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Conductometric Titration for Determination of Trazodone Hydrochloride and Solubility Products of its Ion-Associated Complex Species

Fathy M. Salama, Khalid A.M. Attia, Ragab A. M. Said, Ahmed El-Olemy, Ahmed M. Abdel-raoof *

Pharmaceutical Analytical Chemistry Department, Faculty of Pharmacy, Al-Azhar University, 11751, Nasr City, Cairo, Egypt

***Corresponding author:** Ahmed M. Abdel-raoof, Pharmaceutical Analytical Chemistry Department, Faculty of Pharmacy, Al-Azhar University, 11751, Nasr City, Cairo, Egypt. Tel.: +201092000774*
E-mail address: ahmedmeetyazeed79@yahoo.com

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ABSTRACT

Objectives: Conductometric determination of trazodone hydrochloride, with two precipitating reagents; Ammonium Reineckate and sodium tetraphenylborate has been investigated. **Methods:** The formed ion-associates were studied conductomterically for the determination of solubility products and other functions associated with the process of precipitating trazodone hydrochloride were determined. Moreover, two new maneuvers towards equivalence point detection were carried out. In this study, data processing was performed using numerical derivatization (second derivative) and Boltzmann algorithm. **Results:** Although, the described procedures allowed the determination of trazodone hydrochloride within the range of 2-14 mg using both reagents has less sensitivity other than reported method but has more advantages in simplicity, low cost , relatively short analysis time and direct analysis with good recoveries between 100.01% and 100.07% with relative standard deviation less than 2%. **Conclusion:** These methods were validated and successfully applied for the determination of trazodone hydrochloride in Trittico® tablets. The obtained results were statistically compared with those of the official method by applying t-test and F-value at 95% confidence level and no significant difference was observed regarding accuracy and precision.

Keywords: *Boltzmann sigmoid method; Conductometric; Numerical derivatization; Trazodone hydrochloride*

INTRODUCTION

Trazodone (TZ), (2-{3-[4-(3-chlorophenyl) piperazin-1-yl] propyl}-2H, 3H-[1, 2, 4] triazolo[4, 3-a] pyridin-3-one), is a selective serotonin reuptake inhibitors (SARI) that is used as an antidepressant[1]. The official method for analysis of trazodone hydrochloride based on HPLC using octadecylsilane column and water-0.01 M ammonium phosphate buffer pH 6.0 (60: 40) as mobile phase[2] and potentiometric non-aqueous titration with perchloric acid[3].The analytical methods that are reported for the determination of trazodonehydrochloride in pharmaceutical formulations include spectrophoto-metric methods[4-7] ion-selective electrodes[8,9] voltammetry[10-13] and various chromatographic methods including; HPLC [2,14-17] capillary gas chromatography[18], liquid chromatography–tandem mass spectrometry[19] and instrumental thin layer chromatography[20]. Also, spectrofluorimetric methods have been reported [21,22]. Reineckate salt (RK) which is ammonium tetrathiocyanato-diamminechromate (III) monohydrate and sodium tetraphenylborate(TPB) salt; have been used for quantitative determination of many pharmaceutical compounds using several analytical techniques like spectrophotometric, conductometric and Atomic absorption spectroscopic procedures [23-26].

The current paper based on the use of the use of the differential conductivity methods and Boltzmann sigmoid function "without integration" for locating the equivalence point. The first derivative data were fitted to a built-in nonlinear regression model while approximations to Gaussians of the second derivative data were followed to locate the end point and

Boltzmann models with a perspective of avoiding the uncertainty resulting from locating the endpoint as the break in the conductance-volume curves [27,28]. In addition, we depend on the data obtained from conductometric titration of TZ to calculate the solubility product of the formed ion associates and hence the equilibrium formation constant of the investigated reactions.

MATERIALS AND METHODS

Apparatus

JENWAY model 4510 Conductivity / TDS Meter (451001990), was used in conductance measurements with a dip type conductivity cell of two Pt (non-polarized) electrodes of 1.0 cm^2 in area, rigidly fixed at 1.0 cm apart manufactured in the EU by Barloworld Scientific Ltd, Dunmow, Essex, CM6 3LB. Elementar-Vario El (Germany) was used for elemental analysis (C, H, N, and S) of the ion pairs. Microcal Origin 8.0 (Microcal Software Inc., version 8E) computer program was applied in data treatment for graphical and statistical treatments and calculations.

Chemicals and reagents

All reagents used were of pure grade. Double distilled water was used throughout the experiments.

- Trazodone hydrochloride ($C_{19}H_{22}ClN_5O.HCl$) powder was kindly supplied by Egyptian International Pharmaceutical Industries Company (Eipico) 10th of Ramadan City, Egypt. Its purity was 100.35 ± 0.21 % according to B.P. method [2] (Batch. NO.A0967912).

- Trittico®tablets, labeled to contain 50 mg of trazodonehydrochloride per tablet manufactured by Egyptian International Pharmaceutical Industries Company (Eipico) 10th of Ramadan City, Batch No.1608579 and purchased from the local market.

- Ammonium reineckate [$NH_4(Cr(NH_3)_2(SCN)_4).H2O$] andsodium tetraphenylborate [$NaB(C_6H_5)_4$] (Sigma-Aldrich, Germany).The 1 x 10^{-2}M and 5 x 10^{-3} M (RK,TPB and TZ) solutions were prepared in doubly distilled water.

Working procedure

Procedure for pure pharmaceuticals

A range of volumes containing 2 – 14 mg of the pure TZ solution were transferred into the titration cell and the volume was made with water up to 50 ml. The conductivity cell was immersed in and the solution was titrated with 5 x 10^{-3} M of the titrant (RK or TPB). The conductance was measured 2 minutes subsequent to each addition of the reagent after thorough stirring. The measured values were corrected for volume change to eliminate the effect of dilution on the increase in conductance by means of the following equation, assuming that conductivity is a linear function of dilution:

$$\mathbf{kcorr = kobs[(V_o + V_{added}) / V_o]}$$

Where, kobs, the observed specific conductivity, V_o, the initial volume, and V_{added}, the added volume. The corrected conductivity was then plotted against the volume added of titrant and the second derivative or Boltzmann sigmoid method were used to estimate the end point and the stoichiometric ratios [31-33]. The nominal content of the compound under study was calculated using the following equation:

Amount of the drug (mg) = VMR / N

Where V= volume (ml) of the titrant consumed in the titration, M= relative molecular mass of the analyte, R= molarity of the titrant, and N= number of moles of the titrant consumed per one mole of the analyte.

Stoichiometric ratios determination

A definite volume (5 ml) of 5 x 10^{-3} M TZ was transferred to a 50 ml volumetric flask and made up to the mark with double distilled water. The drug solution was placed in a suitable titrating vessel and the conductivity cell was immersed, then a titrant of 5 x 10^{-3} M of TPB was added from a burette. The solution was stirred for 1-2 min and allowed to attain equilibrium and the end point was determined as previous procedure mentioned before.

Procedure for Tablets

Ten Trittico® tablets were accurately weighed and finely powdered, then a quantity equivalent to 100 mg of trazodone hydrochloride was shaken three times with 25 mL of water for 15 minutes then filtered into 100-mL volumetric flask and the volume was adjusted to the mark with water to obtain a concentration of (1mg mL^{-1}). The nominal content of the active component in tablets was determined as described in the Procedure section.

Ion-associates preparation

Ion-associates synthesis protocol included addition of 10^{-2} M aqueous solution of ion pairing agents (RK and TPB) drop wise to 40 ml of 10^{-2} M TZ solution. The mixture was left to react for 60 min under stirring at room temperature. The resulting precipitate was then filtered off on Whatman filter paper and was then filtered off on Whatman filter paper and washed several times with bidistilled water. The compound was left to dry for 12h at 60 °C, washed with petroleum ether to remove any residual moisture, and then ground to fine powder [29, 30]. Small sample portions were sent to elemental analysis and IR.

Table 1. Elemental analysis data for various TZ-IPs

IP complex	M.Wt calculated	C%		H%		N%		S%		Molecular formula
		calculated	found	calculated	found	calculated	found	calculated	found	
TZ-RK	726.28	38.00	37.47	4.68	4.88	23.13	23.38	17.57	18.30	$[C_{19}H_{22}ClN_5O][C_4H_{12}OCrN_7S_4]$
TZ-PB	691.06	74.66	70.42	6.08	5.83	10.12	9.39	-	-	$[C_{19}H_{22}ClN_5O][C_{24}H_{20}B]$

Table 2. Conductometrically measured solubility (S), solubility products (KSP), and formation constants (k) of various ion-associates

Ion pair complex	λ_oIP	K_S	S	K_{SP}	K=1/K_{SP}
TZ-RK	1.56×10^{8}	4.03	2.58×10^{-5}	6.66×10^{-10}	1.50×10^{9}
TZ-TPB	1.52×10^{8}	3.01	1.97×10^{-5}	3.89×10^{-10}	2.57×10^{9}

Solubility products and other constants determination

Series of solutions of different concentrations ($C = 10^{-5}$-10^{-2} M) were prepared for each of TZ, RK and TPB. The conductivities of these solutions were measured at 25ºC and the specific conductivities (k), corrected for the effect of dilution were calculated and used to obtain the equivalent conductivities (λ) of these solutions.

$$\lambda = 1000\ k / C$$

λ (at a finite concentration) and λo (at infinite dilution) can be related by Onsanger equation [34]:

$$\lambda = \lambda o - (a + b\ \lambda o)\ C^{1/2}$$

Where, (a) and (b) are constants related to the interionic forces (accounting for the electrophoretic and the time of relaxation effect, respectively). Kohlrausch's law of the square root of concentration predicts a nonlinear relation between conductivity and concentration at a lower concentration range. Straight line plots of λ versus $C^{1/2}$, were constructed and the equivalent conductance values at infinite dilution (λo TZ, λo RK and λo TPB, were determined from the intercept of the respective line with the λ axis. The activity coefficients were taken as unity since the solutions were sufficiently dilute. The equivalent conductance values of the IPs under complete dissociation condition (λo IP) were calculated from Kohlrausch's law of independent migration of the ions [27, 34-35].

$$\lambda o\ IP = n\ \lambda o\ TZ + \lambda o\ (ion\ pairing\ agent)$$

Where; n is the stoichiometric ratio. The solubility (S) and the solubility product (Ksp) of a particular ion associate were calculated using the following equations:

$$S = k_s \times 1000 / \lambda oIP$$
$$Ksp = S^2 \text{ for 1: 1 ion associate}$$

Where k_s, is the specific conductivity of a saturated solution of the ion associate, at 25ºC. The saturated IP solutions were prepared by stirring the IP suspensions in bidistilled water for 5h and then left for 24h before measurement [36].

RESULTS AND DISCUSSION

TZ ($C_{19}H_{22}ClN_5O$) is a tertiary amine cation having a high affinity towards the formation of water insoluble ion pair (IP) complexes with the oppositely charged anions such as RK or TPB. Elemental analysis revealed that TZ form ion association with TPB and RK in a stoichiometric ratio of 1: 1 (drug: titrant) **Table 1.** Conductance measurements have been used successfully in quantitative titration systems where the conductance of the solution varied prior to and after the equivalence point. The titration curve, by plotting the change in conductance versus volume of titrant added, represented two straight lines intersecting at the end point. The first segment corresponds to the formed precipitate and the second segment represents the excess of ion pair agent (RK or TPB) **Figure 1**.

In the present study, two proposals were considered. The first plan depended on numerical derivatization of the raw data, while in the second

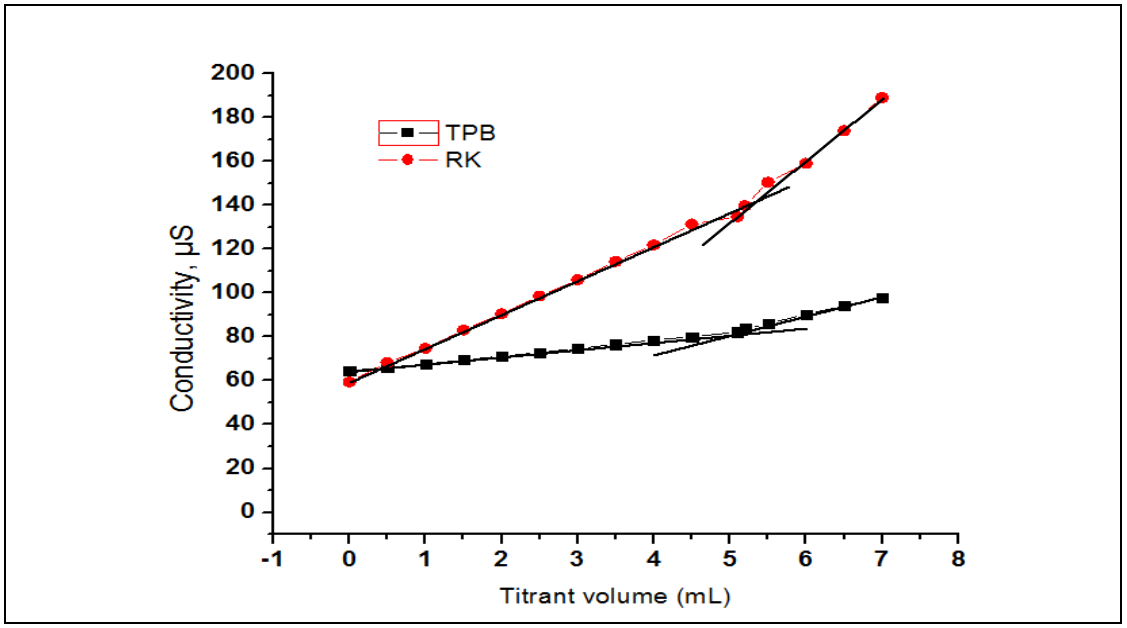


Figure1. Conductometric titration curve of 5mL 5x10^{-3}mol/L TZ titrated with 5x10^{-3}mol/L RK and TPB by conventional procedure for locating the endpoint.

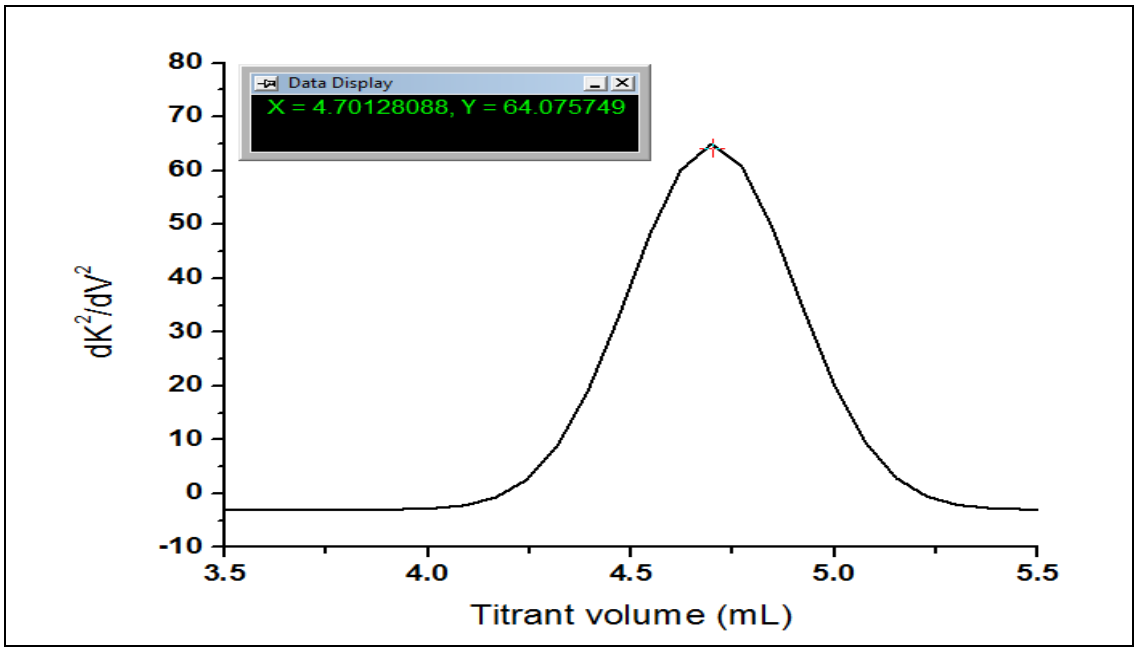


Figure 2. Conductometric titration of 5mL 5x10^{-3}mol/L TZ with 5x10^{-3}mol/LRK applying the numerical second derivative plot (dk^2/dV^2), fitted to Gaussian .Screen reader denotes the endpoint determined by the procedure.

experimental figures were well suited to a Boltzmann sigmoid model. The mathematical differentiation of the obtained conductivity data against the corresponding titrant volume was one of the suggested schemes, **Figures 2, 3**.

Applying the second derivative mode, the endpoint was located as the curve maximum and as defined by the fitting parameters while in the first derivative, the endpoint was located at the halfway point on the fitted line-between two shoulders which is inaccurate in comparison to second derivative. Alternatively, the process of numerical handling of data

Table 3. Recovery study of TZ by adopting standard addition technique using the proposed method

Pharmaceutical taken (mg)	Pure added (mg)	Pure found (mg)		Recovery %	
		RK	TPB	RK	TPB
2	2	1.97	1.99	98.5	99.50
	4	4.03	4.05	100.75	101.25
	6	6.03	5.93	100.5	98.83
	8	8.03	7.91	100.38	98.88
	10	9.99	10.19	99.90	101.9
Mean				100.01	100.07
% RSD				0.896	1.415

data creates unusual behavior represented in a noisy first derivative sigmoid. Noticeably, this problem arises from consolidating the instinctive experimental errors. These errors, in turn, are amplified after the numerical processing. The other proposal, Boltzmann paradigm is based on the fit of the experimental raw data to a simple nonlinear function obtained by direct integration of a Boltzmann type sigmoidal function. Besides, being a nonlinear curve fit in most of software, Boltzmann type sigmoid provides a straightforward and simple correlation between the function parameters and the conductivity-volume curve characters. This model has been described by the following equation [26-28]:

$$f(x) = \frac{A1 - A2}{1 + e^{(X - X0)/dx}} + A2$$

The parameters A_1 and A_2 stand for the asymptotic value for small and large values of x respectively, x_o represents the endpoint and expressed as the central point of transition and dx deals with the width of the transition. **Figures 4, 5** show the determination of equivalence point applying Boltzmann type sigmoid. The mathematical expression of Boltzmann shows the simplicity of this model where the value of x_o is simply obtained as $f(x_o) = (A_1 + A_2)/2$.

Determination of solubility products of the ion-associates

The solubility of an ion-exchanger is one of the main factors controlling the life-span of the sensor incorporating it as a sensing material. Determination of the solubility product of an ion-pair is very important since its reciprocal is approximately equal to the formation constant, which in turns is tightly related to the degree of hydrophobicity of the ion-exchanger. Since, as the hydrophobicity of the IP increases, the leaching rate into the aqueous bathing solution decreases.

Determination of the solubility product of the IP is very important since as the hydrophobicity of the IP increases, the leaching rate into the aqueous bathing solution decreases. The solubility of an ion-exchanger is one of the main factors controlling the life span of the sensor which incorporate it as electroactive material [37-39].

According to Kohlrausch's law of independent migration of the ions, the molar conductivity of an electrolyte equals the sum of the molar conductivities of the cations and the anions [29,40]. The equivalent conductance (λ) of an ion is the conductance of a solution of unspecified volume containing one gram-equivalent and measured between electrodes 1 cm apart. Due to interionic effects, (λ) is concentration dependent, and the limiting ionic equivalent conductance (λo) at infinite dilution (no disturbing effect on the mobilities of ions other than solvent and temperature) reaches its maximum value and used for comparison purposes. The magnitude of (λo) is determined by the charge, the solvent viscosity, size and the magnitude of the applied potential. λo TZ, λo RK and λo TPB can be determined from the intercept of the respective line with the λ axis) from straight line plots of λ versus $C^{1/2}$ **Figure 6**. Hence, the equivalent conductance of the solvated IPs (λo IP) at infinite dilution could be calculated as follow:

$$\lambda oTZ\text{-}RK = \lambda oDex + \lambda o\ RK$$
$$\lambda oTZ\text{-}TPB = \lambda oDex + \lambda oTPB$$

The solubility products (Ksp) of the ion associates were determined conductometrically and found to be 6.66×10^{-10} and 3.89×10^{-10} for TZ-RK and TZ-TPB, respectively **Table 2**. The very low solubility of TZ-TPB IP ($S = 1.97\times10^{-5}$ M) and consequently, the high formation constant value ($k = 2.57\times10^{9}$), revealed that the degree of completeness of the reaction was more than 99.9%. At equilibrium, the solubility product

Table 4. Determination of TZ in Trittico® 50 mg tablets using the proposed RK and TPB methods compared to the official method [2]

Parameters	Proposed method		Official method [2]
	RK	TPB	
n*	5	5	5
$\bar{X}$**	100.80	100.19	99.14
%RSD	1.331	1.025	0.742
t***	2.428(2.306)	1.857(2.306)	____
F***	3.324(6.388)	1.949(6.388)	____

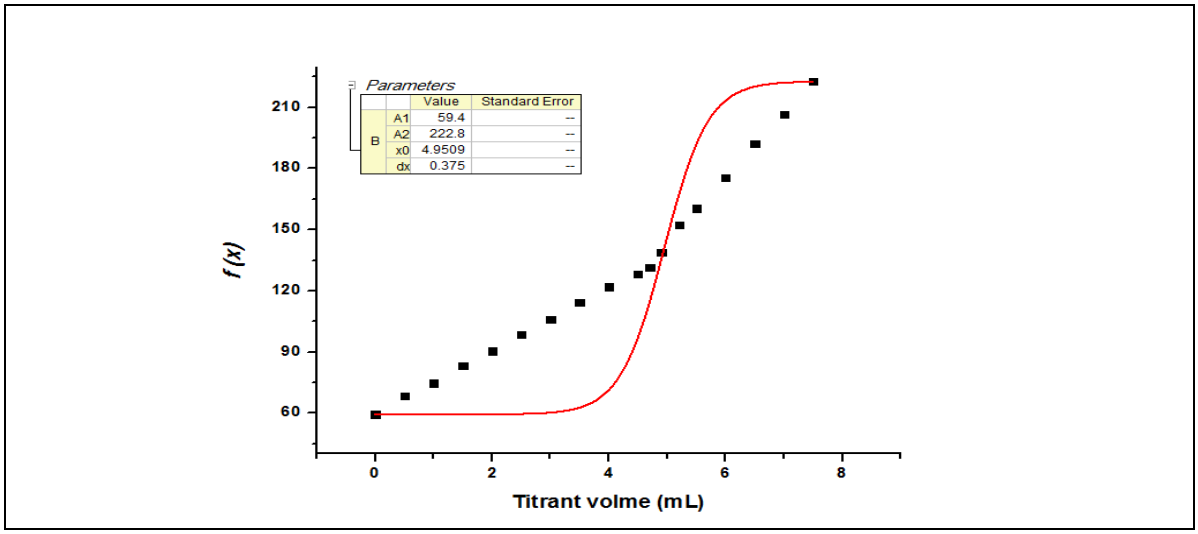


Figure 4. Conductometric titration of 5mL 5x10^{-3}mol/L TZ with 5x10^{-3}mol/L RK applying the Boltzmann sigmoid method f(x). Value of x_0 stands for the equivalence point determined using Boltzmann model.

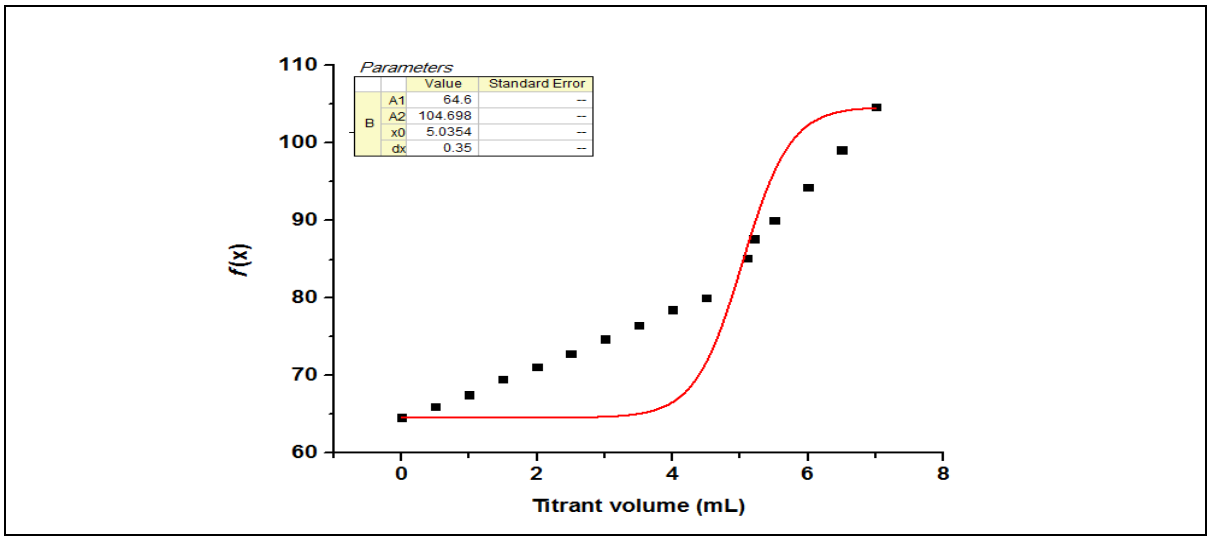


Figure 5. Conductometric titration of 5mL 5x10^{-3}mol/L TZ with 5x10^{-3}mol/L TPB applying the Boltzmann sigmoid method f(x). Value of x_0 stands for the equivalence point determined using Boltzmann model.

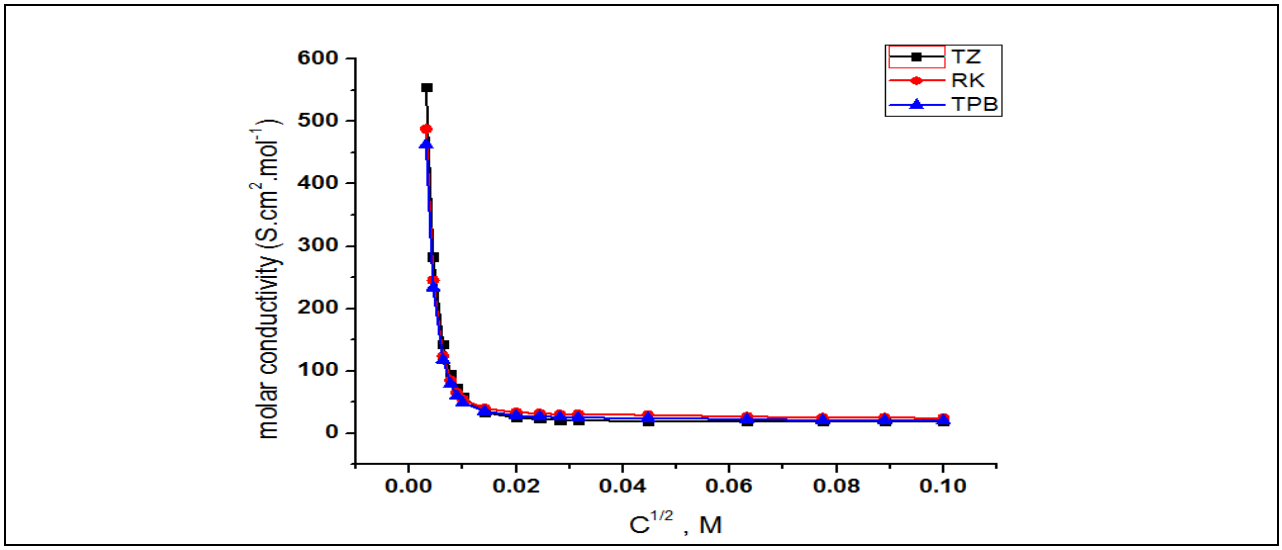
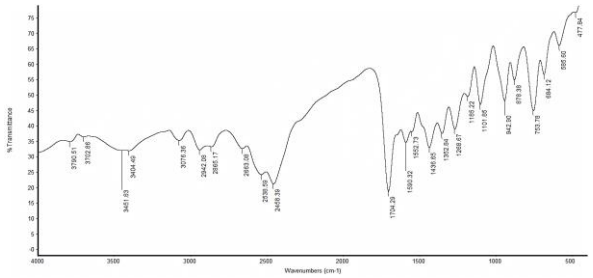
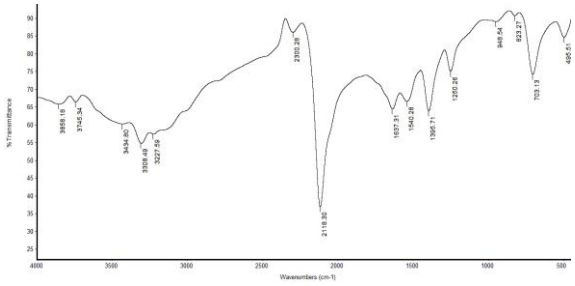


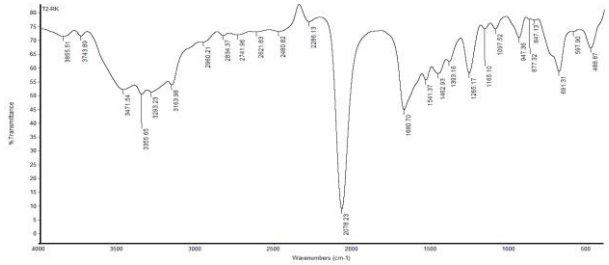
Figure 6. Molar conductance vs. the square root of concentration $C^{0.5}$ for TZ, RK and TPB



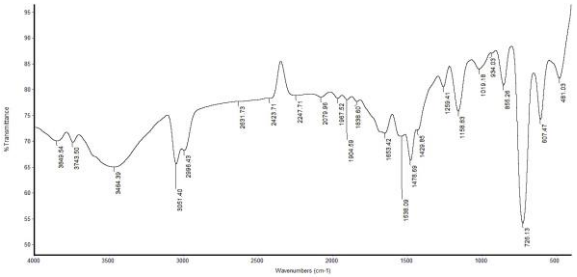
7-a. IR spectra of TZ



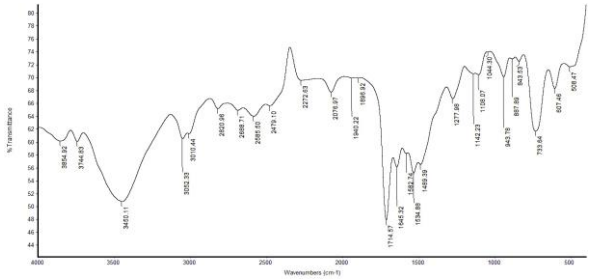
7-b. IR spectra of RK



7-c. IR spectra of RK-TZ



7-d. IR spectra of TPB



7-e. IR spectra of TPB-TZ

Figure 7(a-e). IR spectra of TZ, RK, TPB free ligands and the ion associates.

of the undissociated IP in water (the intrinsic solubility) was omitted as this term makes a negligible contribution to the total solubility because the IPs were sparingly soluble in water and their saturated solutions were, therefore, very dilute [38,39,41].

IR Spectra

The IR spectrum of Amm.Rt has a characteristic band at 2118 cm-1 due to ν(CN) "in the Cr-NCS link" stretching vibration, a band at 703 cm^{-1} due to νsym (C-S) and at 495 cm^{-1} due to δ(NCS) deformation vibration [42]. The IR spectrum of the formed ion associate shows a weak band corresponding to *ν*CH (aliphatic) at 2960 cm^{-1}. The band corresponding to the stretching vibrations of C=O shifted to a lower frequency by ~ 24 cm^{-1}. In addition, the peak due to *ν*NCS is shifted to a lower frequency by 40 cm^{-1}. Peaks due to *νsym*(C-S) and *δ* (NCS) appear at 691 and 489 cm^{-1} respectively. The above IR interpretation indicates that an ion associate has been formed between TZ and RK. On the other hand, the band corresponding to the stretching vibrations of C=O in TZ intact was shifted to a higher frequency in ion associate complex (TZ-TPB) by ~ 10 cm^{-1} and the strong band peak in TPB at 726 (four monosubstituted benzene rings attached to boron atom) was shifted to a higher frequency in ion associate complex by ~ 7 $cm^{-1}$and the band peak belongs to B-C at 1019 cm^{-1} was disappeared at an ion associate complex. The above IR interpretation indicates that an ion associate has been formed between TZ and TPB. **Figure 7(a-e)** shows the IR spectra of the free ligands as well as the ion associate.

Determination of TZ in pure form and pharmaceutical preparations

The proposed method was applied for the determination of TZ in the pharmaceutical formulation, Trittico® tablets. Satisfactory results were obtained in good agreement with the label claim, and the results of the standard addition technique indicate no interference from excipients and additives **Table 3**. The result was given in **Table 4** shows that the proposed method is satisfactorily accurate and precise. The accuracy and reproducibility with respect to the official[2] method were assessed by performing student's t and F tests, respectively.

CONCLUSION

The conductometric methods are characterized by low cost and simplicity, conductometric titrations are especially useful for very dilute solutions as the percentage change in conductance is independent of concentration and measurements need not be made close to the equivalence point. The proposed methods are simple, rapid and inexpensive. So, it is a good alternative to the other reported methods and to the high-cost HPLC methods.

Conflict of Interest

The authors declare that they don't have any conflict of interest.

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