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Validated Derivative Spectrophotometric Methods for the Analysis of Lincomycin in Bulk and Dosage Forms

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Abstract: Spectrophotometric methods (zero-order, first order and second order derivative) were evaluted for the analysis of lincomycin in bulk and pharmaceutical formulation. The first and second derivative methods were found more suitable for the analysis as zero order spectrum showed non-defined λ_{max}. Regression analysis for first and second derivative obeyed Beer's law over the concentration range 3-15µg/ml with good correlation coefficient (not less than 0.999). The two methods were proved to be simple and sensitive (LOD and LOQ: 0.676µg/ml and 2.28µg/ml, respectively). Selectivity was reflected by the obtained recovery percentage results (100.3 ± 1.3%; n=3).

Keywords: Lincomycin, Derivative, Spectrophotometry.

INTRODUCTION

The acceptability of analytical method corresponds directly to the performance characteristics that indicate its suitability and reliability for the intended analytical applications. From the commencement of official pharmaceutical analysis, analytical assay methods were included in the compendial monographs with the aim to characterize the quality of bulk drug materials by setting limits of their active ingredient content. In recent

years, the assay methods in the monographs include titrimetric, spectrometry, chromatography, and capillary electrophoresis; also the electro analytical methods can be seen in the literature [1].

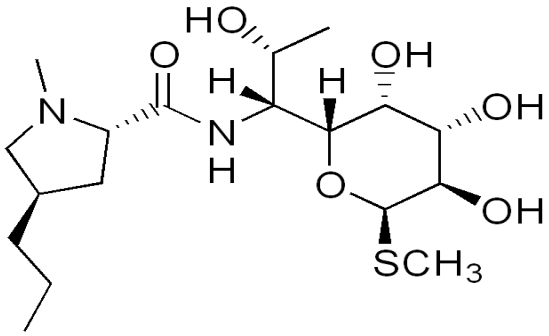

Figure 1: Lincomycin structure.

Spectroscopy is a most powerful tool available for the study of atomic and molecular structure/s and is used in the analysis of wide range of samples. It is the branch of science dealing with the study of interaction between electromagnetic radiation and matter. Derivative spectrophotometry is an analytical technique which consists in differentiating normal spectrum by mathematical transformation of spectral curve into a derivative (first–or higher derivatives). This technique usually improves resolution bands, eliminates the influence of background or matrix and provides more defined fingerprints than traditional ordinary or direct absorbance spectra, since it enhances the detectability of minor spectral features [2].

Lincomycin (**Figure 1**) is a lincosamide antibiotic, primarily used in animals to treat gram-positive and anaerobic bacterial infections.

It is particularly useful to treat infections in animals that are allergic to penicillins and cephalosporines [3]. Literature review revealed many chromatographic and spectrophotometric methods for the determination of lincomycin in pharmaceutical formulations and biological fluids [4-9]. Most of the reported methods are either lack sensitivity or require sophisticated instruments. Therefore, the aim of our study was to develop simple, sensitive and accurate derivative spectrophotometric methods suitable for the routine analysis of lincomycin in bulk and pharmaceutical formulations.

EXPERIMENTAL

Instruments and apparatus: All UV measurements were done on UV-1800. Model UV-1800ENG240V, Shimadzu Corporation, Koyoto, Japan. Weighing was done using Balance: Kern ALS 120-4, Germany.

MATERIALS

Standard and sample: Lincomycin standard were kindly provided by SFDA (Saudi food and drug administration), KSA. Lincocin® injection solution, 600mg/2ml, Pfizer pharmaceutical USA.

Preparation of stock solution: Distilled water was the diluent solvent used in all the experimental work.

Lincomycin standard solution: An accurately weighed quantity of lincomycin standard (300 mg) was dissolved in 30ml distilled water. The solution was transferred into 100 ml volumetric flask and volume was

then completed to mark with the diluent solvent. 1 ml of the resultant solution was further diluted to 100 ml (solution A; 30µg/ml)

Sample solution: One ml of lincomycin injection solution was accurately pipetted and transferred into100 ml volumetric flask. The volume was completed to mark with the solvent. 1 ml was further diluted to 100 ml with the solvent (solution B; 30µg/ml).

Procedures:

UV spectra of lincomycin standard: The zero, 1st and 2nd derivative spectra of lincomycin were recorded over the range 200-400 nm.

Construction of calibration curves: Serial dilutions were made from solution A by transferring accurately measured volumes (1–5mL) to a set of 10mL volumetric flasks. The volumes were completed to mark with diluent to obtain concentration range 3-15 µg/ml. The zero, 1st and 2nd derivative spectra were recorded over the range 200-400 nm against blank (the diluent). The calibration curve was constructed by plotting the obtained absorbance values against drug concentrations. The above procedure was repeated using solution B instead of A to construct the sample calibration curve.

Assay: Three different absorbance values for lincomycin sample were divided by its respective value of standard absorbance to get three values of content percent, then the mean and SD were calculated**.** Alternatively, the slope ratio method for the constructed curves was applied to determine the content percent.

Validation of the proposed method

Linearity: LOD, LOQ, slope error and intercept error were calculated from the concentration range, absorbance data and slope [10].

Accuracy: Recovery percentage was calculated as following**,** 3ml of solution B was mixed with 3ml of solution A. The volume was completed to 10 ml with diluent. 3ml of each solution A and B were transferred to separate stoppered glass tubes and the volumes were completed to 10ml with diluent. The absorbance of the above solutions was measured and the recovery was calculated from the following equation [11]:

$$Recovery = \frac{\text{Amix} - Asam}{Astd}$$

Where A_{mix} = total absorbance of mixture (standard + sample solution), A_{Sam} = absorbance of sample solution, and A_{Std} = absorbance of standard solution. The t-value was also calculated at 95% confident limit for two degrees of freedom using the following formula [12] and compared to the tabulated one.

$$\text{t-value} = (x - \mu).\ \sqrt{n}\ /\ s$$

Where u= known true mean (considering injection content as 100%),

x=mean content of the sample,

n= number of samples,

SD= standard deviation of the assay

Precision: The reproducibility and repeatability of the developed method were obtained by the follow-up of within-day and between-day data for four concentrations within the linearity range. The RSD% value of each concentration was calculated.

RESULTS AND DISCUSSION

Lincomycin standard solutions were analyzed using zero order and derivative spectrophotometry. Better resolution was obtained with first and second derivative using distilled water as solvent. Maximum absorbance wavelength of lincomycin were observed at 194 nm. 206 nm and 212 nm in Zero order, first order and second order spectrum respectively, (**Figures 2, 3** & **4**)

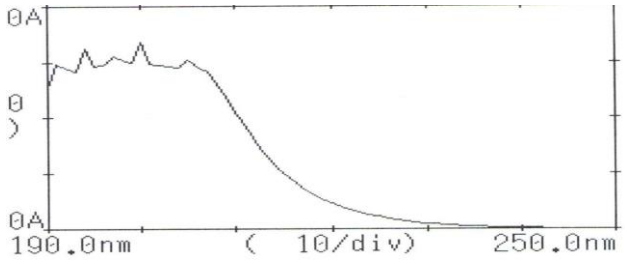

Figure 2: Zero-order Spectrum of lincomycin (12µg/ml, 194nm).

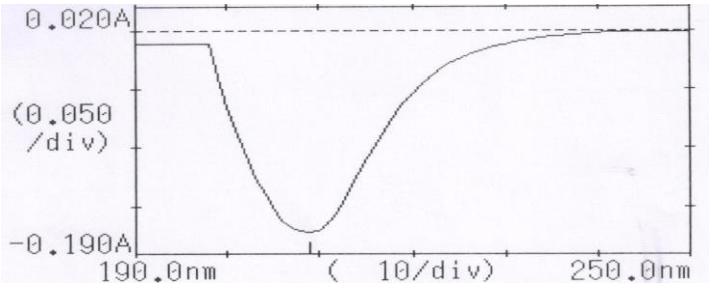

Figure 3: First derivative spectrum of Lincomycin (12µg/ml, 208).

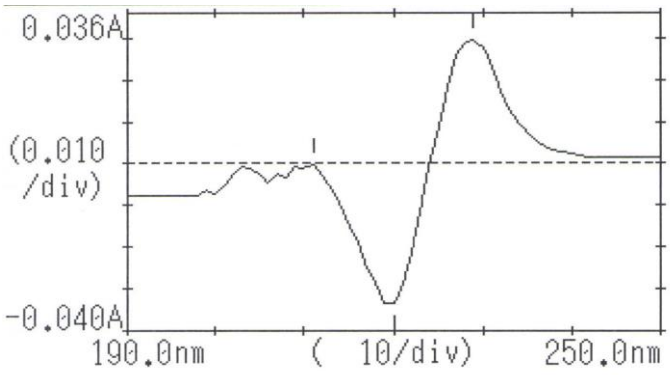

Figure 4: Second derivative spectrum of Lincomycin (12µg/ml, 230nm).

VALIDATION OF THE DEVELOPED METHOD

Linearity: The calibration curve was constructed using the developed method. It was found to obey Beer's law within the concentration range of 3-15 µg/ml with correlation coefficient values not less than 0.999. The regression analysis data was calculated at 95% confidence (**Table-1**).

Table-1: Linearity data of the developed spectrophotometric methods.

Parameter	First derivative	Second derivative
λ_{max}	208nm	230nm
Concentration range	3-15µg/ml	3-15µg/ml
Slope ± ts_b	0.0173 ±0.0013	0.0026 ±0.0003
Intercept ± ts_a	0.00019 ±0.012	-0.0003 ±0.0023
LOD	0.676µg/ml	0.693 µg/ml
LOQ	2.28 µg/ml	2.31 µg/ml
R	0.999	0.999

The results obtained (low value of slope and intercept errors, low SB values of response and LOD and LOQ values) reflected the sensitivity and the reliability of the method. The slope consistency was calculated at 95% confidence limit.

Accuracy: The accuracy of the procedure and freedom of interference by the excipients were confirmed by the results obtained for recovery testing of added amount of authentic drugs to sample solutions in the ratio of 1:1. The obtained results showed good recovery (100.3 ± 1.3; n=3), which indicates the accuracy of the developed methods.

As the calculated t- value at 95% confidence limit (0.14) was less than the tabulated one (4.3), the developed methods proved to be accurate.

Precision: The precision of the developed methods was confirmed by the calculated RSD values, which were found to be within the accepted limit (less than 2%) (**Table-2**)

Table-2: Precision data as evaluated by RSD%.

Concentration µg/ml	Within day precision RSD%; n =3	Between days precision RSD%; n =3
6	1.30	1.30
9	0.90	1.10
12	0.30	0.26

Assay: The developed derivative spectrophotometric method was applied for the drug uniformity testing in lincomycin injection where good assay results were obtained (99.90 ± 1.46, n = 3).

CONCLUSION

The developed methods were proved to be applicable for lincomycin routine analysis in bulk and dosage forms. It was validated according to VICH guidelines and found to be simple, rapid and cost-effective. The developed methods are under investigation to evaluate their stability indicating properties to study the stability of lincomycin under stress conditions.

REFERENCES

1. Z.A. Al Othman and N. Rahman. *Arabian Journal of Chemistry*. 2013. (http://dx.doi.org /10.1016/ j.arabjc.2013.04.016)
2. F.S. Rojas and C.B. Ojeda. *Anal Chim Acta*. 2009, **635** (1), 22-44.
3. Drugs.com (www.drugs.com/cdi/lincomycin.html).
4. S. Naveed, S. Naz Shah, F. Qamar, N. Waheed and Safeena Nazeer. *International Journal of Pharm. Drug Dev*. 2014, **1** (2), 10-12.
5. P. Rajeevkumar and N. Subramanian. *International Journal of Chem Tech Research*. 2010, **2** (4), 2052-2055.
6. M. Dousa, Z. Sikac, M. Halama, K. Lemr. *Journal of Pharmaceutical and Biomedical Analysis*. 2006, **4**: 981-986.
7. K. Vučićević-Prčetić, R. Cservenák, N. Radulović. *Journal of pharmaceutical and biomedical analysis*. 2011, **4**: 736-742.
8. P. Czarniak, M. Boddy and J.D. Hughes. *Journal of drug design, development and therapy*. 2016, **10**: 1029-1034.
9. M.N. Abualhasan, N. Batrawi, O.B. Sutcliffe and A. Zaid. *Scientia Pharmaceutica*. 2012, **4**: 977-986·
10. S.W. Shantier and E.A. Gad Kariem. *Elixir Pharmacy*. 2013, **59**: 15471-15473.
11. M.M. Elimam, S.W. Shantier, E.A. Gadkariem, M.A. Mohamed. *Int J Pharm Pharm Sci*. 2016, **8** (4): 347-349.
12. S.W. Shantier, E.A. Gadkariem and R. Mohamed. *World Journal of Pharmaceutical Research*. 2015, **4** (4): 234-240.

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