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Two spectrophotometric methods for the determination of azithromycin and roxithromycin in pharmaceutical preparations

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ABSTRACT

Two new and simple spectrophotometric procedures have been proposed and validated for estimation of two important macrolide antibiotics namely, azithromycin dihydrate and roxithromycin. Method I depends on complex formation between any of the two drugs and copper in acidic medium where the absorbances of the produced complexes are measured at 250 and 264 nm with linearity ranges of 1.0-100.0 and 2.0-130.0 µg/mL for the two drugs, respectively. Method II depends on the reaction of these drugs with *N*-bromosuccinimide forming a product which is yellow colored, measured at 264 and 278 nm, with linearity ranges of 2.0-140.0 and 3.0-160.0 µg/mL for azithromycin dihydrate and roxithromycin, respectively. The proposed methods were subjected to detailed validation procedure; moreover they were used for the estimation of the concerned drugs in their different dosage forms. Study of the reactions stoichiometry was carried out; furthermore, a reaction mechanism proposal was presented.

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1. Introduction

Azithromycin dihydrate (AZT) is a nitrogen-containing macrolide. It is chemically known as (2*R*,3*S*,4*R*,5*R*,8*R*,10*R*,11*R*, 12*S*, 13*S*, 14*R*)-13-[(2, 6-dideoxy-3-*C*-methyl-3-*o*-methyl-a-L-*ribo*-hexopyranosyl)oxy]-2-ethyl-3, 4,10-trihydroxy-3,5,6,8,10, 12, 14-heptamethyl-11-[[3,4,6-trideoxy-3-(dimethylamino)-b-D-*xylo*-hexopyranosyl]oxy] -1-oxa-6-azacyclopentadecan-15-one [1] (Figure 1 (a)). It is used for treating respiratory-tract, soft-tissue and skin infections. It is also used in case of typhoid and trachoma [2].

Roxithromycin (ROX) is a macrolide antibiotic obtained from erythromycin. It is designated chemically as (3*R*,4*S*,5*S*,6*R*, 7*R*,9*R*,11*S*,12*R*,13*S*,14*R*)-4-[(2, 6-dideoxy-3-*C*-methyl-3-*o*-methyl-a-L-*ribo*-hexopyranosyl)oxy]-14-ethyl-7,12, 13-trihydroxy-10-[(*E*)-[(2-methoxyethoxy)methoxy]imino]-3, 5, 7, 9, 11, 13-hexamethyl-6-[[3, 4, 6-trideoxy-3-(dimethylamino)-b-D-*xylo*-hexopyranosyl]oxy]oxacyclotetradecan-2-one (Erythromycin 9-(*E*)-[*O*-[(2-methoxyethoxy)methyl]oxime]) [1] (Figure 1 (b)). Its uses include the treatment *Legionella* infections. When combined with neomycin, it is utilized for the counteractive action of surgical infection in patients having gut surgery [2].

Literature survey revealed different methods for estimation of AZT or ROX whether alone or in combination with other drugs. As for AZT, it has been determined by high performance liquid chromatography (HPLC) [3-6], liquid chromatography-mass detection (LC-MS/MS) [7,8], ultra-performance liquid chromatography (UPLC) [9], micellar liquid chromatography (MLC) [10], spectroflourimetry [11] and UV-Visible spectrophotometry [12-17] while ROX has been estimated by HPLC [18,19], LC-MS/MS [20], spectroflourimetry [21,22] and UV-Visible spectrophotometry [23].

There is also a spectrophotometric method for estimation of AZT and ROX that was performed in our laboratory for their estimation in pharmaceutical preparations [24]. Till now, no methods have been reported for spectrophotometric estimation by using metal complexation or *N*-bromosuccinimide for any of the two drugs which encouraged us to develop these methods. Spectrophotometry is a very important technique in drug analysis due to its availability in most laboratories, simplicity and low cost. The developed methods have simple procedures and wider determination ranges, also they are rapid and convenient in comparison with the reported spectrophotometric methods [12-14,23,24], so can be applied in quality control laboratories for routine analysis.

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Table 1. Assay parameters for determination of AZT and ROX by Method I and II.

Method	Assay parameters	AZT	ROX
I	Volume of metal ion	2 mL	3 mL
	Buffer PH	6 (borate buffer)	5.5 (acetate buffer)
	Volume of Buffer solution	2 mL	1.5 mL
	λ_{max}	250 nm	264 nm
II	Volume of NBS	0.5 mL	1 mL
	Heating Temperature	40 °C	50 °C
	Heating Time	20 min.	20 min.
	Diluting Solvent	Methanol	Methanol
	λ_{max}	264 nm	278 nm

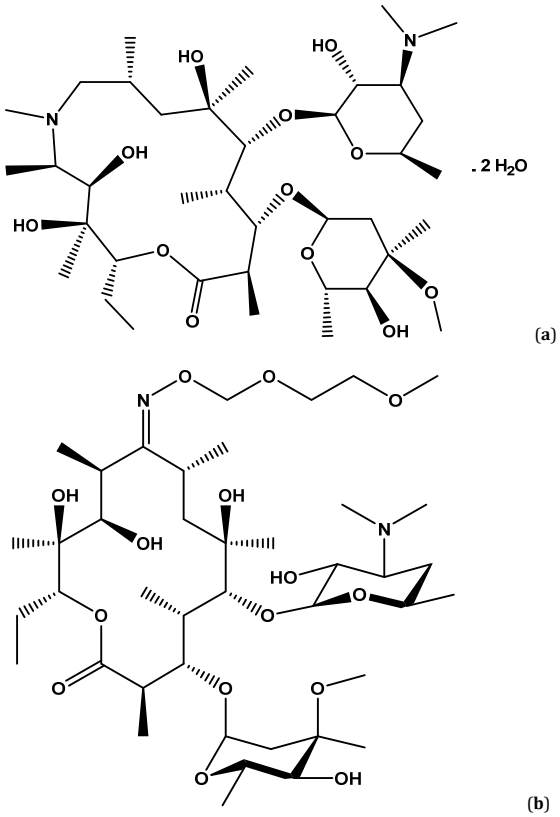


Figure 1. Structural formula of (**a**) AZT and (**b**) ROX.

2. Experimental

2.1. Instrumentations

A double-beam Shimadzu UV-Vis spectrophotometer (Kyoto, Japan) model UV 1601 PC (1.0 cm quartz cells) was used for all absorbance measurements. A Jenway 3503 digital pH meter (Stone, Staffs, UK) was used for adjustment of the pH.

2.2. Materials

Azithromycin dihydrate (Batch No.1B07007) sample was obtained from Amoun Pharmaceutical Co., El-Obour City, Cairo, Egypt. It is certified to have a potency of 100.8%. Roxithromycin (Batch No. 011006B) sample was supplied by Alkan Pharma Co., 6th of October City, Egypt. It is certified to have a potency of 98%. Xithrone® tablets contain 500 mg of Azithromycin/tablet (Batch No. 153132) products of Amoun Pharma Co., Cairo, Egypt. Roxicin® tablets contain 150 mg of ROX/tablet (Batch No. 101124) products of El-Obour Modern Pharmaceutical industries Co., Cairo, Egypt. Zithrocan® capsules labeled to contain 524.06 mg of Azithromycin dihydrate/tablet (Batch No. 131) produced by Hikma Pharma Co., 6th of October City, Egypt. Xithrone® suspension contains 200 mg Azithromycin/5 mL, (Batch No. 155359), products of Amoun Pharma Co., Cairo, Egypt. All dosage forms were obtained from municipal pharmacy.

2.3. Reagents and Chemicals

All reagents and chemicals were of analytical grade, solvents were of spectroscopic grade and the water utilized in the study was distilled. Aqueous solution of copper sulphate ($CuSO_4 \cdot 5H_2O$), obtained from El Nasr Co. (ADWIC, Cairo, Egypt) of concentration (5×10^{-3} M), acetate buffer solution (0.2 M) was obtained by adjusting the pH of sodium acetate trihydrate (0.2 M) with acetic acid (0.2 M) [25], El Nasr Co. (ADWIC, Cairo, Egypt), borate buffer solution (0.2 M) was obtained by adjusting the pH of boric acid (0.2 M) with NaOH (0.2 M), El Nasr Co. (ADWIC, Cairo, Egypt) and *N*-bromosuccinimide (NBS) with purity 98%, (0.056 M) was freshly prepared in distilled water, Sisco Research Laboratories, Mumbai, India.

2.4. Standard solution

Stock solutions of AZT and ROX containing 200.0 µg/mL were obtained by dissolving 20.0 mg of the pure drug in 100 mL of methanol. With the same solvent, further dilution was done to get the required concentration. The solution was stable for at least 7 days when kept in the refrigerator.

2.5. General procedures

2.5.1. Construction of calibration graphs

2.5.1.1. Method I

Accurate volumes of AZT and ROX standard solutions were measured and transferred into a series of 10.0 mL volumetric flasks. Borate or acetate buffer with optimum pH were added to each flask with their specified volumes then the specified volume of copper solution was added (Table 1). Distilled water was used to complete solutions to the mark and thoroughly mixed. The absorbance of each solution was measured at the specific λ_{max} (Table 1) against a reagent blank. The absorbance values were plotted against drug concentrations (µg/mL) to get the calibration plots; alternatively the corresponding regression equations were derived.

2.5.1.2. Method II

Accurate volumes of AZT and ROX standard solutions were measured and transferred into a series of test tubes. The specified volumes of NBS were added to each test tube (Table 1) then heated in water bath using the specified time and temperature mentioned in Table 1. After cooling, the solutions were quantitatively transferred into a series of 10.0 mL volumetric flasks and completed to the mark with methanol. The solutions were thoroughly mixed. The absorbance of the produced colored product was measured at the specific λ_{max} (Table 1) against blank solution. The calibration graph was obtained by same steps as Method I.

Table 2. Analytical performance data for the proposed methods.

Parameter	Method IA	Method IB	Method IIA	Method IIB
Concentration range (µg/mL)	1.0 -100	2.0-130	2.0-140	3.0-160
Limit of detection LOD (µg/mL)	0.7604	0.9050	0.6938	1.4696
Limit of Quantitation LOQ (µg/mL)	2.3043	2.7425	2.1026	4.4533
Regression equation * y=a+bx	$y = 0.0078x + 0.0859$	$y = 0.0047x + 0.1268$	$y = 0.0106x + 0.093$	$y = 0.0083x + 0.0317$
Correlation coefficient	0.9999	0.9999	0.9998	0.9999
($S_{y/x}$)	0.0032	0.0024	0.0041	0.0061
(S_a)	0.0018	0.0013	0.0022	0.0037
(S_b)	0.0001	0.0000	0.0001	0.0001
%RSD	0.553	0.697	0.596	0.504
%Error (%RSD/√n)	0.176	0.220	0.188	0.159
$A^{\%}$ ($dl.g^{-1}.cm^{-1}$)	78.37	46.99	105.76	82.55
Molar absorptivity ($L.mol.^{-1}cm^{-1}$)	6152.19	3933.21	8302.49	6910.10

* *y*: absorbance; *a*: intercept; *x*: Concentration (µg/mL); *b*: slope.

2.5.2. Procedure for tablets

Ten tablets were accurately weighed, finely pulverized and thoroughly mixed. An accurately weighed amount of the powdered tablets equivalent to 20.0 mg of AZT or ROX was transferred into a 100.0 mL conical flask and about 40 mL of methanol were added. The contents of the flask were sonicated for 30 minutes, filtered into 100 mL volumetric flask and completed to the mark with the same solvent. Different aliquots of the filtrate were accurately transferred into 10.0 mL volumetric flasks. Then, proceed as described under "Construction of calibration graphs for Method I and II". From the corresponding regression equations, the nominal contents of tablets were determined.

2.5.3. Procedure for capsules

The content of 10 capsules were weighed and thoroughly mixed. A weighed quantity of the powder equivalent to 20.0 mg AZT was transferred into a 100.0 mL volumetric flask and the volume was made up to the mark with methanol. The contents of the flask were sonicated for 30 minutes, filtered and different volumes of the filtrate were quantitatively transferred into 10.0 mL volumetric flask. The procedure was followed as described under "Construction of the calibration graph for Method I and II". From the corresponding regression equations, the nominal contents of the capsules were calculated.

2.5.4. Procedure for suspension

An accurate volume of the freshly reconstituted oral suspension equivalent to 20.0 mg of AZT was extracted with about 25 mL of methanol, sonicated for about 45 min, left for a time in a refrigerator to allow any insoluble matter to settle down then filtered into a 100.0 mL volumetric flask. The solution was then completed to volume with methanol. This extract was further diluted with methanol. The procedure was completed as mentioned for preparing the calibration plots. From the corresponding regression equation, the nominal contents of the suspension were determined.

3. Results and discussion

3.1. Method I

It was found that the binary complexes of AZT or ROX with metal ions have not been studied yet. This encouraged us to develop this method. As they may have an effect on bio-availability of AZT and ROX as some metal ions are present in relatively appreciable concentration in biological fluids. The absorption spectra of AZT in borate buffer and ROX in acetate buffer of optimum pH value exhibits maximum absorbance at 219 and 198 nm, respectively. Upon complexation with Cu^{2+} ions, the maximum absorbances were obtained at 250 and 264 nm for AZT and ROX, respectively, as illustrated in Figure 2 and 3.

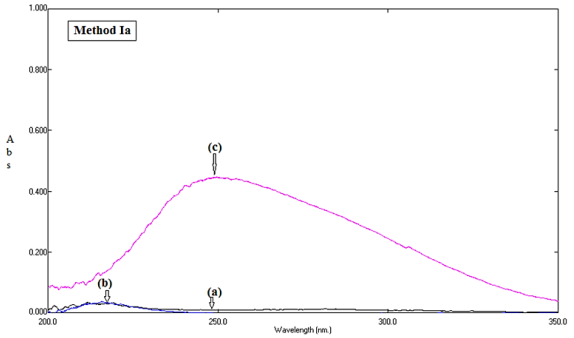


Figure 2. Absorption spectra of (a) AZT (30.0 µg/mL) in methanol, (b) AZT (30.0 µg/mL) in borate buffer of pH 6 and (c) AZT (30.0 µg/mL) complex with copper in borate buffer of pH 6.

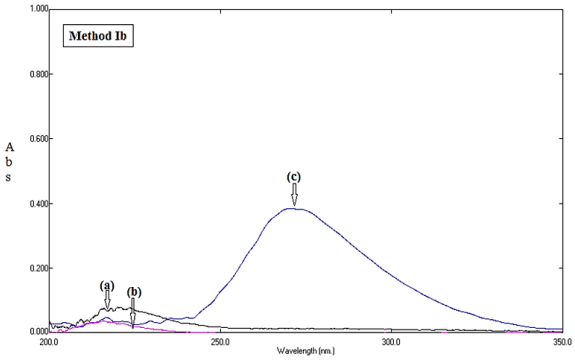


Figure 3. Absorption spectra of (a) ROX (30.0 µg/mL) in methanol, (b) ROX (30.0 µg/mL) in acetate buffer of pH 5.5, and (c) ROX (30.0 µg/mL) complex with copper in acetate buffer of pH 5.5.

3.1.1. Optimization of experimental conditions

It was found that AZT and ROX form stable complexes with Cu^{2+}. The experimental conditions influencing the formation of complexes and their stability were studied and optimized.

3.1.2. pH

The influence of pH was observed over the pH range of 2.5-10.0. It was found that optimum pH is 6.0 for AZT, and 5.5 for ROX.

3.1.3. Metal ion concentration

The influence of copper ion concentration was studied using increasing volumes of Cu^{2+} solution (5×10^{-3} M). It was

found that the optimum volumes of Cu^{2+} are 2 and 3 mL for AZT and ROX, respectively.

3.1.4. Buffer volume

The effect of the buffer volume was also observed. It was observed that 2.0 mL of the borate buffer and 1.5 mL of acetate buffer was used for maximum absorbance of AZT and ROX-copper complexes, respectively.

3.1.5. Temperature

The influence of temperature was investigated. The maximum absorbance was obtained at room temperature and decreased by increasing temperature. It was also found that the complex is formed instantaneously and is stable for at least two hours.

3.1.6. Diluting solvents

The effect of diluting solvents on the absorbance of the produced complexes was studied including distilled water, methanol, acetonitrile, dimethylformamide and butanol. Distilled water was found to be the best diluting solvent giving the maximum absorbance, which adds another advantage to the proposed method.

3.1.7. Order of addition

The effect of order of addition of buffer solution and copper was also studied. It was found that the maximum absorbance was obtained by addition of buffer solution before copper for both drugs.

3.2. Method II

N-Bromosuccinimide is a very useful and important reagent [26]. It has a strong brominating ability [27]. It has been reported that tertiary amine reacts with *N*-bromo-succinimide to produce colored intermediate, which on hydrolysis gives aldehydes and brominated secondary amine whereas *N*-bromosuccinimide is irreversibly reduced to succinimide [28]. Since both AZT and ROX contain tertiary amino groups, they are susceptible to react with NBS yielding a colored product with λ_{max} of 264 and 278 nm for AZT and ROX, respectively, as presented in Figure 4-5 [29].

3.2.1. Optimization of experimental conditions

The optimum conditions for the proposed methods have been studied and optimized including: concentration of NBS, heating temperature, heating time and nature of diluting solvent (Table 1).

3.2.2. Concentration of NBS

The impact of NBS concentration on the color development was studied using increasing volumes of NBS solution (0.056 M) from 0.2 to 3.0 mL. The highest absorbance was obtained with 0.5 mL for AZT and 1.0 mL for ROX that remained unchanged with increasing the amount of NBS.

3.2.3. Heating temperature

The influence of heating temperature on the colored product absorbance was observed from room temperature up to boiling temperature. It was observed that 40 and 50 °C were the optimum temperature giving the maximum absorbance for AZT and ROX, respectively.

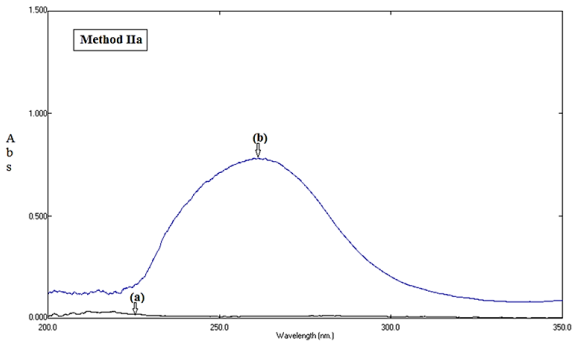


Figure 4. Absorption spectra of (a) AZT (60.0 µg/mL) in methanol, (b) Reaction product of AZT (60.0 µg/mL) with NBS.

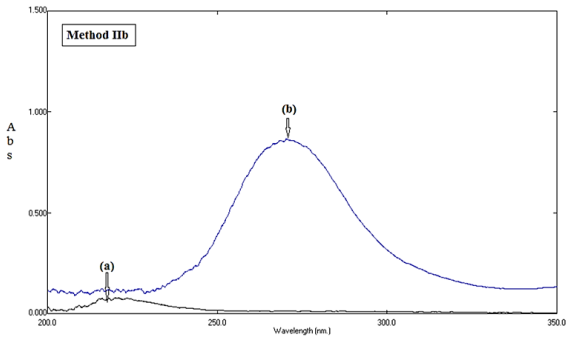


Figure 5. Absorption spectra of (a) ROX (60.0 µg/mL) in methanol and (b) Reaction product of ROX (60.0 µg/mL) with NBS.

3.2.4. Heating time

The influence of heating time was also observed in from 10-60 minutes. The maximum absorbance was obtained after 20 minutes of heating at optimum temperature in case of both drugs.

3.2.5. Diluting solvents

The diluting solvents mentioned in Method I was also studied to select the appropriate one. Methanol is the best diluting solvent giving the maximum absorbance values.

3.3. Methods validation

The proposed procedures were validated according to ICHQ2 (R1) recommendations [30], including the following validation parameters.

3.4. Linearity and range

The calibration plots obtained by plotting the absorbance *versus* the final concentrations (µg/mL) were found to be linear over the concentration ranges presented in Table 2. The validity of the developed methods were proven by statistical regression line [31] and it was found that percentage relative standard deviation (%RSD) was small while correlation coefficient values (r) approach unity (Table 2) indicating the good linearity of calibration plots.

3.5. LOQ and LOD

LOQ and LOD were determined using the following equations [30]:

$$LOQ = 10 \times S_a/b \quad (1)$$

Table 3. Application of the proposed methods to the determination of AZT and ROX in pure form.

Parameter	Method IA		Method IB		Method IIA		Method IIB		Comparison method [24]	
	Conc. taken (µg/mL)	% Found [a]	Conc. taken (µg/mL)	% Found [a]	Conc. taken (µg/mL)	% Found [a]	Conc. taken (µg/mL)	% Found [a]	% Found [a] for ROX	% Found [a] for AZT
	1.0	100.00	2.0	99.05	2.0	99.06	3.0	99.60	100.4	99.66
	5.0	100.13	5.0	99.00	5.0	100.94	5.0	99.42	99.35	99.94
	10.0	99.42	10.0	99.15	10.0	100.47	10.0	100.80	100.5	100.42
	20.0	100.04	20.0	99.04	20.0	99.53	20.0	99.06	100.0	99.80
	40.0	100.67	40.0	100.43	40.0	99.43	40.0	99.14		
	50.0	101.05	80.0	100.59	60.0	99.84	60.0	99.19		
	60.0	101.30	100.0	100.68	80.0	99.29	100.0	99.23		
	80.0	100.02	120.0	99.86	100.0	99.72	120.0	99.68		
	90.0	100.30	130.0	99.27	120.0	100.39	160.0	99.34		
	100.0	100.53			140.0	99.53				
Mean±SD		100.35±0.56		99.72±0.70		99.82±0.60		99.51±0.50	100.05±0.51	99.96±0.33
t	0.0519(2.262) [b]		0.7034(2.306) [b]		0.383(2.262) [b]		0.207(2.306) [b]			
F	2.879(8.81) [b]		1.883(8.85) [b]		3.305(8.81) [b]		1.0404(8.85) [b]			

[a] Each result is the average of three separate determinations.
[b] Values between brackets are the tabulated t and F values, at $p = 0.05$ [31].

Table 4. Precision data of the proposed methods for the determination of AZT and ROX in pure form.

Method	Intra-day precision				Inter-day precision			
	Conc. taken (µg/mL)	Mean |%found|±SD	%RSD	%Error	Conc. taken (µg/mL)	Mean |%found|±SD	%RSD	%Error
IA	40.0	100.41±0.51	0.50	0.29	40.0	100.01±0.46	0.46	0.27
	60.0	100.48±0.62	0.61	0.31	60.0	100.16±0.55	0.55	0.32
	80.0	100.38±0.87	0.87	0.50	80.0	100.19±0.44	0.44	0.25
IB	40.0	100.07±0.18	0.18	0.10	40.0	99.46±0.55	0.55	0.32
	60.0	99.91±0.69	0.69	0.40	60.0	100.01±0.19	0.19	0.11
	80.0	99.91±0.18	0.18	0.10	80.0	99.99±0.34	0.34	0.20
IIA	40.0	100.06±0.34	0.34	0.20	40.0	100.18±0.25	0.25	0.15
	60.0	100.46±0.80	0.80	0.31	60.0	99.96±0.27	0.27	0.15
	80.0	100.5±0.71	0.71	0.41	80.0	100.30±0.50	0.50	0.29
IIB	60.0	100.46±0.43	0.43	0.31	60.0	100.04±0.33	0.33	0.19
	80.0	99.80±0.80	0.80	0.46	80.0	99.69±0.60	0.60	0.35
	100.0	99.85±0.53	0.53	0.31	100.0	100.30±0.62	0.62	0.36

$$LOD = 3.3 \times S_a/b \quad (2)$$

These data are outlined in Table 2.

3.6. Accuracy

Statistical evaluation of the developed method using student *t*-test and the variance ratio *F*-test [31] demonstrated no significance differences between the performance of the proposed and comparison methods [24] (Table 3). It is to be mentioned that the comparison method [24] depends on a binary complex formation between the studied drugs and eosin Y in aqueous buffered medium.

3.7. Precision

Through evaluating three concentrations of AZT and ROX on three sequential time intervals, intraday precision was determined. Furthermore, repeated analysis of three concentrations for three successive days, allowed interday precision to be demonstrated. Intraday and interday precision values are outlined in Table 4. The very small values of RSD demonstrate the high repeatability and intermediate precision of the developed procedures.

3.8. Robustness

It was established by the consistency of the absorbance with the deliberately small changes in the different experimental conditions. For Method I, these changes include: buffer volume (optimum volume±0.5), pH (optimum pH±0.2), metal ion volume (optimum volume±0.2). While in case of Method II, these minor changes include: volume of NBS (optimum volume±0.2), heating temperature (optimum temperature±2 °C), time of heating (optimum time±2 minutes).These small changes didn't influence the absorbance value obtained by the developed methods, demonstrating the robustness of these methods.

3.9. Specificity

It was demonstrated by investigation of any interference from the common tablet, capsule and suspension excipients. No interference was produced during the application of the developed procedures.

3.10. Pharmaceutical applications

The developed procedures were used to estimate the drugs in their dosage forms. The results obtained were statistically compared with those of a reference method [24]. No significant difference was found between the developed and comparison method regarding *t* and *F* values as shown in Table 5.

3.11. Reactions stoichiometry and mechanism

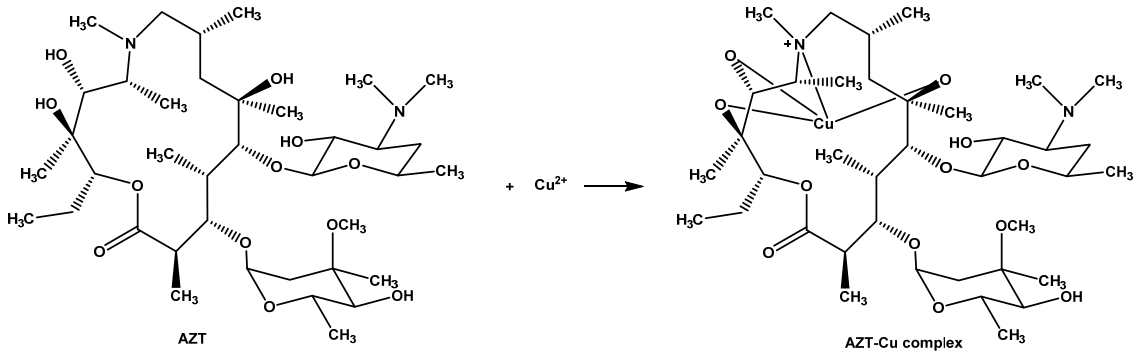
Limiting logarithmic method [32] was used to study the reaction stoichiometry between AZT or ROX and copper or NBS. For Method I, curves of log absorbance *versus* either log[Cu^{2+}] or log[Drug] resulted in straight lines with slopes of 0.7240/0.9384 for AZT and 0.7048/ 0.7349 for ROX (Figure 6 and 7). So, it is demonstrated that the reaction take place in 1:1 ratio. A reaction mechanism proposal depending on the molar reactivity between AZT or ROX and copper is presented in Scheme 1 and 2. For method II, a similar approach was followed where the slopes were 0.7258/0.8102 for AZT and 0.91964/ 1.1949 for ROX (Figure 8 and 9) indicating that the reaction takes place in 1:1 ratio. The reaction mechanism proposal presented in Scheme 3.

Table 5. Application of the proposed and comparison methods to the determination of AZT and ROX in different dosage forms.

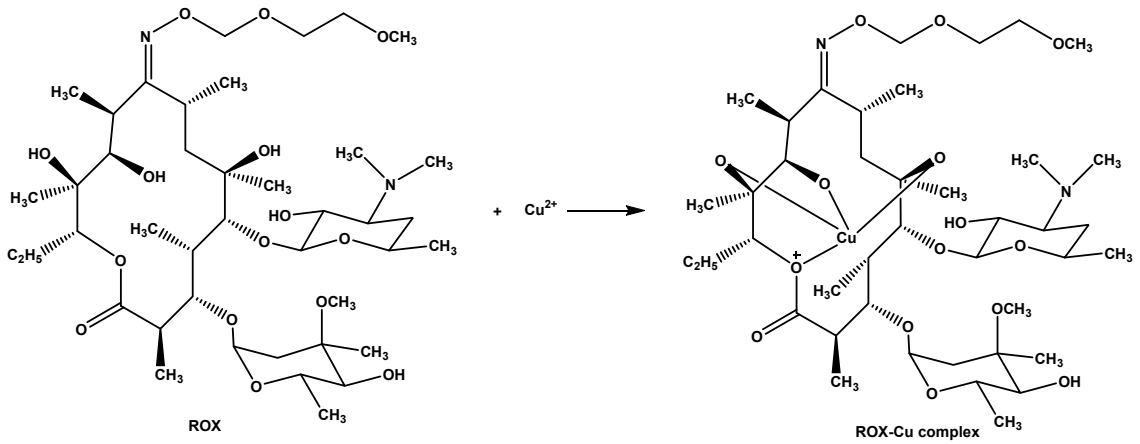
Pharmaceutical Preparation	Conc. taken (µg/mL)	Method I % Found [a]	Method II % Found [a]	Comparison method [24] % Found [a]
Xithrone tablets	20.0	100.00	100.18	99.04
(500 mg AZT/tablet)	40.0	98.19	99.40	99.31
	60.0	102.32	100.09	101.77
	80.0	100.36	101.50	99.17
	100.0	99.71	99.40	-
Mean±SD		100.12±1.48	100.11±0.86	99.82±1.30
t		0.369(2.776) [b]	0.598(2.776) [b]	-
F		1.688(9.12) [b]	2.206(9.12) [b]	-
Roxicin tablets	20.0	99.77	100.98	99.34
(150 mg ROX/tablet)	40.0	100.11	98.27	101.51
	60.0	101.59	100.23	99.12
	80.0	99.26	98.94	100.12
	100.0	100.59	100.07	-
Mean±SD		100.26±0.89	99.70±1.08	100.02±1.08
t		0.217(2.776) [b]	0.511(2.776) [b]	-
F		1.165(9.12) [b]	1.286(9.12) [b]	-
Zithrocan capsules	20.0	99.86	99.69	101.49
(500 mg AZT/capsule)	40.0	99.21	100.41	99.31
	60.0	100.27	99.23	99.44
	80.0	98.77	99.14	100.40
	100.0	99.88	100.10	-
Mean±SD		99.60±0.60	99.71±0.55	100.16±1.01
t		1.044(2.776) [b]	0.93(2.776) [b]	-
F		2.295(9.12) [b]	3.033(9.12) [b]	-
Xithrone suspension	20.0	99.67	99.90	99.35
(200 mg AZT/5 mL)	40.0	100.66	98.91	99.79
	60.0	99.16	99.69	100.92
	80.0	98.96	100.16	99.57
	100.0	100.15	99.29	-
Mean±SD		99.72±0.70	99.59±0.50	99.91±0.70
t		0.571(2.776) [b]	0.55(2.776) [b]	-
F		1.183(9.12) [b]	1.681(9.12) [b]	

[a] Each result is the average of three separate determinations.

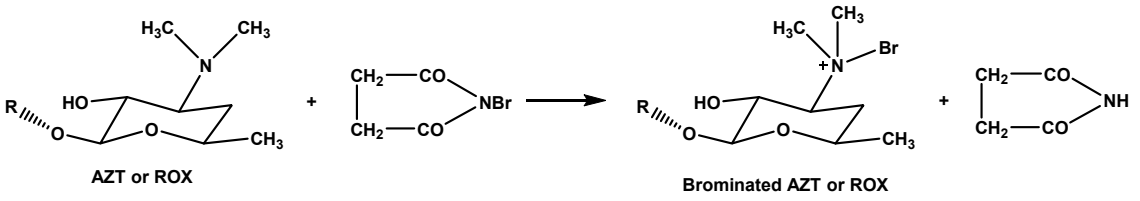
[b] Values between brackets are the tabulated t and F values, at $p = 0.05$ [31].



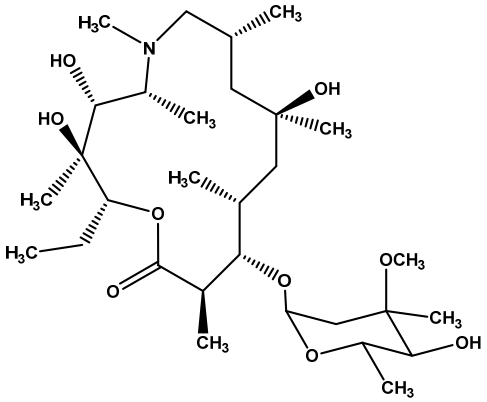
Scheme 1



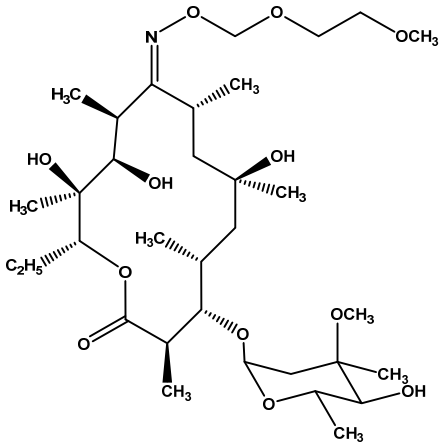
Scheme 2



R for AZT =



R for ROX =



Scheme 3

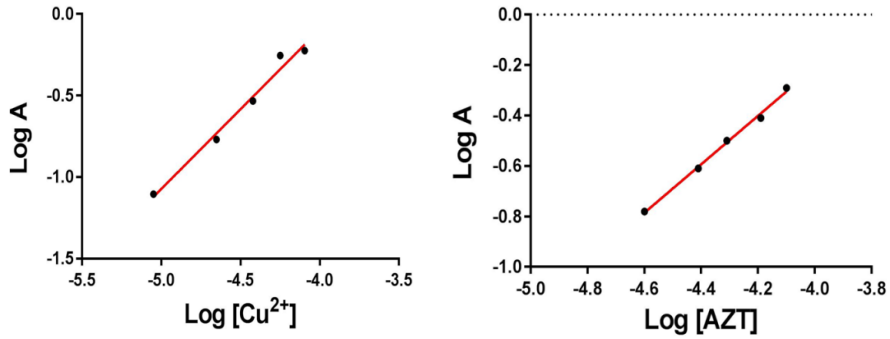


Figure 6. Limiting logarithmic plots for the molar reactivity of AZT with Cu^{2+}.

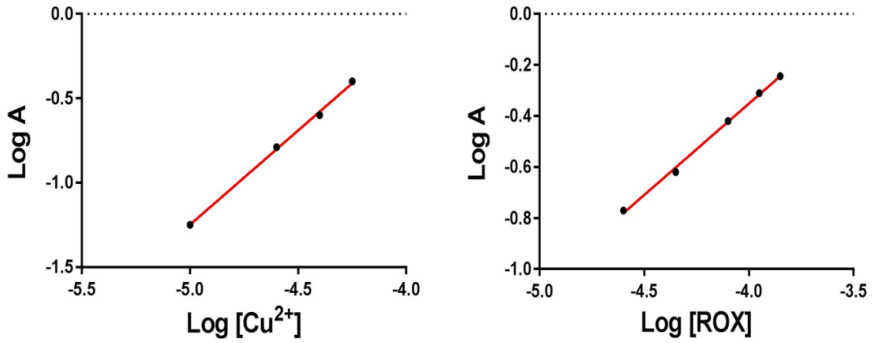


Figure 7. Limiting logarithmic plots for the molar reactivity of ROX with Cu^{2+}.

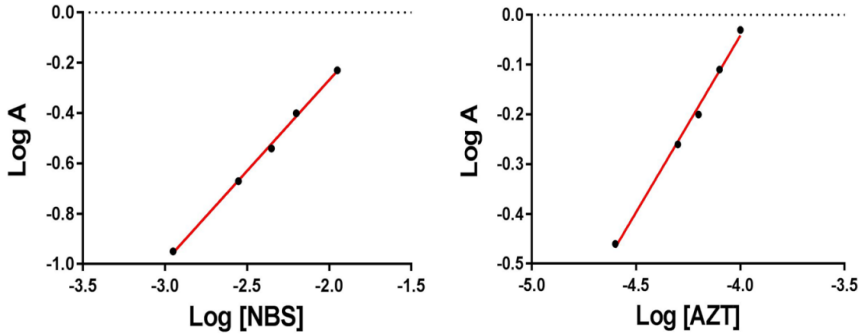


Figure 8. Limiting logarithmic plots for the molar reactivity of AZT with NBS.

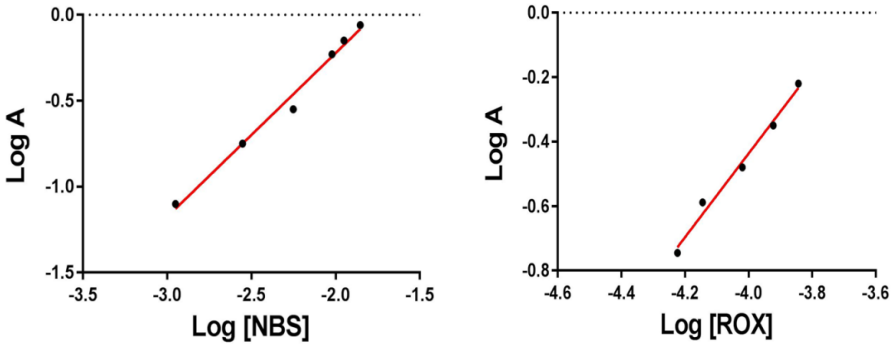


Figure 9. Limiting logarithmic plots for the molar reactivity of ROX with NBS.

4. Conclusion

Two simple, sensitive, specific and inexpensive spectrophotometric procedures are proposed for estimation of AZT and ROX in their dosage forms with no interference from common excipients. Furthermore, the developed procedures are rapid and do not need complexed requirements needed in chromatographic methods. In addition, they offer reproducible results and have the advantage of being simple and convenient. So, they can be used in quality control laboratories for routine analysis.

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