A Novel Spectrophotometric Methods for Quantitative Determination of Ciprofloxacin Hydrochloride and Tinidazole in Tablets using Hydrotrropic Solubilizing Agent

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ABSTRACT

Two accurate, precise, sensitive and economical procedures for simultaneous determination of ciprofloxacin hydrochloride and tinidazole in tablet dosage form have been developed. In the present investigation, 2.0 M urea solution (hydrotrropic solubilizing agent) was employed to solubilize, ciprofloxacin (a poorly water soluble drug) from fine powder of its tablets to carryout spectrophotometric analysis. The methods employed were simultaneous equation method (method I), absorbance ratio method (method II). The result showed that Beers’ law was obeyed in concentration range of 5-50 µg/ml with good linearity (r²>0.99) for both the drugs in both methods. The recoveries were within 99.42-101.27% for ciprofloxacin hydrochloride and 99.61-101.81% for tinidazole. Precision was good with acceptable limits of detection (LOD) and quantitation (LOQ) for both compounds. The average content of the compounds were 100.03 and 101.05% in method (I), 99.55 and 101.05% in method (II) for ciprofloxacin hydrochloride and tinidazole respectively. The optimized methods showed good reproducibility and recovery with standard deviation of < 1.0% and percent relative standard deviation less then 2.0%.

Key words: Simultaneous equation method, Absorbance ratio method, Ciprofloxacin hydrochloride, Tinidazole, Hydrotrropic agent

INTRODUCTION

The term hydrotropy has been used to designate the increase in solubility of various substances in water due to the presence of large amounts of additives1. Various organic solvents have been employed for the solubilization of poorly water soluble drugs for spectrophotometric estimations. Drawbacks of organic solvents include higher cost, toxicity, pollution and error in analysis due to volatility. The primary objective of this study was to employ hydrotrropic solubilizing agents for the selected drugs to preclude the use of organic solvents. Chemically Ciprofloxacin Hydrochloride (CPH) is (1-cyclopropyl-6-fluoro-1, 4-dihydro-4-oxo-7-(1-piperazinyl)-3-quinolinecarboxylic acid) is fluoro quinolones and antimicrobials with potent activity against a broad spectrum of bacteria2-4. Literature survey revealed that chromatographic method was reported for its estimation from tablet formulation and spectrophotometric methods for estimation in combination dosage forms. Chemically Tinidazole (TZ) is (1-(2-ethylsulfonylethyl)-2-methyl-5-nitro-imidazo[1,2-a]pyridine), antiprotozoal and anti-bacterial drugs and have wide range of applications. This class of hydrotropic agents has better characteristics than the other organic solvents. They are eco-friendly, rapid, precise and accurate methods to analyze the drugs simultaneously. Combined dosage form, hence an attempt has been made to develop simple, sensitive, eco-friendly methods for estimation in combined dosage forms that are being used either alone or in combination for the treatment of dysentery of amoebic, bacterial or mixed origin.

Experimental

The term hydrotropy has been used to designate the increase in solubility of various substances in water due to the presence of large amounts of additives5. Various organic solvents have been employed for the solubilization of poorly water soluble drugs for spectrophotometric estimations. But so far no spectroscopic methods has been reported for simultaneous estimation of CPH and TZ in combined dosage form, hence an attempt has been made to develop simple, sensitive, economical, rapid, precise and accurate methods to analyze the drugs simultaneously.

MATERIALS AND METHODS

Instruments

UV-Visible double beam spectrophotometer, Shimadzu model-1700 having spectral bandwidth 3 nm and of wavelength accuracy ±1 nm, with 1 cm quartz cells was used. All weighing were done on electronic balance (Shimadzu, Model AY- 120).

Reagents and Chemicals

Analytically Pure samples of CPH and TZ was obtained as gift sample from Zest Pharma Pvt. Ltd, Indore (MP), India and were used as such without further purification. The tablet dosage form, Cipracil TZ (contain CPH 500mg, TZ 600 mg) was procured from the local market, Indore, India. 2.0 M urea was selected as hydrotrropic solubilizing agent. All other material used was of analytical reagent grade.

Methods

Vierordt’s simultaneous equation method (Method I)

For selection of analytical wavelength for Simultaneous equation method6-11 the wavelength

\[ \lambda_{max}^1 \text{ is based on the absorption of drugs X and Y at the wavelength maxima of the other. The} \]

\[ Q = \lambda^1 \text{ and } \lambda^2 \text{ are the absorbances of samples at the 272 and 321 nm respectively.} \]

\[ C_y = \frac{A_y}{A_x} \text{ and } C_x = \frac{A_x}{A_y} \text{ are the absorbances of CPH at } \lambda_y \text{ and } \lambda_x \text{ respectively; } C_y = \frac{Q_m-Q_y}{Q_x-Q_y} \times A_x \text{ and } C_x = \frac{Q_m-Q_x}{Q_y-Q_x} \times A_y. \]

Graphical absorbance ratio Q-Analysis method (Method II)

In absorption ratio method12-13 absorbances of both the drugs were calculated at two selected wavelengths, among which } \lambda_x \text{ is the wavelength of isoabsorptive point of both drugs, and } \lambda_y \text{ is the } \lambda_{max} \text{ of either drug among both drugs. From the overlain spectra (Figure 3) wavelength 291 (isoabsorption point) and 321(} \lambda_{max} \text{ of TZ) were selected for study. The absorbances at 291 nm and 272 nm for CPH were obtained and similarly for TZ absorbances are measured at 291 nm and 321 nm. The concentration of the individual components were calculated by using the following equations: } C_x = \frac{Q_m-Q_y}{Q_x-Q_y} \times A_x \text{ and } C_y = \frac{Q_m-Q_y}{Q_x-Q_y} \times A_y. \text{ The absorbance analysis was performed by using Eqn-1 and Eqn-2. Where } \lambda_y \text{ and } \lambda_x \text{ are the concentrations of CPH and TZ respectively in the diluted sample, and } A_x \text{ and } A_y \text{ are the absorbances of CPH at } \lambda_x \text{ and } \lambda_y \text{ respectively; Cx and Cy are the concentration of CPH and TZ respectively.}

Preliminary solubility studies of drugs

Solubility of both drugs was determined at 28 ± 2 °C. An excess amount of drug was added to a 50 ml of distilled water in test tube. The tube was warmed to 50°C and kept for stirring for 30 min. The volume of the supernatant was measured, and the solubility of each drug was calculated.

Preparation of standard solution for calibration curves of CPH and TZ

About 50mg each of CPH and TZ were accurately weighed and transferred to 50ml of volumetric flask separately. 40 ml, 2.0 M urea was used to solubilize after shaking for 10 to 15 minutes. Rest of the volume was made up with distilled water to get solution of 1000 µg/ml. Stock solutions of 100 µg/ml of each drug were prepared by further dilution and scanned over the range of 400nm-300nm in the spectrum mode to get the overlain spectra of both drugs. The spectra exhibit major absorbance maxima at 272 nm and 321 nm for CPH and TZ respectively. Beers-Lambert law obeyed in the range of 5-50 µg/ml and 5-45 µg/ml for CPH and TZ respectively. Six mixed standards 5,10,15,20,25,30 for CPH; and 30,25,20,15,10,5 for TZ

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were prepared from stock solutions of CPH and TZ for further study.

Analysis of Tablet Formulation
Twenty tablets (brand name- Cipract TZ) were taken and their average weight was determined, they were crushed to fine powder. Then powder equivalent to 50 mg (101.91 mg) of TZ was taken in 50ml volumetric flask and 40 ml, 2.0 M urea was used to solubilize after shaking for 10 to 15 minutes. Rest of the volume was made up with distilled water to get solution of 1000µg/ml. Stock solutions of 100µg/ml of each drugs were prepared by further dilution. The supernatant liquid was transferred to 50ml of volumetric flask through a whatman No-41 filter paper. The residue was washed twice with water and the combined filtrate was made up to 50ml mark with water. The above solution was further diluted to get a solution containing 10µg/ml of CPH and 12 µg/ml of TZ. The above binary mixture was analyzed at appropriate wavelengths and values of the absorptions were substituted in the respective formulas (Eqn.1,2,3,4) to obtain the content of CPH and TZ. CPH and TZ was determined from their calibration curve plotted between absorption difference and concentration. The results of analysis were given in Table 1.

Table 1: Result of pharmaceutical formulation analysis

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Method-A CPH</th>
<th>Method-B CPH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TZ</td>
<td>CPH</td>
</tr>
<tr>
<td>Label claim (mg/Tab)</td>
<td>500</td>
<td>500</td>
</tr>
<tr>
<td>Found (mg/Tab)</td>
<td>499.87</td>
<td>600.09</td>
</tr>
<tr>
<td>Drug content*</td>
<td>100.03</td>
<td>100.82</td>
</tr>
<tr>
<td>±S.D</td>
<td>0.384</td>
<td>0.530</td>
</tr>
<tr>
<td>%COV</td>
<td>0.331</td>
<td>0.284</td>
</tr>
<tr>
<td>SE</td>
<td>0.403</td>
<td>0.203</td>
</tr>
</tbody>
</table>

*Value for drug content (%) are the mean of six estimation, Method-A: Vierordt's simultaneous equation method, Method-B: Graphical absorbance ratio Q-Analysis method, S.D: Standard deviation, COV: Coefficient of variance and S.E: Standard error.

Recovery studies
To check the accuracy of the developed methods and to study the interference of formulation additives, analytical recovery experiments was carried out by standard addition method. From that total amount of drug found and percentage recovery was calculated. The results were reported in Table 2.

Table 2: Result of Recovery studies

<table>
<thead>
<tr>
<th>Method</th>
<th>Drug</th>
<th>Amount (mg/ml) taken</th>
<th>% Recovery±S.D</th>
<th>COV%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Method-A</td>
<td>CPH</td>
<td>30 5</td>
<td>99.42±0.442</td>
<td>0.321</td>
</tr>
<tr>
<td></td>
<td>TZ</td>
<td>60 10</td>
<td>101.18±0.820</td>
<td>0.521</td>
</tr>
<tr>
<td>Method-B</td>
<td>CPH</td>
<td>90 15</td>
<td>99.3±0.001</td>
<td>0.119</td>
</tr>
<tr>
<td></td>
<td>TZ</td>
<td>60 15</td>
<td>100.01±0.010</td>
<td>0.104</td>
</tr>
<tr>
<td></td>
<td></td>
<td>90 15</td>
<td>100.27±0.711</td>
<td>0.471</td>
</tr>
<tr>
<td></td>
<td></td>
<td>60 10</td>
<td>101.18±0.420</td>
<td>0.291</td>
</tr>
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<td></td>
<td></td>
<td>60 10</td>
<td>101.8±0.307</td>
<td>0.108</td>
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<td></td>
<td></td>
<td>90 15</td>
<td>100.99±0.182</td>
<td>0.222</td>
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<td></td>
<td></td>
<td>30 5</td>
<td>100.26±0.326</td>
<td>0.209</td>
</tr>
<tr>
<td></td>
<td></td>
<td>60 10</td>
<td>101.32±0.621</td>
<td>0.491</td>
</tr>
<tr>
<td></td>
<td></td>
<td>90 15</td>
<td>99.61±0.536</td>
<td>0.203</td>
</tr>
</tbody>
</table>

%Recovery is mean of three estimation, Method-A: Vierordt’s simultaneous equation method, Method-B: Graphical absorbance ratio Q-Analysis method, S.D: Standard deviation, COV: Coefficient of variance.

Validation of the developed methods
The developed methods for simultaneous estimation of CPH and TZ were validated as per ICH guidelines.

Accuracy
To check the accuracy of the developed methods and to study the interference of formulation additives, analytical recovery experiments was carried out by standard addition method. Total amount of drug found and percentage recovery was calculated and results were reported in Table 2.
RESULTS AND DISCUSSION

All UV spectrophotometric methods were found to be simple, accurate, economic and rapid for simultaneous estimation of CPH and TZ in tablet dosage form. By performing these methods it was found that both drugs shown good regression value at their respective wavelengths and the recoveries were within 99.42-101.27% for CPH and 99.61-101.81% for TZ. Precision was good with acceptable limits of detection (LOD) and quantitation (LOQ) for both compounds. The optimized methods showed good reproducibility and recovery with standard deviation of < 1.0% and percent relative standard deviation less than 2.0%.

Since urea do not interfere above 245 nm, therefore other poorly water-soluble drugs can also be estimated above 245 nm by hydrotropy avoiding the use of organic solvents. There was no interference of urea and commonly used additives present in tablet formulations. A critical evaluation of the proposed methods was performed by statistical analysis of the experimental data. In order to demonstrate the validity and applicability of the proposed methods, recovery studies were performed by analyzing synthetic mixture of CPH and TZ with different composition ratio.

Hence, the proposed methods could be successfully applied to the determination of CPH and TZ in the commercially available bulk and tablet dosage form. Thus, it may be concluded that the proposed methods of analysis are new, simple, cost-effective, environmentally friendly, safe, accurate and reproducible. Definitely, there is further scope of 2.0 M urea solution as solubilizing agent for other poorly water-soluble drugs. There was no interference of urea in the estimation. The proposed method can be successfully employed in the routine analysis of Poorly water soluble drugs in pharmaceutical dosage forms.

CONCLUSION

It is thus concluded that the proposed method is new, simple, cost effective, accurate, safe, free from pollution and precise and can be successfully employed in the routine analysis of Poorly water soluble drugs in pharmaceutical dosage forms.

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REFERENCES

13. Patel,PU.; Shujah, BN.; Patel,CM.; Patel,MM.; Patel,GC.; Patel,GM.; Simultaneous spectrophotometric estimation of Gatifloxacin and Ofloxacin in mixture, Indian J. Pharm. Sci.; 2005; 40:149-152.
15. Shinde, VM.; Desai, BS.; Tendulkar, DM.; Selective determination of fluoroquinolone derivative from tablets by phase HPLC, Indian Drugs.;1996;35:715-717.