

SIMULTANEOUS ESTIMATION OF MOXIFLOXACIN HCL AND PREDNISOLONE ACETATE FROM EYE DROPS

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ABSTRACT

Moxifloxacin HCl (MOX) and Prednisolone acetate (PRD) are combined in ophthalmic formulations to provide antibacterial and anti-inflammatory activity, respectively. The present study describes the development and validation of two simple UV-visible spectrophotometric procedures for their simultaneous estimation in bulk drug mixtures and eye-drop formulation. A Shimadzu UV-1800 spectrophotometer was used. Standard solutions were prepared in methanol and diluted with distilled water. Overlaid spectra were used to select analytical wavelengths. For the simultaneous-equation method, 288 nm and 247 nm were selected as the respective λ_{max} values of MOX and PRD. Both analytes showed linear responses over 2–12 $\mu\text{g/mL}$. Specific absorptivity values were established and used to construct simultaneous equations for calculation of both

components. For the Q-absorbance-ratio method, the thesis reports an isoabsorptive wavelength in the vicinity of 273–277 nm; the method was applied together with the individual drug maxima. The methods were assessed using assay, recovery, intra-day precision, inter-day precision, repeatability and ruggedness. In the marketed eye-drop sample, the simultaneous-equation procedure gave 99.93% of label claim for MOX and 100.77% for PRD, while the Q-absorbance-ratio procedure gave 100.44% and 101.01%, respectively. Recovery for the simultaneous-equation method was reported as 100.44% for MOX and 100.11% for PRD; the Q method gave 100.25% and 99.90%, respectively. Reported precision values were below 2% RSD. The study therefore demonstrates that both procedures can be used for routine simultaneous assay of the selected ophthalmic combination where UV spectrophotometric analysis is appropriate.

KEYWORDS: Moxifloxacin hydrochloride; Prednisolone acetate; UV-visible spectrophotometry; Simultaneous equation; Q-absorbance ratio; Ophthalmic formulation.

1. INTRODUCTION

Pharmaceutical analysis is essential for establishing the identity, strength, purity and quality of active pharmaceutical ingredients and finished dosage forms. The thesis emphasizes that quantitative analytical methods are particularly important in pharmaceutical manufacturing because raw materials and finished products must meet predefined quality requirements. Instrumental methods such as UV-visible spectrophotometry and chromatographic techniques are widely used because they can provide reproducible quantitative measurements.

Moxifloxacin hydrochloride is a fourth-generation fluoroquinolone antibacterial agent. Its antibacterial activity is associated with inhibition of bacterial DNA gyrase and topoisomerase IV. Prednisolone acetate is a corticosteroid used for its anti-inflammatory activity. In an ophthalmic combination, the two components address different therapeutic aspects of an eye condition. The thesis identifies the combination as an ophthalmic formulation available in the Indian market and proposes simultaneous analytical estimation as a quality-control requirement.

For multicomponent formulations, individual estimation may require separate analytical procedures or chromatographic separation. UV-visible spectrophotometry offers a comparatively simple alternative when the absorption characteristics of the components permit mathematical resolution. The thesis therefore investigated simultaneous-equation and Q-absorbance-ratio approaches for MOX and PRD.

2. OBJECTIVE

The objective was to develop and validate simple, precise and accurate UV-visible spectrophotometric methods for simultaneous estimation of moxifloxacin hydrochloride and prednisolone acetate in bulk mixture and ophthalmic dosage form. The study specifically evaluated linearity, accuracy/recovery, precision, repeatability and applicability to a marketed eye-drop formulation.

3. MATERIALS AND METHODS

3.1 Instrumentation and reagents

A Shimadzu UV-1800 UV-visible spectrophotometer was used. Moxifloxacin hydrochloride and prednisolone acetate reference materials were used for preparation of standards.

Methanol and distilled water were used for solution preparation. The marketed ophthalmic product evaluated in the thesis was identified as Combace, with an average volume of 5 mL.

3.2 Standard stock solutions

Accurately weighed 10 mg quantities of MOX and PRD were transferred separately to 100-mL volumetric flasks containing approximately 50 mL methanol. The solutions were diluted to volume with distilled water to obtain 100 µg/mL stock solutions of each drug.

3.3 Spectral study and wavelength selection

Aliquots of the individual stock solutions were diluted to 10 µg/mL and scanned from 400 to 200 nm against the solvent blank using a 1-cm cell. The overlain spectra were used to select wavelengths. The experimental method section identifies 288 nm as the λ_{max} of MOX and 247 nm as the λ_{max} of PRD for the simultaneous-equation method. The thesis contains a minor inconsistency in later narrative descriptions of the maxima and Q-method isoabsorptive wavelength; therefore, this article follows the wavelengths specified in the method-development section and tabulated experimental work.

3.4 Linearity

For each drug, standard solutions covering 2, 4, 6, 8, 10 and 12 µg/mL were prepared in distilled water. Absorbance was measured at the selected wavelength and calibration relationships were assessed from absorbance versus concentration. The thesis reports linearity over 2–12 µg/mL for both analytes.

3.5 Simultaneous-equation method

Specific absorptivity, $E(1\%, 1\text{ cm})$, values were determined at both selected wavelengths. The mean values reported for the simultaneous-equation study were approximately 922.4 and 830.2 for MOX at 288 and 247 nm, respectively, and 601.8 and 854.6 for PRD at the same wavelengths. For a two-component mixture, absorbance at each wavelength is represented as the sum of the absorbances contributed by MOX and PRD. The two equations were solved simultaneously to obtain the concentrations of the individual components.

3.6 Q-absorbance-ratio method

The second procedure used the absorbance-ratio principle. The thesis reports an isoabsorptive point around 273–277 nm in different sections; the detailed Q-method table uses 273 nm, while the concluding narrative mentions 273.0 nm and an earlier results paragraph mentions

277 nm. The method combines absorbance at the isoabsorptive wavelength with absorbance at a drug-specific maximum to calculate the concentrations of both components. Because the source contains these wavelength inconsistencies, the numerical results are reported as documented rather than silently corrected.

3.7 Sample preparation

For bulk-mixture analysis, quantities equivalent to 10 mg MOX and 20 mg PRD were dissolved in methanol and diluted with distilled water. An aliquot was further diluted to obtain approximately 4 µg/mL MOX and 8 µg/mL PRD. For the marketed eye drops, a measured volume equivalent to 5 mg MOX and 10 mg PRD was transferred to a 100-mL volumetric flask containing 50 mL methanol, shaken for 20 min, diluted with distilled water and filtered through Whatman No. 41 paper. The filtrate was diluted to approximately 4 µg/mL MOX and 8 µg/mL PRD before spectrophotometric measurement.

3.8 Method validation

Validation was performed using linearity, accuracy, precision and ruggedness. Accuracy was assessed by standard addition at 80%, 100% and 120% levels. Intra-day precision was evaluated at multiple concentrations on the same day, and inter-day precision over consecutive days. Repeatability was assessed using six replicate measurements. The thesis reports that the procedures were also examined for operator-related ruggedness.

4. RESULTS AND DISCUSSION

4.1 Spectral characteristics and linearity

The individual standard spectra permitted selection of wavelengths suitable for simultaneous analysis. The calibration data show a progressive increase in absorbance with concentration for both compounds. For MOX at 288 nm, mean absorbance increased from 0.195 at 2 µg/mL to 1.107 at 12 µg/mL. For PRD at 247 nm, absorbance increased from 0.141 at 2 µg/mL to 1.098 at 12 µg/mL. The replicate %RSD values in the calibration data were low, generally below 0.8%, indicating good measurement repeatability within the calibration experiment.

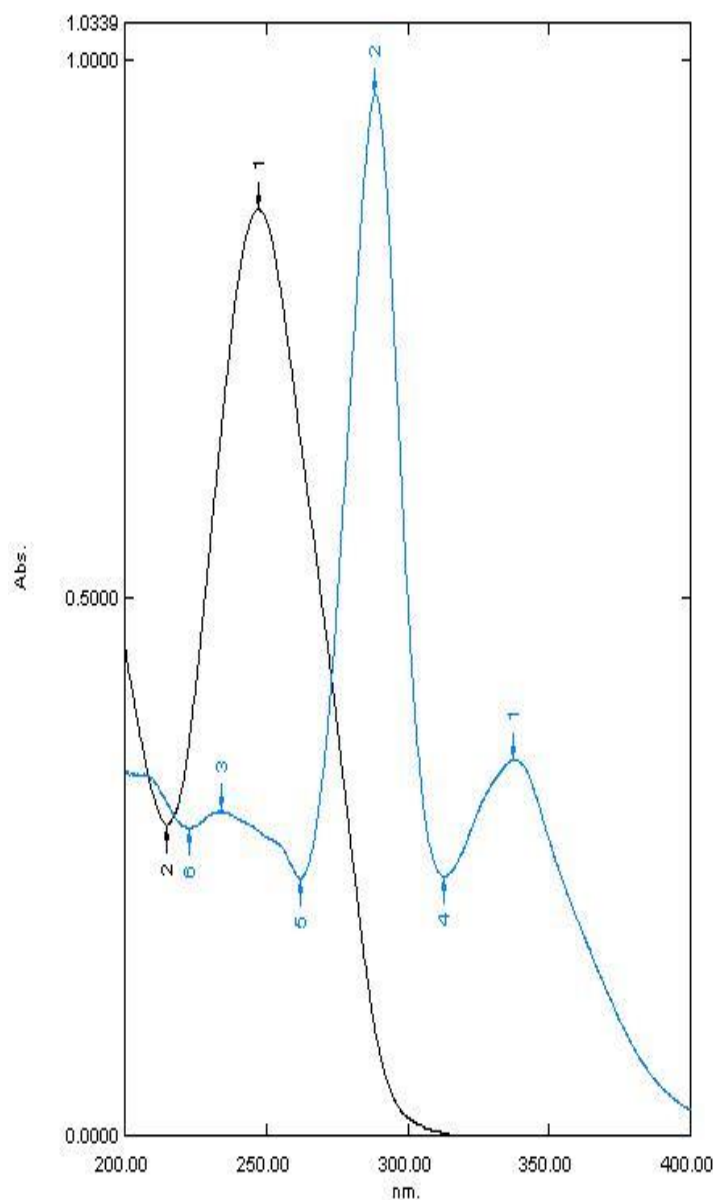


Figure 2: Overlain UV spectra of the two analytes from the source thesis, showing the spectral characteristics used for method development.

Figure note: The figures are reproduced from the submitted thesis results/method-development material; numerical values and wavelength assignments in the manuscript follow the thesis tables and detailed method section.

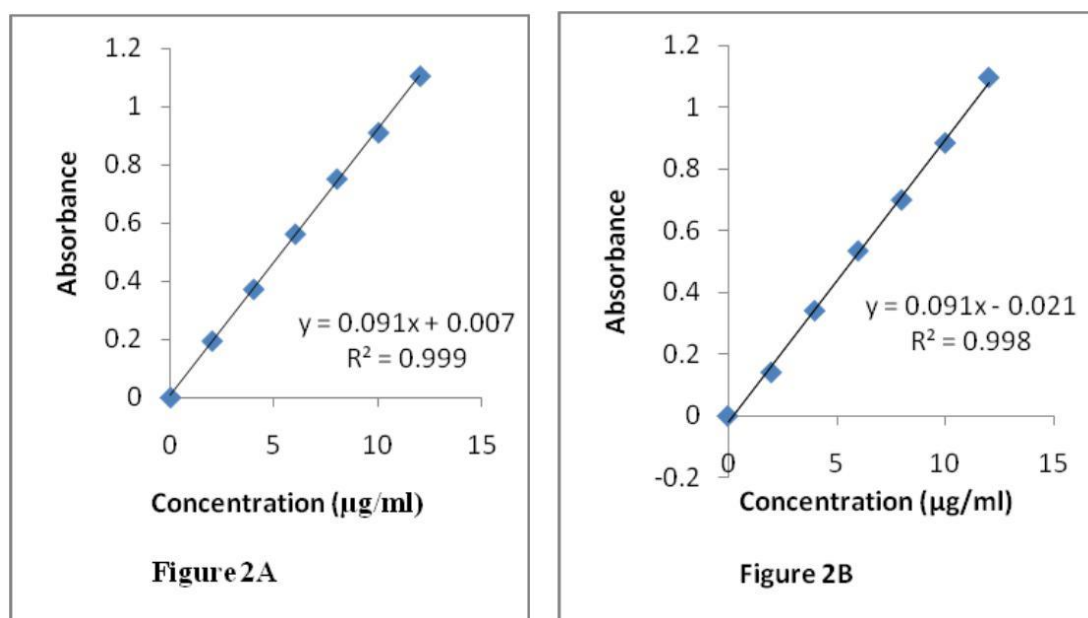


Figure 1: Calibration curves for moxifloxacin hydrochloride (MOX) and prednisolone acetate (PRD) reported in the source thesis.

Drug	Wavelength	Range (µg/mL)	Representative absorbance range
MOX	288 nm	2–12	0.195–1.107
PRD	247 nm	2–12	0.141–1.098

The study therefore established a common working range of 2–12 µg/mL for both analytes. This range encompasses the diluted concentrations used for analysis of the eye-drop formulation.

4.2 Simultaneous-Equation Method: Bulk Mixture And Formulation

In a laboratory mixture containing 4 µg/mL MOX and 8 µg/mL PRD, the simultaneous-equation method gave mean estimated amounts of 4.05 and 8.10 µg/mL, corresponding to 101.28% and 101.25%, respectively. The reported RSD values were low, supporting reproducibility of the calculation procedure.

Application to the marketed eye-drop sample gave a mean assay of 99.93% of label claim for MOX and 100.77% for PRD. These values were close to the stated formulation strengths. The corresponding mean standard deviations were 1.123% and 0.629% when expressed as percentage of label claim, with reported %RSD values of 1.124% and 0.624%, respectively.

Parameter	MOX	PRD
Marketed formulation assay (% label claim)	99.93	100.77

Recovery (overall reported value, %)	100.44	100.11
Intra-day precision (%RSD)	0.63	0.36
Inter-day precision (%RSD)	1.02	0.39
Repeatability (%RSD)	0.73	0.89

4.3 Accuracy and precision

Recovery experiments were performed by adding known quantities of standard at 80%, 100% and 120% levels to preanalysed samples. The individual recovery values reported in the source for MOX were 99.10%, 100.83% and 101.40%, while PRD showed 98.95%, 101.66% and 99.72%. The thesis summarizes the overall recovery as 100.44% for MOX and 100.11% for PRD. The associated RSD values were within the stated acceptance expectation of less than 2%.

Precision data likewise remained below 2% RSD. The reported intra-day values were 0.63% for MOX and 0.36% for PRD, while inter-day values were 1.02% and 0.39%, respectively. Repeatability values were 0.73% and 0.89%. These results indicate limited variation under the tested conditions.

4.4 Q-absorbance-ratio method

The second method was developed using the absorbance-ratio concept. The thesis reports MOX and PRD assay values of 100.44% and 101.01% of label claim, respectively, in the ophthalmic formulation. Recovery was reported as 100.25% for MOX and 99.90% for PRD. Precision was also below 2% RSD, with intra-day values of 0.32% and 0.43%, inter-day values of 0.23% and 0.45%, and repeatability values of 0.34% and 0.39% for MOX and PRD, respectively.

Parameter	MOX	PRD
Marketed formulation assay (% label claim)	100.44	101.01
Recovery (%)	100.25	99.90
Intra-day precision (%RSD)	0.32	0.43
Inter-day precision (%RSD)	0.23	0.45
Repeatability (%RSD)	0.34	0.39

4.5 Comparison of the two procedures

Both procedures produced assay results close to 100% of label claim and recovery values close to 100%. The simultaneous-equation method uses the two individual λ_{max} wavelengths and the measured absorptivity coefficients, whereas the Q method uses an isoabsorptive

wavelength together with a component-specific wavelength. The two approaches therefore provide mathematically different ways of resolving the same two-component UV response without chromatographic separation.

The reported precision values indicate that both procedures were reproducible in the tested laboratory conditions. The thesis describes both as simple, precise and accurate for simultaneous determination. Because the study was designed around UV-visible measurements, the methods may be attractive where routine assay needs do not require chromatographic separation; however, the thesis does not establish that either method is stability-indicating or suitable for resolving degradation products.

5. CONCLUSION

Two UV-visible spectrophotometric procedures were developed for simultaneous estimation of moxifloxacin hydrochloride and prednisolone acetate in bulk mixture and ophthalmic dosage form. The simultaneous-equation method used 288 and 247 nm and showed linearity from 2 to 12 µg/mL for both drugs. In the marketed eye-drop formulation, the method yielded 99.93% MOX and 100.77% PRD of label claim, with recovery and precision results within the limits reported in the thesis. The Q-absorbance-ratio method provided assay values of 100.44% and 101.01% for MOX and PRD, respectively, with similarly low variability.

Taken together, the source study supports the use of these UV spectrophotometric approaches as straightforward quantitative procedures for the selected combination under the investigated conditions. The principal analytical contribution is the simultaneous resolution of two absorbing components without a chromatographic separation step. Future work, if required, should independently confirm the wavelength selection and Q-method isoabsorptive point, and may include specificity against degradation products, expanded robustness testing and comparison with a validated chromatographic reference method.

6. LIMITATIONS

The present article is a condensed research manuscript based strictly on the submitted thesis. The source contains a few internal inconsistencies in the description of wavelength assignments and the Q-method isoabsorptive wavelength (273 versus 277 nm in different sections). These have been explicitly identified rather than reconciled by inference. The source also describes validation principally through linearity, accuracy, precision, repeatability and ruggedness; detailed LOD/LOQ and stability-indicating results are not

presented in the reported experimental results summarized here. Accordingly, this article does not claim broader validation characteristics beyond those documented.

7. REFERENCES

1. Dewani AP et al. Development and Validation of RP-HPLC Method for the Determination of Moxifloxacin in Presence of Its Degradation Products. *American-Eurasian Journal of Scientific Research*. 2011; 6(4): 192–200.
2. Sahu SK et al. Spectrophotometric Estimation of Moxifloxacin in Bulk and its Pharmaceutical Formulations. *Pharmacologyonline*. 2011; 2: 491–502.
3. Lobna MA et al. Development and Validation of Spectrophotometric, Atomic Absorption and Kinetic Methods for Determination of Moxifloxacin Hydrochloride. *Analytical Chemistry Insights*. 2011; 6: 67–78.
4. Ashok R et al. Development and validation of analytical method for estimation of prednisolone in bulk and tablets using UV-Visible spectroscopy. *International Journal of Pharmacy and Pharmaceutical Sciences*. 2011; 3(4): 184–186.
5. Dewani AP et al. Absorption ratio method for the estimation of moxifloxacin HCl and ketorolac tromethamine in their combined dosage form by UV-Visible spectroscopy. *International Journal of Pharmaceutical Research & Development*. 2011; 3(7): 21–26.
6. Dhilon V et al. Validated HPTLC method for estimation of moxifloxacin hydrochloride in tablets. *Pharmaceutical Methods*. 2010; 1(1): 54–56.
7. Kurakula M et al. Estimation of Prednisolone in Proliposomal formulation using RP-HPLC method. *International Journal of Research in Pharmaceutical and Biomedical Sciences*. 2011; 2(4): 1663–1669.
8. Chitlange SS et al. Development and Validation of Spectrophotometric and HPLC Method for the Simultaneous Estimation of Salbutamol Sulphate and Prednisolone in Tablet Dosage Form. *Journal of Analytical & Bioanalytical Techniques*. 2011.
9. Malhotra D et al. Validated UV Spectroscopic method for simultaneous estimation of azithromycin and prednisolone. *International Journal of Pharmacy and Pharmaceutical Sciences*. 2011; 3(4): 299–302.
10. Musharraf SG et al. Stress degradation studies and development of stability-indicating TLC-densitometry method for determination of prednisolone acetate and chloramphenicol. *Chemistry Central Journal*. 2012; 6: 7.

11. Pavan P et al. Spectrophotometric Methods for Simultaneous Estimation of Moxifloxacin HCl and Ketorolac Tromethamine. Asian Journal of Research in Chemistry. 2010; 3(3): 767.
12. Najma S et al. High-Performance Liquid Chromatography Assay for Moxifloxacin in Bulk, Pharmaceutical Formulations and Serum. Journal of the Chinese Chemical Society. 2010; 57.
13. Misra M et al. A simple and validated UV-Spectroscopic method for estimation of moxifloxacin in bulk and formulation. Journal of Global Pharm Technology. 2010; 2(6): 21–27.
14. Source note: *This manuscript is condensed from the submitted thesis “Simultaneous Estimation of Moxifloxacin HCl and Prednisolone Acetate from Eye Drops.” The source reports the experimental procedures, validation design and numerical results summarized above.* □turn0file0□L943-L966