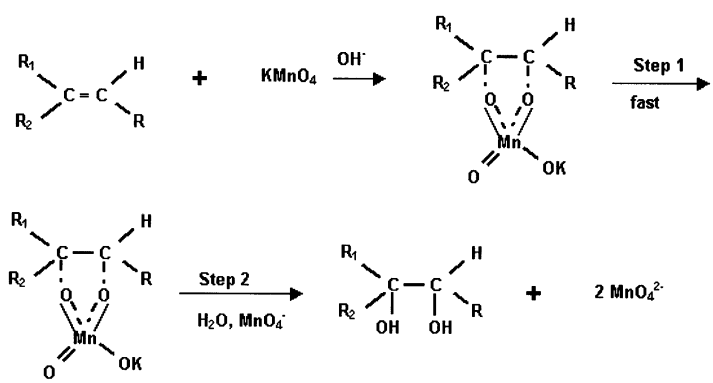
3.2. Evaluation of the kinetic methods

The quantitation of triprolidine under the optimized experimental conditions outlined above, where the $KMnO_4$ concentration was about 40 times the initial concentration of triprolidine, would result in a pseudo-zero order reaction with respect to $KMnO_4$. However, the rate will be directly proportional to triprolidine concentration in a pseudo-first order rate equation as follows:

$$\text{Rate} = k' \text{ [triprolidine]} \quad (4)$$

where k' is the pseudo-first order rate constant.

Eq. (4) was the basis for several experiments, which were run to obtain triprolidine concentrations using the rate data. Rate constant, constant concentration and fixed-time method [20,21] were tried and the most suitable analytical method was selected taking into account the applicability, the sensitivity (i.e. the slope of the calibration graph), the correlation coefficient (r) and the intercept.



Scheme 1. Proposed reaction between triprolidine and $KMnO_4$ in 0.05 M NaOH at room temperature.

3.2.1. Rate-constant method

Graphs of log (absorbance) versus time for triprolidine concentration in the range of 2.4×10^{-5}–1.2×10^{-4} M (8–40 μg ml^{-1}) were plotted and all appeared to be rectilinear.

Pseudo-first order rate constants corresponding to different triprolidine concentrations (C) were calculated from the slopes multiplied by -2.303 and are presented in Table 3. Regression of (C) versus k' gave the equation:

$$K' = 2.401 \times 10^{-2} - 394.759C,\ r = 0.858$$

The value of (r) indicates poor linearity, which is probably due to slight changes in the temperature of the reaction. Results obtained are summarized in the performance table.

3.2.2. Fixed-concentration method

Reaction rates were determined for different triprolidine concentrations in the range 3×10^{-5}–1.2×10^{-4} M. A preselected value for the absorbance was fixed and the time was measured in seconds. The reciprocal of time (i.e. $1/t$) versus the initial concentration of triprolidine (Table 4) was plotted and the following equation for the calibration graph was worked out by linear regression:

$$1/t = -4.44 \times 10^{-3} + 174.81C,\ r = 0.9997$$

The range of triprolidine concentration giving the most satisfactory calibration graph with the above equation was limited (9 μg ml^{-1}–40 mg ml^{-1}) and therefore this method was abandoned. The results obtained are summarized in Table 5.

Table 3
Values of k' calculated from slopes of log A versus t graphs multiplied by -2.303 for different concentrations of triprolidine at constant concentration of $KMnO_4$ (3.04×10^{-3} M) and 0.05 M NaOH, at room temperature

K' (s^{-1})	[Triprolidine] (M)
2.578×10^{-2}	0.24×10^{-4}
2.829×10^{-2}	0.30×10^{-4}
4.433×10^{-2}	0.36×10^{-4}
6.161×10^{-2}	0.60×10^{-4}
6.195×10^{-2}	0.90×10^{-4}
6.420×10^{-2}	1.20×10^{-4}

Table 4
Values of reciprocal of time taken at fixed absorbance for different rates of various concentrations of triprolidine at 3.04×10^{-3} M $KMnO_4$ and 0.05 M NaOH at room temperature

$1/t$ (s^{-1})	[Triprolidine] (M)
5.780×10^{-2}	0.30×10^{-4}
10.904×10^{-2}	0.36×10^{-4}
37.600×10^{-2}	0.60×10^{-4}
68.600×10^{-2}	0.90×10^{-4}
100.190×10^{-2}	1.20×10^{-4}

3.2.3. Initial-rate method: pseudo zero-order method

In this method, graphs of the rate (at the beginning of the reaction) versus triprolidine concentration were not easy to obtain, because the first step of the reaction was too fast to follow, so tangents of the curve were not easy to draw. This method was therefore abandoned.

3.2.4. Fixed-time method

Reaction rates were determined for different concentration of triprolidine. Calibration graphs of the absorbance versus initial concentration of triprolidine were obtained at fixed times of 5, 10, 15, 10 and 25 min with the calibration equations shown in Table 1. It is clear that both the slopes and intercepts increase with time. The best correlation coefficient and more reaction products (indicated by higher absorbance readings as shown in Fig. 2) were obtained for a fixed time of 20 min. Therefore, a fixed time of 20 min was chosen as the most suitable time for measurements. The detection limit [22] was 3×10^{-6} M (0.9 μg ml^{-1}), while the quantification limit was 1.8×10^{-5} M (6 μg ml^{-1}).

3.2.4.1. Accuracy and precision. Five replicate determinations at different concentration levels were carried out to test the precision and accuracy of the proposed method. The recovery was 99.80%. The relative standard deviation R.S.D. ($n = 5$) at 12 μg ml^{-1} and the percentage relative error were 0.97 and 1.01%, respectively. The ruggedness of the proposed method was studied by evaluating the coefficient of variation (five replicates) at 12

Table 5
Performance of the results for the determination of triprolidine by the proposed methods[a]

		Rate constant method	Fixed concentration method	Fixed time method
1	Linearity range (μg ml^{-1})	8–40	9–40	6–40
2	Regression equation	$K' = 2.4\times10^{-2} - 394.76C$	$1/t = -4.44\times10^{-3} + 174.81C$	$A = 0.027 + 3.38\times10^{3}C$
3	Correlation coefficient	0.858	0.9997	0.9999
4	R.S.D. (%)	2.3	1.1	0.97
5	Recovery (%)	97.41	98.17	99.80
6	LOD (μg ml^{-1})	4.8	6	0.9
7	LOQ (μg ml^{-1})	8	10	6
8	$S_{y/x}$*			4.18×10^{-3}
	S_a**			0.103
	S_b***			4.45

[a] C, Molar concentration; LOD, lower detection limit; LOQ, lower quantification limit.
* Standard deviation of the residuals.
** Standard deviation of the intercept.
*** Standard deviation of the slope.

μg ml^{-1} for the drug determination during 1 and 5 days. It was found to be 0.96 and 1.03%, respectively.

4. Applications

The fixed-time method was applied to determine triprolidine in the supplied drug formulation in tablet form. The concentration of triprolidine was calculated using the corresponding calibration equation shown in Table 1 at a fixed time of 20 min.

5. Conclusion

The results obtained for the analysis of triprolidine in drug formulation employed were compared with those obtained with the official BP method (Table 6).

The Student's t-test and F-test values for the 95% confidence level did not exceed the theoretical values of 2.306, and 6.39 for t- and F-tests, respectively indicating no significant difference between the accuracy and precision of the two methods.

The kinetically based method proposed in this work for the quantitation of triprolidine is selective in Actifed tablets as interference with pseudoephedrine and excipients did not occur. The tolerance limit (concentration of the interference substance that give 3% relative error) for the presence of pseudoephedrine in determination of triprolidine was found to be 256 μg ml^{-1} pseudoephedrine in 10 μg ml^{-1} triprolidine solutions. The method is also a direct method and more sensitive compared to the USP method. Furthermore, the proposed method does not require elaborate treatment and tedious extraction procedures usually associated with the USP and the BP methods.

Furthermore, the method can be used as a stability indicating assay due to the fact that the double bond in the propane moiety of the molecule is considered to be the weakest point in

Table 6
Determination of triprolidine in Actifed* tablets, by kinetic and BP official methods

	% Recovery		
	Kinetic method		BP method
Mean*	99.82 ± 0.93		99.59 ± 1.11
S.D. (%)	0.97		1.01
t-value**		0.367	
F-value**		1.084	

* Average of five determinations.
** Theoretical values of t and F at $P = 0.05$ are 2.306 and 6.39, respectively.

the triprolidine molecule, as well as the fact that the method is based on the oxidation of this double bond.

References

[1] J.E.F. Reynolds, Martindale The Extra Pharmacopoeia, 31st edn, Pharmaceutical Press, London, 1996, pp. 1053–1054.
[2] A. Avenoso, G. Facciola, G.M. Campo, A. Fazio, E. Spina, J. Chromatogr. B Biomed. Appl. 714 (2) (1998) 299–308.
[3] J.L. Chawla, R.A. Sodhi, R.T. Sane, Indian Drugs 34 (6) (1997) 339–345.
[4] D.D. Orsi, L. Gagliardi, A. Balasco, D. Tonelli, Chromatographia 43 (9-10) (1996) 496–500.
[5] M.J. Akhtar, S. Khan, M. Hafiz, J. Pharm. Biomed. Anal. 12 (3) (1994) 379–382.
[6] S.A. Volpidelli, F. Centorrino, P.R. Puopob, J. Kando, F.R. Frankenburg, R.J. Baldessarini, J.G. Flood, Clin. Chem. 39 (8) (1993) 1656–1659.
[7] E.B. Hasen, T.A. Getek, W.A. Korfmacher, J. Anal. Toxicol. 13 (3) (1989) 185–187.
[8] S.S. Yang, R.K. Gilpin, J. Chromatogr. Sci. 26 (8) (1988) 416–420.
[9] E. Dinc, F. Onur, Stp Pharma Sci. 8 (3) (1998) 203–208.
[10] A.G. Davidson, L.M. Mkoji, J. Pharm. Biomed. Anal. 6 (5) (1988) 449–460.
[11] R. Jones, Analyst 112 (11) (1987) 1495–1498.
[12] P.J. Gemperline, S.E. Boyette, K. Tyndal, Appl. Spectrosc. 41 (3) (1987) 454–459.
[13] C.S. Sastry, A.S. Tipirneni, M.V. Suryanarayana, J. Pharm. Biomed. Anal. 8 (3) (1990) 287–292.
[14] E. Bishop, W. Hussein, Analyst 109 (7) (1984) 912–913.
[15] G. Kanoute, E.G. Nivaud, M. Guernet, J. Pharm. Biomed. Anal. 6 (6-8) (1988) 977–981.
[16] The British Pharmacopoeia (BP) 1993, HMSO, London, 1993, pp. 1 (689, 1143).
[17] The United States Pharmacopoeia (USP) XXIII (NF XVIII) US Pharmacopeial Convention, Twinbook Parkway, Rockville, 1995 pp. 1605–1666.
[18] J. Roso, Advanced Physiochemical Experiments, Sir Issac Pitman and Sons, London, 1964, p. 67.
[19] P. Sykes, A Guidebook to Mechanisms in Organic Chemistry, 4th edn, Longman, London, 1978, p. 186.
[20] H.A. Laitinen, W.E. Harris, Chemical Analysis, 2nd edn, McGraw-Hill, New York, 1975.
[21] K.B. Yatsimirskii, Kinetic Methods of Analysis, Pergamon Press, Oxford, 1966.
[22] J.C. Miller, J.N. Miller, Statistics for Analytical Chemistry, Wiley, New York, 1984.