SIMULTANEOUS UV SPECTROPHOTOMETRIC METHODS FOR ESTIMATION OF CEFADROXIL AND PROBENCID IN TABLET DOSAGE FORM

Mayur S. Jain\(^1\), Sunil R. Bavaskar\(^1\), Shashikant D. Barhate\(^1\), Jintendra D. Fegade\(^2\)

\(^1\)Shree Sureshdada Jain Institute of Pharmaceutical Education and Research, Jammer, Dist Jalgaon, Maharashtra, India.
\(^2\)TVE’S College of Pharmacy, Nehru Vidyanagar, Distt Jalgaon, Faizpur, Maharashtra 425 503, India.

ABSTRACT

Two methods for simultaneous estimation of Cefadroxil and Probencid in combined tablet dosage form have been developed. The first UV spectrophotometric method was a determination using the simultaneous equation method at 233 nm and 247 nm. The second UV spectrophotometric method is the Q-analysis (absorption ratio) method, which involves the formation of absorbance equation at 242 nm (isobestic point) and at 247 nm the maximum absorption of Probencid. The linearity ranges for Cefadroxil and Probencid both were 10-60μg/ml respectively. The accuracy of the methods was assessed by recovery studies was found to be 99.43±0.75 and 99.69±0.40 for simultaneous equation method and 99.23±0.34 and 99.56±0.16 for absorption ratio method for Cefadroxil and Probencid respectively. These methods are simple, accurate and rapid; those require no preliminary separation and can therefore be used for routine analysis of both drugs in quality control laboratories.

Keywords: Cefadroxil, Probencid, Q-analysis spectrophotometric, Simultaneous equation method.

INTRODUCTION

Cefadroxil chemically a 7-[[2-amino -2-(4-hydroxyphenyl) acetyl] amino]-3-methyl- 8-oxo-5-thia-1-azabicyclo [4.2.0] oct-2-ene-2-carboxylic acid Cefadroxil [1].

Chemical structure of Cefadroxil

Cefadroxil, a first-generation cephalosporin antibiotic, is used to treat urinary tract infections, skin and skin structure infections, pharyngitis and tonsillitis. Like all beta-lactam antibiotics, cefadroxil binds to specific penicillin-binding proteins (PBPs) located inside the bacterial cell wall, causing the inhibition of the third and last stage of bacterial cell wall synthesis. Cell lysis is then mediated by bacterial cell wall autolytic enzymes such as autolysins; it is possible that cefadroxil interferes with an autolysin inhibitor. Literature survey revealed that cefadroxil was qualitatively assayed in biological fluids either individually or in presence of other antibacterial drugs using liquid chromatography 5, other new methods and using hydrotope are also there for the determination of cefadroxil [2-4].

Probenecid is an uricosoronic agent used in gout therapy. When Cefadroxil is co-administered with Probenecid, the renal excretion of Cefadroxil is inhibited. The combination is used in gastrointestinal tract and respiratory tract infections. The prototypical uricosuric agent. It inhibits the renal excretion of organic anions and reduces tubular reabsorption of urate. Probenecid has also been used to treat patients with renal impairment, and, because it reduces the renal tubular excretion of other
drugs, has been used as an adjunct to antibacterial therapy. [PubChem] . The mechanism by which probenecid inhibits renal tubular transport is not known, but the drug may inhibit transport enzymes that require a source of high energy phosphate bonds and/or non-specifically interfere with substrate access to protein receptor sites on the kidney tubular.

Chemical structure of Probencid

EXPERIMENTAL

Pharmaceutically pure samples of CEF and PROB were obtained as gifts from Curex Pharmaceuticals Industries Ltd Jalgaon. Methanol was used as solvent in the study. Double beam UV/Vis spectrophotometer Shimadzu model 1800 with a pair of 10mm matched quartz cells was used to measure absorbance of the resulting solution.

Preparation of standard stock solution

Accurately 10 mg each of CEF and PROB was weighed separately and transferred to two different 100ml volumetric flask. Volume was made up to the mark with Methanol. The standard stock solutions (100μg/ml) were further diluted separately to obtain working standard of concentration 10μg/ml of CEF and PROB each.

Study of spectra and selection of wavelengths:

Each working standard solution was scanned between the range 200-400 nm in 1 cm cell against blank. Maximum absorbing wavelength of CEF and PROB were selected from spectral data and isobestic wavelength selected from overlain spectra of zero order. The λ max for CEF, PROB and isobestic point was 233nm, 247nm and 242nm respectively.

Method I

In quantitative estimation of two components by simultaneous equation method, absorbances were measured at the maximum absorption wavelengths of two drugs. From the spectra of CEF and PROB absorbances were measured at selected wavelengths i.e. 233nm (λ1) and 247nm (λ2) the maximum absorption of CEF and PROB respectively. The absorptivity coefficients of each drug at both wavelengths were determined. The concentration of each drug in laboratory mixture and tablet formulation was determined by substituting the absorbance and Absorptivity coefficient in the following sets of equations.

Validating the results

The methods were validated with respect to linearity, limit of detection (LOD), limit of quantification (LOQ), precision, accuracy and ruggedness. To study accuracy of the developed methods, recovery studies were carried out using standard addition method at three different levels. Percent recovery and low relative standard deviation for six replicates of sample solution was less than 2%, which met the acceptance criteria established for spectrophotometric methods. Ruggedness of the proposed
method was determined by analysis of sample solution prepared by proposed methods between different days. The percent relative standard deviation was found to be less than 2% showed ruggedness of the spectrophotometric methods. The results obtained are summarized in Tables

The overlain spectra of CEF and PROB exhibit \( \lambda_{\text{max}} \) of 233 nm and 247 nm for CEF and PROB respectively which are quite separated from each other. Additionally one is absorptive point was observed at 243 nm. This wavelength was selected for simultaneous estimation of

CEF and PROB for Q value analysis and it is assumed to be sensitive wavelength. Standard calibration curves for CEF and PROB were linear with correlation coefficients \( (r) \) values in the range of 0.9933 – 0.9956 at all the selected wavelengths and the values were average of three readings with standard deviation in the range of 0.16 – 0.34. The calibration curves were repeated three times in a day and the average % RSD was found to be 0.034 for CEF and 0.16 for PROB; similarly the method was repeated for three different days and average % RSD was found to be 0.033 for CEF and 0.17 for PROB. The accuracy of the methods was confirmed by recovery studies from tablet at three different levels of standard additions; recovery in the range of 99.76 – 100% justifies the accuracy of method.

Table 1. Linear regression analysis of calibration curves with their respective absorptivity values:

| Parameter                        | Method First | CEF  | PROB | Method Second | CEF  | PROB |
|----------------------------------|--------------|------|------|---------------|------|------|
| Correlation coefficient \((r)\)  | CEF          | 0.9978| 0.9921| CEF           | 0.9959| 0.9995|
| Molar Absorptivity (lit/mole/cm) | CEF          | 1.2202| 0.0752| CEF           | 1.2202| 0.0752|
| Slope                            | CEF          | 0.0421| 0.0327| CEF           | 0.0293| 0.0308|
| Intercept                        | CEF          | 0.0094| 0.0165| CEF           | 0.0016| 0.0092|

Table 2. Results of recovery studies

| Level of recovery % | Amount of pure drug added (mg) | Simultaneous equation method % recovery | Absorbance ratio method % recovery |
|---------------------|--------------------------------|----------------------------------------|----------------------------------|
|                     | CEF  | PROB | CEF  | PROB | CEF  | PROB | CEF  | PROB |
| 80                  | 200  | 200  | 99.44| 99.72| 99.40| 99.67|
| 100                 | 250  | 250  | 99.35| 99.65| 99.23| 99.89|
| 120                 | 300  | 300  | 99.50| 99.72| 99.89| 99.56|
| Mean % recovery     |      |      | 99.43| 99.69| 99.23| 99.56|
| SD*                 | 0.75 | 0.040| 0.34 | 0.16 |
| CV**                | 0.75 | 0.040| 0.034| 0.016|

*SD = Standard deviation ** CV = coefficient of variance
Table 3. Results of analysis of tablet formulation

| Drugs | Simultaneous equation method | Absorbance ratio method |
|-------|-----------------------------|-------------------------|
|       | % ± SD(n=6)                 | % ± SD(n=6)             |
| CEF   | 100.01±0.017                | 100.06±0.015            |
| PROB  | 100.02±0.088                | 100.02±0.012            |

Table 4. Results of intermediate precisions:

| Day    | Method I          | Method II       |
|--------|-------------------|-----------------|
|        | % Label claim estimated (Mean ±%R.S.D*) | % Label claim estimated (Mean ±%R.S.D*) |
|        | CEF | PROB | CEF | PROB |
| Intraday | 99.2±0.15 | 99.5±0.15 | 99.5±0.16 | 99.67±0.12 |
| Interday | 99.6±0.55 | 99.9±0.40 | 99.3±0.015 | 99.43±0.14 |

*R.S.D. = Relative standard deviation

CONCLUSION

The proposed UV spectrophotometric methods are a simple, accurate, precise, rapid and economical for the simultaneous estimation of CEF and PROB in tablet dosage form. The proposed methods use inexpensive reagents, solvents and instruments that are available in laboratories. Hence, these methods can be conveniently adopted for the routine analysis in quality control laboratories.

ACKNOWLEDGEMENT

Authors are thankful to the Curex Pharmaceutical Jalgaon for giving gift samples of CEF and PROB respectively. Also thankful to Principal of Shree Sureshdada Jain institute of Pharmaceutical Education and Research, Jammer for performing experimental Work.

REFERENCES

1. Anonymous. www.drugbank.ca/drugs/DB01140,
2. Shukla Ravi, Patel Asha, Soni M, Modi Vishesh and Jaliwala Y. Quantitative spectrophotometric estimation of cefadroxil using hydrotrropic solubilization technique. Asian journal of pharmaceutics, 2, 2008, 4-8.
3. Blanchin MD, Kok WT and Fabre H. New detection modes for the determination of cephalosporins and their decomposition products. Chromatographia, 24(1), 1987, 625-627.
4. Badawy SS, Abdel-Gawad FM and Ibrahim MM. Spectrophotometric Studies on Determination of Cefadroxil with Copper(II) and Vanadium(V) in Sulphuric Acid Medium. Analytical letters, 26(3), 1993, 487-497.
5. Butler D: Wartime tactic doubles power of scarce bird-flu drug. Nature, 438(7064), 2005.