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Quantitation of Ranitidine in Pharmaceuticals by Titrimetry and Spectrophotometry Using Potassium Dichromate as the Oxidimetric Reagent

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Four new methods are described for the determination of ranitidine hydrochloride (RNH) in bulk drug and in formulations employing titrimetric and spectrophotometric techniques and using potassium dichromate as the oxidimetric reagent. In titrimetry (method A), RNH is treated with a measured excess of dichromate in acid medium, and the unreacted oxidant is back titrated with iron(II) ammonium sulfate. The three spectrophotometric methods are also based on the oxidation of RNH by a known excess of dichromate under acidic conditions followed by the determination of surplus oxidant by three different reaction schemes. In one procedure (method B), the residual dichromate is treated with diphenylcarbazide and the absorbance measured at 540 nm. Calculated amount of iron(II) is added to residual dichromate and the resulting iron(III) is complexed with thiocyanate and measured at 470 nm (method C). Method D involves reduction of unreacted dichromate by a calculated amount of iron(II) and estimation of residual iron(II) as its orthophenanthroline complex after raising the pH, and measuring the absorbance at 510 nm. In all the methods, the amount of dichromate reacted corresponds to the drug content. The experimental conditions are optimized. The titrimetric procedure is applicable over 5-10 mg range. In spectrophotometric methods, Beer's law is obeyed in the ranges 5-50, 5-80, and 10-100 μg ml^{-1} for method B, method C, and method D, respectively. The methods were validated for accuracy, precision and recovery. The proposed methods were applied to the analysis of RNH in the tablet and the injection forms, and the results were in agreement with those obtained by the reference method.

Keywords: Ranitidine, Determination, Titrimetry, Spectrophotometry, Dichromate, Formulations

INTRODUCTION

Ranitidine hydrochloride (RNH), chemically is N,N-dimethyl-5-[2-(1-methylamine-2-nitrovinyl)-ethylthiomethyl] furfurylamine hydrochloride (Fig. 1). It is a H_2-receptor antagonist and is widely used in short term treatment of duodenal ulcer and in the management of hypersecretory conditions [1]. Several techniques such as proton magnetic resonance spectroscopy [2], near infrared reflectance spectrometry [3], scintillation proximity assay [4], flow injection fluorimetry [5], polarography [6,7], differential pulse polarography [8], capillary electrophoresis [9], liquid chromatography [10], and high performance liquid chromatography [11-15], have been used for the determination of RNH in pharmaceuticals. These techniques require sophisticated instruments and expensive reagents, and involve

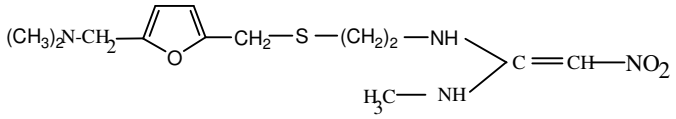

Fig. 1. Structure of drug.

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Table 1. Comparison of the Existing Spectrophotometric Methods with the Proposed Methods for RNH

Ref.	Reagents employed	Linear range ($\mu g\ ml^{-1}$)	ϵ ($l\ mol^{-1}\ cm^{-1}$)	Remarks
[17]	F-C reagent	40-240	-	Less sensitive
[18]	$KMnO_4$/NBS-azine dyes	5-30	5.2×10^3	Involves extraction, uses unstable reagent and has narrow range of linear response
		0.5-4.0	1.8×10^4	
		0.4-2.8	4.2×10^4	
		0.4-2.8	7.2×10^4	
[19]	Iron(III)-MBTH	5-18	-	30 min contact time, uses expensive reagent
[20]	Iron(III)-MBTH	0.4-6.0	-	-do-
[21]	DCBC	10-50	-	Involves boiling for 20 min
[22]	$NaNO_2$	0.3-12 $mg\ ml^{-1}$	-	Involves flow injection automated assembly, least sensitive
[23]	Rose bengal	2-12	-	Involves extraction
[24]	Bromothymol blue	1-20	-	Involves extraction
Present methods	$K_2Cr_2O_7$-diphenylcarbazide	5-50	3.4×10^4	Use stable oxidant, employ non-stringent working conditions, shorter contact time, no heating or extraction step, more sensitive
	$K_2Cr_2O_7$-iron(II) thiocyanate	5-80	2.3×10^4	
	$K_2Cr_2O_7$-iron(II) orthophenanthroline	10-100	1.2×10^4	

F.C. Folin-Ciocalteau; NBS. N-bromosuccinimide; MBTH 3-methyl-2-benzothiazolinone hydrazone; DCBC. 3,5-Dichloro-p-benzoquinone chlorimide.

several manipulation steps and derivatization reactions.

Literature survey revealed that the only titrimetric method [16] reported for RNH requires 300 mg of drug for each titration. There are several reports of the determination of RNH by spectrophotometry involving the use of Folin-Ciocalteu reagent [17], azine dyes [18], 3-methyl-2-benzothiazoline hydrazone-iron(III) [19,20], 3,5-dichlorop-benzoquinone chlorimine [21], sodium nitrite [22], rose bengal [23] and bromothymol blue [24]. These methods are based on redox, coupling, charge-transfer complexation, nitrosation and ion-pair complexation reactions. However, the reported spectrophotometric methods suffer from one or more disadvantage including poor sensitivity, a complicated and time-consuming procedure, extraction step or the need for expensive or undesirable chemicals (Table 1).

In this paper, we demonstrate the use of titrimetric and spectrophotometric methods, which circumvent the problems encountered in the methods previously reported. The four methods are based on the oxidation of RNH by a known excess of dichromate followed by the determination of unreacted oxidant by back titration with iron(II) ammonium sulfate and by spectrophotometry *via* different reaction schemes. The methods yielded accurate, rapid and reproducible results for four different commercial products with two different dosages of tablets and one dosage of injections. The results obtained by the proposed methods were compared with those obtained by the reference method.

EXPERIMENTAL

Apparatus

A Systronics model 106 digital spectrophotometer

equipped with 1 cm quartz cells was used for all absorbance measurements.

Reagents and Materials

All chemicals were of analytical-reagent grade and solutions were made in distilled water.

A 0.01 M potassium dichromate solution was prepared by dissolving 2.94 g of chemical (S. d. Fine Chem. Ltd., Mumbai, India) in water and diluting to 1 l in a calibrated flask, and used for titrimetric work. For spectrophotometric investigations, a stock solution equivalent to 1000 µg ml^{-1} $K_2Cr_2O_7$ was prepared by dissolving 100 mg of chemical in water and diluting to 100 ml in a calibrated flask. Working concentrations of 30, 50, and 90 µg ml^{-1} $K_2Cr_2O_7$ were prepared by appropriate dilution of stock solution with water and used in the methods B, C, and D, respectively.

Iron(II) ammonium sulfate solution (0.06 M) was prepared by dissolving about 12 g of chemical (S. d. Fine Chem. Ltd., Mumbai) in 500 ml of water containing 2 ml of 2 M H_2SO_4, and used in titrimetry. This was standardized with pure potassium dichromate [25] and diluted to get 400 µg ml^{-1} and 700 µg ml^{-1} solutions for use in spectrophotometric methods C and D, respectively. A 0.25% diphenylcarbazide solution was prepared by dissolving 0.25 g of reagent (BDH Chemicals Ltd., Poole, England) in 100 ml of 1:1 acetone:water.

Potassium thiocyanate (2 M) was prepared by dissolving about 19.44 g of chemical (S. d. Fine Chem. Ltd., Mumbai) in 100 ml of water. Orthophenanthroline (0.25%) solution was prepared by dissolving 0.25 g of reagent (S. d. Fine Chem. Ltd., Mumbai) in 100 ml of water with the aid of heat. Barium diphenylaminesulphonate indicator (1%) was prepared by dissolving 100 mg of indicator (BDH Chemicals Ltd., Poole, England) in 10 ml of water with the aid of heat. Sulfuric acid (5 M) was prepared by adding 139 ml of concentrated acid (S. d. Fine Chem. Mumbai, India, sp. gr. 1.86) to 361 ml of water with cooling, for use in methods A and D. This was diluted appropriately with water to 1 M acid for use in method B. A 5 M Hydrochloric acid was prepared by diluting 111 ml of concentrated acid (S. d. Fine Chem. Ltd., Mumbai, sp. gr. 1.18) to 250 ml with water and used in method C. Ammonia (1:1) was prepared by diluting 50 ml of strong ammonia (Qualigens Fine Chemicals, Glaxo India Ltd., Mumbai, India, 25%) with 50 ml of water for use in method D. Syrupy phosphoric acid acid (S. d. Fine Chem. Ltd., Mumbai, India) was used in method A.

Pharmaceutical grade RNH which is reported to be 99.8% pure was procured from Glaxo SmithKline Pharmaceuticals Ltd., Nasik, India. To prepare a stock solution containing 1 mg ml^{-1} RNH, 250 mg of pure drug was dissolved in 20 ml of water in a beaker to which were added 10 ml of 5 M hydrochloric acid and 500 mg of zinc dust and the beaker was set aside for 20 min. Then, filtered using glass wool, and the filtrate and the washings were collected in a 250 ml calibrated flask, and diluted to the mark with water and mixed. This solution was used in titrimetric work. The stock solution (1000 µg ml^{-1} RNH) was diluted appropriately to obtain working concentrations of 100 and 200 µg ml^{-1} RNH for spectrophotometric study by method B, and methods C and D, respectively.

General Procedures

Titrimetry (Method A). A 10 ml aliquot of pure drug solution containing 5-10 mg of reduced RNH was accurately measured into a 100 ml titration flask. The solution was acidified by adding 3 ml of 5 M sulfuric acid, and total volume adjusted to 15 ml by adding water, Ten ml of 0.01 M $K_2Cr_2O_7$ solution was added by means of pipette, the contents were mixed well and the flask let stand for 15 min with occasional swirling. Finally, 2 ml of syrupy phosphoric acid and 5 drops of barium diphenylamine sulfonate indicator were added and the unreacted dichromate titrated with 0.06 M FAS solution. A blank was run under identical conditions and the amount of drug in the transferred aliquot was calculated from:

$$\text{Amount (mg)} = (B - S)\, M_w\, R/0.333$$

where B = volume of FAS solution consumed in the blank titration in ml, S = volume of FAS solution consumed in the sample titration in ml, M_w = relative molecular mass of drug and R = concentration of dichromate solution, M.

Spectrophotometry (Method B). Different aliquots (0.0, 0.5, 1.0,……5.0 ml) of standard 100 µg ml^{-1} RNH (reduced) solution were accurately measured into a series of 10 ml calibrated flasks by means of a burette and the total volume was adjusted to 5 ml by adding water. To each flask was added 1 ml of 5 M sulfuric acid followed by 1 ml of 30 µg

ml^{-1} $K_2Cr_2O_7$ solution. The contents were mixed well and the flasks were let to stand for 10 min with intermittent shaking. Then, 1.0 ml of 0.25% DPC solution was added, the volume was diluted to the mark with water and mixed well. The absorbance of each solution was measured at 540 nm against a water blank.

Spectrophotometry (Method C). Known aliquots (0.0, 0.25, 0.5, 1.0,…..4.0 ml) of standard 200 µg ml^{-1} RNH (reduced) solution were accurately transferred into a series of 10 ml calibrated flasks by means of a micro burette and the total volume was brought to 4 ml by adding water. Two ml of 5 M hydrochloric acid was added to each flask followed by 1 ml of 50 µg ml^{-1} $K_2Cr_2O_7$ solution. The reactants were mixed well and allowed to stand for 10 min with occasional shaking. Then, 1 ml of 400 µg ml^{-1} FAS solution was added, mixed well and let to stand for 1 min. Lastly, 1 ml of 2 M potassium thiocyanate solution was added, the volume was diluted to the mark with water, mixed well and absorbance of the resulting solution measured at 470 nm against a water blank.

Spectrophotometry (Method D). Varying volumes of standard drug solution (0.0, 0.5, 1.0 ….5.0 ml; 200 µg ml^{-1} RNH) were accurately measured and transferred into a series of 10 ml calibrated flasks and the volume was brought to 5 ml by adding water. The solution was acidified by adding 1 ml of 5 M sulfuric acid; and 1 ml of 90 µg ml^{-1} $K_2Cr_2O_7$ was then added to each flask. The flasks were kept aside for 10 min with periodic shaking. Then, 1 ml of 700 µg ml^{-1} FAS was added and mixed. After 1 min, 1 ml of 0.25% orthophenanthroline solution was added to each flask and the volume was diluted to the mark with 1:1 ammonia solution and mixed. The absorbance of the solution was measured at 510 nm against a reagent blank after 10 min.

In methods B and C, the decreasing values of absorbance were plotted against the concentration of RNH, and in method D, increasing absorbance values were plotted as a function of RNH concentration, to obtain the calibration graphs. The concentration of the unknown was read from the calibration graph or computed from the regression equation derived from the Beer's law data.

Procedure for formulations. Preparations containing RNH were purchased from local commercial sources and subjected to analysis. A quantity of finely ground tablet powder or an aliquot of injection solution equivalent to 100 mg of RNH was accurately weighed, measured into a 100 ml beaker and mixed with 20 ml of water, 10 ml of 5 M hydrochloric acid and 100 mg of zinc dust, and kept aside for 20 min. Then, filtered using Whatmann No. 42 filter paper, the residue was washed with water and the filtrate and washings were collected in a 100 ml calibrated flask and the volume was diluted to the mark with water. A suitable aliquot (5 ml) of the tablet, extract or injection solution was analyzed by titrimetry. The tablet extract injection solution (1000 µg ml^{-1} RNH) was diluted stepwise with water to get 100 and 200 µg ml^{-1} RNH solutions (reduced) for analysis by spectrophotometric methods B, C, and D, respectively.

RESULTS AND DISCUSSION

Potassium dichromate has been a useful oxidizing agent in the determination of a number of substances of pharmaceutical importance [26-30]. All the four methods described here are indirect and are based on the determination of residual dichromate after allowing the reaction between a measured excess of dichromate and RNH to go to completion under acidic conditions. In titrimetry, the reaction is followed by back titration of unreacted dichromate while the spectrophotometric methods entail well-known color reactions for chromium(VI), iron(II) or iron(III).

Method Development and Optimization of Experimental Conditions

Method A. Direct titration of RNH with dichromate was not feasible. Hence, preliminary experiments were conducted to find out the optimum conditions for the indirect titration. The oxidation reaction between RNH and dichromate was found to occur when the reactants were mixed in acid medium and allowed to stand for some time. However, the rate of the reaction at room temperature (30 ± 2 °C) was found to be depend on the nature of acid and its concentration, and standing time. The reaction was found to be slow in HCl medium. However, it was quantitative with a stoichiometry of 1:0.333 (RNH:$K_2Cr_2O_7$) at 0.6 M H_2SO_4 concentration over all.

From this stoichiometry, it is implied that reduced RNH is oxidized to its sulfoxide. This stoichiometry was achieved with a standing time of 15 min and longer contact times

Table 2. Evaluation of Accuracy and Precision

Method	RNH taken ($\mu g\ ml^{-1}$)	RNH found ($\mu g\ ml^{-1}$) [b]	Range ($\mu g\ ml^{-1}$)	RE (%)	SD ($\mu g\ ml^{-1}$)	RSD (%)	CL[c]
A[a]	5.0	4.9	0.2	1.6	0.16	3.25	4.9 ± 0.1
	7.0	6.9	0.3	2.0	0.22	3.10	6.9 ± 0.2
	9.0	8.7	0.5	3.5	0.26	3.00	8.7 ± 0.2
B	10.0	9.8	0.6	1.5	0.23	2.34	9.8 ± 0.2
	20.0	19.5	1.3	2.6	0.48	2.46	19.5 ± 0.4
	30.0	30.0	1.4	1.0	0.43	1.44	30.0 ± 0.4
C	20.0	20.4	2.1	2.0	0.58	2.84	20.4 ± 0.5
	40.0	40.3	2.1	0.6	0.69	1.71	40.3 ± 0.6
	60.0	61.2	1.2	2.0	0.43	0.70	61.2 ± 0.4
D	30.0	29.5	2.8	1.7	0.62	2.10	29.5 ± 0.6
	50.0	48.8	3.2	2.3	0.87	1.78	48.8 ± 0.8
	80.0	78.7	1.2	1.7	1.22	1.55	78.7± 1.1

RE = relative error, SD = standard deviation, RSD = relative standard deviation, CL = confidence limit. [a]In method A, amount taken/found, range and SD are in mg. [b]Mean value of seven determinations. [c]At 95% confidence level and for six degrees of freedom.

resulting in the consumption of a little more dichromate. But no significant stoichiometry was obtained up to 30 min. Hence, it is necessary that the residual dichromate be back titrated immediately after 15 min contact time to get precise results. Using 10 ml of 0.01 M $K_2Cr_2O_7$ under the stated experimental conditions, 5-10 mg of RNH can be determined with high accuracy and precision (Table 3). The relation between the amount of drug and the endpoint was also examined. The linearity is apparent from the correlation coefficient of -0.9918 (n = 7) implying that the reaction occurs stoichiometrically in the range studied (5-10 mg). Inconsistent results were obtained when RNH was used as such; however, the reduced RNH gave satisfactory results.

Method B. One of the earliest spectrophotometric methods developed for chromium(VI) [31] is based on the formation of intense purple color with diphenylcarbazide (DPC) in sulfuric acid medium, and measurable at 540 nm. Use is made of the ability of chromium(IV) to oxidize the RNH and form a colored product with DPC, to develop an indirect spectrophotometric method for RNH.

RNH when added in increasing concentrations to a fixed concentration of dichromate consumes the latter and there will be a concomitant decrease in the latter's concentration. On adding diphenylcarbazide to decreasing concentrations of dichromate, the absorbance decreases with increasing concentration of RNH (Fig. 2.), which is corroborated by the correlation coefficient of -0.998. Preliminary experiments revealed that 3 $\mu g\ ml^{-1}$ $K_2Cr_2O_7$ could be determined under the specified acid and reagent concentrations. Hence, different amounts of RNH were reacted with 1 ml of 30 $\mu g\ ml^{-1}$ $K_2Cr_2O_7$, and after the oxidation reaction was complete, unreacted dichromate were determined by treating with diphenylcarbazide, and measuring the absorbance at 540 nm. This enabled to fix the concentration range of RNH that could be determined by the method.

Since the original procedure for chromium(VI) using diphenylcarbazide employed 0.1 M sulfuric acid concentration, 1 ml of 1 M acid in a total volume of 7 ml was

Table 3. Quantitation Parameters of Spectrophotometric Methods

Parameter	Method B	Method C	Method D
λ_{max} (nm)	540	470	510
Beer's law limits (µg ml^{-1})	5-50	5-80	10-100
Molar absorptivity (l mol^{-1} cm^{-1})	3.4×10^4	2.2×10^4	1.2×10^4
Sandell sensitivity (µg cm^{-2})	0.093	0.138	0.257
Limit of detection (µg ml^{-1})	0.83	1.59	3.07
Limit of quantification (µg ml^{-1})	2.52	4.83	9.28
Regression equation, Y[a]			
Intercept (a)	0.605	0.641	0.005
Slope (b)	- 0.011	-0.007	0.004
Correlation coefficient (r)	- 0.998	-0.994	0.999

[a]Y = a + bX where Y is the absorbance in a cell of 1cm path length and X concentration in µg ml^{-1}.

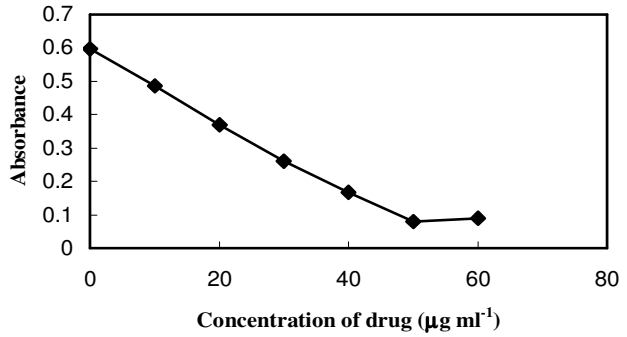

Fig. 2. Beer's law for method B.

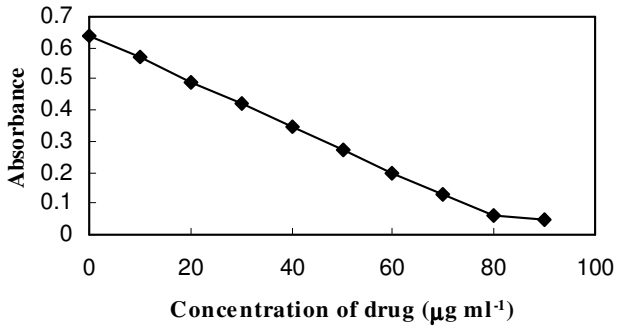

Fig. 3. Beer's law curve for method C.

used for the oxidation reaction, and the same was maintained for the estimation of residual dichromate. Under the stated experimental conditions, the reaction between RNH and dichromate was complete in 10 min and contact times up to 30 min had no effect on the absorbance of the color, which was stable up to 60 min thereafter.

Method C. Complex formation between iron(III) and thiocyanate is a well-established reaction that is widely used for trace level determination of iron [32]. The present method is based on the oxidation of RNH by a known excess of dichromate in sulphuric acid medium, reducing the unreacted dichromate with iron(II), and subsequent formation of iron (III)-thiocyanate complex which is measured at 470 nm. When a fixed concentration of dichromate is reacted with increasing concentrations of RNH, there occurs a concomitant fall in the former's concentration. When the unreacted dichromate is reduced by a fixed concentration of iron(II), there will be a proportional decrease in the resultant iron(III) concentration. This is observed as a proportional decrease in the absorbance of iron(III)-thiocyanate complex on increasing the concentration of RNH (Fig. 3) which formed the basis of determination.

Two blanks were prepared for this study. The reagent blank which contained optimum concentrations of all the reagents except RNH gave maximum absorbance (Fig. 3). The other blank was prepared in the absence of $K_2Cr_2O_7$ and drug to determine the contribution of other reagents to the absorbance of the system. Since the absorbance of the second blank was negligible, the absorbance measurement was made against water blank. Chromium(III) formed in the reaction is

so small (max. concentration is ~1 µg ml^{-1}) that it does not interfere in the measurement at 470 nm.

The conditions for the determination of iron(III) with thiocyanate are well established [32]. Hence, the various conditions involved in the oxidation of RNH by dichromate and its subsequent reduction by iron(II) were optimized.

Nitric acid or hydrochloric acid media can also be used for the complex formation reaction [32]. Apprehending that nitric acid, being an oxidizing agent, would interfere with the oxidation step, it was not tried. Sulfuric acid medium, although convenient for oxidation step, was not preferred since it is reported to reduce the color intensity of iron(III) thiocyanate complex. A 1 M hydrochloric acid concentration was found adequate for the oxidation as well as complexation steps.

Because of non-linearity at higher concentrations [32], 5.5 µg ml^{-1} was taken as the upper limit of iron(III) that could be determined by thiocyanate method. Stoichiometrically, 48.25 µg of $K_2Cr_2O_7$ would be required to produce it from 386.1 µg of FAS. However, slightly larger amounts (50 µg $K_2Cr_2O_7$ and 400 µg of FAS) were used to ensure quantitative reaction. Although a fixed amount of FAS is not really needed, large amounts are undesirable since iron(II) tends to undergo aerial oxidation. Hence, a fixed amount (400 µg) of FAS, enough to reduce the total $K_2Cr_2O_7$, was employed in the investigation.

The oxidation of RNH by dichromate was complete in 10 min and subsequent oxidation of iron(II) to iron(III) by residual dichromate, and the complex formation reaction with thiocyanate were instantaneous under the described experimental conditions. The developed color was stable up to 60 min in the presence of the reaction products.

Method D. The colorimetric method based on the complexation reaction between iron(II) and orthophenanthroline forming the red tris(*o*-phenanthroline) iron(II) chelate ion, called ferroin, continues to be the most sensitive and the most widely used procedure for the determination of iron in a variety of materials [33]. This reaction coupled with the oxidizing property of dichromate has been made use in developing an indirect spectrophotometric method for RNH. The drug in varying concentrations when reacted with a fixed and known concentration of dichromate in sulfuric acid medium consumes the latter in proportionate concentrations for its oxidation and there will be a concomitant decrease in dichromate amount. When the decreasing concentrations of oxidant are treated with a fixed and known concentration of iron(II) in the same acidic conditions, there will be a proportional increase in the concentration of iron(II). This is shown by the increase absorbance of the orthorphenanthroline complex resulting from the residual iron(II). The absorbance measured at 510 nm is found to increase linearly with the concentration of RNH (Fig. 4) serving as the basis for the determination of drug.

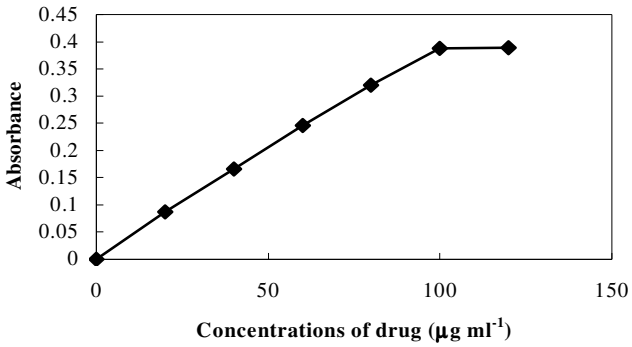

Fig. 4. Beer's law curve for method D.

The optimum conditions were established based on the formation of maximum color through variation of parameters such as nature of acid and its concentration, reaction time and volume of ammonia required to raise the pH to about 4.

Since the oxidation step was slow in hydrochloric acid or phosphoric acid medium, sulfuric acid was selected as the medium for the oxidation of RNH by dichromate. One ml of 5 M sulfuric acid in a total volume of 7 ml was found to effect oxidation in a reasonable time of 10 min. Higher concentrations were seldom used since they would require large volumes of ammonia to raise the pH to a level (~4) required for iron(II)-orthophenanthroline complex formation.

Fixing 10 µg ml^{-1} as the upper limit of iron(II) that could be determined by orthophenanthroline method, 700 µg of FAS was used in this study. Stoichiometrically, this would react with 87.46 µg of $K_2Cr_2O_7$. However, a slightly higher amount (90 µg) of oxidant was used to ensure complete oxidation of iron(II) and to produce a colorless blank.

The volume of 1:1 ammonia was not critical since the stability and sensitivity of ferroin are unaffected over a wide

pH range. However, 1 ml of 1:1 ammonia was used to raise the pH to about 4. Under the described experimental conditions, ferroin complex was found to be stable for several hours.

Analytical Parameters of Spectrophotometric Methods

A linear relation was found between absorbance at λ_{max} and concentration ranges given in Table 3 for the three methods. The apparent molar absorptivity and Sandell sensitivity values together with the limits of detection (LOD) and quantification (LOQ) compiled in Table 3 are indicative of the high sensitivity of the proposed methods. The LOD and LOQ were calculated using LOD = 3.3 σ/S and LOQ = 10 σ/S, where σ is the standard deviation of seven blank determinations and S is the slope of the calibration curve.

Method Validation

The accuracy and precision of the methods were evaluated by performing seven replicate seven replicate analysis of pure drug solution at three levels (within the working ranges). The relative error (%), an indicator of accuracy, was within 2.5% and within-day precision which is also called the repeatability expressed in relative standard deviation (%) was less than 3% indicating high accuracy and repeatability of the methods. The reproducibility of the methods, also called the day-to-day precision, was evaluated by performing replicate analyses on pure drug solution at three levels over a period of five days preparing all solutions afresh each day. The day-to-day RSD values were less than 4% reflecting the usefulness of the methods in routine use.

Application

Commercially available tablets and injections were successfully analyzed for the RNH content by the proposed methods. The assay results are presented in Table 4. As can be seen from the Table 4, the results obtained agreed with the label claim and also those of the reference method [34], which consisted of the measurement of the absorbance of the tablet extract or injection solution in 0.1 M hydrochloric acid at 225 nm. The performance of the proposed methods was judged further by the Student's t-test for accuracy and F-test for precision. At the 95% confidence level, the calculated t- and F-values did not exceed the tabulated values (t = 2.77 and F = 6.39) suggesting that the methods are as accurate and precise as the reference method.

The accuracy and validity of the proposed methods were further ascertained by performing recovery studies. The pre-analyzed tablet powder/injection solution was spiked with pure RNH at three levels and the total was found by the proposed methods. Each determination was repeated three times. The results summarized in Table 5 reveal good recoveries of pure drug added and non-interference from tablet excipients such as talc, starch, gelatin, gum acacia, calcium carbonate, calcium gluconate, calcium dihydrogenortho-phosphate, sodium alginate and magnesium stearate. This is also clear from the assay results presented in Table 4.

CONCLUSIONS

Four new methods using potassium dichromate as the oxidimetric reagent have been developed based on different reaction schemes. Titration method, although applicable over a narrow range (5-10 mg), has a better sensitivity compared to the only titrimetric method available for ranitidine. The three spectrophotometric methods have no procedural or instrumental wrangles which are common with the existing procedures (Table 1). The methods offer wide dynamic linear ranges of applicability, highly stable colored species and shorter contact times, and are free from either heating or extraction steps. The methods are more sensitive than many of the existing methods and are comparable to the procedure employing $KMnO_4$ or N-bromosuccinimide (NBS) and azines dyes [18]. The relative advantage of the proposed methods is that it uses potassium dichromate which is stable in solution unlike $KMnO_4$ or NBS. These advantages coupled with high accuracy and precision render the methods suitable for routine analysis.

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Table 4. Results of Analysis of Formulations Containing RNH and Statistical Comparison with Reference Method

Brand name and dosage form	Label claim (mg/tablet or ml)	%found ± SD				
		Reference method	Method A	Method B	Method C	Method D
Tablets	150	99.1 ± 0.9	100.4 ± 1.7	99.8 ± 2.1	98.6 ± 1.6	101.1± 1.9
Ranitin[a]			t = 1.60	t = 0.75	t = 0.63	T = 2.19
			F = 3.12	F = 5.08	F = 2.82	F = 3.91
	300	102.5 ± 1.1	101.9 ± 2.8	101.0 ± 0.8	102.2 ± 1.9	103.3 ± 2.2
			t = 0.54	t = 2.65	t = 0.40	t = 0.69
			F = 6.48	F = 2.17	F = 3.46	F = 4.00
Histac[b]	150	97.8 ± 1.0	96.9 ± 1.6	98.2 ± 0.8	99.1 ± 1.7	98.4 ± 1.4
			t = 1.11	t = 0.55	t = 1.52	t = 0.68
			F = 2.80	F = 1.36	F = 3.08	F = 2.16
	300	100.3 ± 1.4	101.6 ± 2.1	101.1 ± 2.0	100.7 ± 2.7	100.7 ± 2.6
			t = 1.13	t = 0.74	t = 0.32	t = 0.26
			F = 2.10	F = 1.90	F = 3.51	F = 3.30
Zinetac[c]	150	98.5 ± 0.8	99.7 ± 1.3	100.0 ± 1.8	99.3 ± 1.4	98.2 ± 1.7
			t = 1.72	t = 1.79	t = 1.16	t = 0.43
			F = 2.19	F = 4.68	F = 2.56	F = 4.09
	300	99.7 ± 1.4	100.3 ± 2.7	101.0 ± 0.7	100.9 ± 2.0	100.3 ± 1.3
			t = 0.47	t = 2.00	t = 1.11	t = 0.73
			F = 4.06	F = 3.57	F = 2.08	F = 1.13
Aciloc[d]	150	102.7 ± 1.0	101.7 ± 1.6	103.2 ± 0.9	102.7 ± 1.9	101.7 ± 0.7
			t = 1.12	t = 0.83	t = 0.098	t = 1.84
			F = 2.43	F = 1.22	F = 3.19	F = 2.34
	300	100.4 ± 1.3	99.6 ± 2.7	101.1 ± 1.6	102.0 ± 0.7	100.9 ± 1.4
			t=0.68	t = 0.78	t = 2.40	t = 0.50
			F=3.94	F = 1.46	F = 3.28	F = 1.03
Injections	25	103.2 ± 1.1	104.5 ± 1.6	102.7 ± 0.6	103.7 ± 2.8	104.3 ± 1.4
Ranitin[a]			t = 1.57	t = 0.86	t = 0.46	t = 1.46
			F = 2.13	F = 3.21	F = 6.18	F = 1.64
Histac[b]	25	97.6 ± 1.0	98.3 ± 2.0	96.7 ± 1.4	97.34 ± 1.56	99.0 ± 0.6
			t = 0.74	t = 1.15	t = 0.36	t = 2.73
			F = 4.08	F = 2.16	F = 2.53	F = 3.06
Ranitidine[c]	25	101.7 ± 1.3	102.3 ± 1.3	101.0 ± 2.7	101.1 ± 0.9	102.4 ± 1.7
			t = 0.68	t = 0.73	t = 0.91	t = 0.70
			F = 1.09	F = 2.94	F = 2.15	F = 1.91
Aciloc[d]	25	99.3 ± 1.1	100.6 ± 1.8	101.0 ± 1.3	100.6 ± 1.4	101.1 ± 1.2
			t = 1.50	t = 2.36	t = 1.63	t = 2.62
			F = 2.76	F = 1.41	F = 1.79	F = 1.24

Percent found is the mean value of five determinations ψ Marketed by: [a]Torrent pharmaceuticals. [b]Ranbaxy Chemicals. [c]Glaxo Smithkline Pharm. Ltd. [d]Cadila Pharmaceuticals.

Table 5. Results of Recovery Study by Standard-Addition Method

Formulation studied	Method A				Method B			
	RNH in sample (mg)	Pure RNH added (mg)	Total found (mg)	Pure RNH recovered (%)	RNH in sample (µg)	Pure RNH added (µg)	Total found (µg)	Pure RNH recovered (%)
Ranitin	2.5	3.0	5.6	103.6	49.9	100	148.6	98.6
tablets	2.5	4.0	6.5	98.8	49.9	200	252.5	101.3
(150 mg)	2.5	5.0	7.6	102.3	49.9	300	364.2	104.8
Ranitin	2.6	2.5	5.3	106.7	51.3	100	153.0	101.7
injections	2.6	5.0	7.8	103.3	51.3	200	256.4	102.5
(25 mg)	2.6	7.5	10.2	101.5	51.3	300	353.5	100.7

Formulation studied	Method C				Method D			
	RNH in sample (µg)	Pure RNH added (µg)	Total found (µg)	Pure RNH recovered (%)	RNH in sample (µg)	Pure RNH added (µg)	Total found (µg)	Pure RNH recovered (%)
Ranitin	49.3	150	194.2	96.6	101.1	200	308.4	103.6
tablets	49.3	300	341.2	97.3	101.1	400	534.0	108.2
(150 mg)	49.3	450	501.8	100.6	101.1	600	727.1	104.3
Ranitin	51.9	150	205.8	102.6	104.3	200	319.0	107.3
injections	51.9	300	368.7	105.7	104.3	400	517.4	103.3
(25 mg)	51.9	450	503.7	100.4	104.3	600	714.8	101.7

Percent recovered is the mean value of three determinations.

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