Development and Validation of Spectrophotometric Method for Simultaneous Estimation of Lornoxicam and Paracetamol in Tablet Dosage Form by Dual Wavelength Method

Shailendra Bindaiya, Santosh Kushwaha, Ameeta Argal
Department of Pharmaceutical Chemistry, Rajeev Gandhi College of Pharmacy, Bhopal

Abstract

A precise, accurate, sensitive spectrophotometric method has been developed and validated for the simultaneous estimation of Lornoxicam and Paracetamol in tablet formulation. This method was based on UV-spectrophotometric determination of two drugs, using dual wavelength method. It involves measurement of absorbance difference at 286nm and 230.5nm were consider for estimation of Lornoxicam, at 257nm and 282nm were consider for estimation of Paracetamol. in 0.1 N NaOH. The linearity was observed in the concentration range of 8-40 µg/ml & 10-50 µg/ml for Lornoxicam and Paracetamol respectively. The accuracy and precision of the method was determined and validated stastically. The method showed good reproducibility and recovery with % RSD less than 2. The method was validated according to ICH guidelines.

Keywords: Lornoxicam, Paracetamol, Simultaneous estimation, dual wavelength method, Validation

Introduction

Lornoxicam (6-Chloro-4-hydroxy-2-methyl-N-2-pyridinyl-2H-thieno, 3-e]-1, 2-thiazine-3-carboxamide 1, 1-dioxide, 9CI Chlorotenoxicam) is a non-steroidal anti-inflammatory drug (NSAID) of the oxicam class with analgesic, anti-inflammatory and antipyretic properties. It is available in oral and parenteral formulations. Lornoxicam inhibits prostaglandin biosynthesis by blocking the enzyme cyclooxygenase.Lornoxicam inhibits both isoforms in the same concentration range, that is, COX-1 inhibition: COX-2 inhibition = 1. It readily penetrates into the synovial fluid. Synovial fluid: plasma AUC ratio is 0.5 after administration of 4 mg twice daily.

Paracetamol (N- 4-hydroxyphenyl acetamide) is a Analgesic-antipyretic drugs with poor anti-inflammatory action, belongs to Para-aminophenol derivative categories of NSAIDs. The main mechanism of action of paracetamol is considered to be the inhibition of cyclooxygenase (COX), recent findings suggest that it is highly selective for COX-2.Because of its selectivity for COX-2 it does not significantly inhibit the production of the pro-clotting thromboxanes.

Experimental

Apparatus -

Spectrophotometric analysis was carried out on a Shimadzu 1700 double beam spectrophotometer with fixed slit width (2 nm) and 10 mm matched quatz cells.

Chemicals and Reagents -

Lornoxicam and Paracetamol were kindly supplied by Lupin Laboratories Mumbai. A pharmaceutical preparation (label claim Lornoxicam 8 mg and Paracetamol 500 mg) were manufactured and supplied by Lupin Laboratories (Mumbai, India). NaOH (S.D. Fine Chemicals Ltd., Mumbai).

Preparation of calibration curve

Stock solutions were prepared by dissolving Lornoxicam and Paracetamol in 0.1 N NaOH. The standard solutions were prepared by dilution of stock solutions in selected solvent to reach concentration ranges of 8 – 40 and 10 – 50 μg mL-1 for Lornoxicam and Paracetamol respectively.

Assay procedure for tablets -

Twenty tablets (Neucam-P, Lupin Laboratories Mumbai) were weighed and ground to a fine powder. An accurately weighed powder sample equivalent to 0.8 mg of Lornoxicam and 50 mg Paracetamol were transferred to 100 ml of volumetric flask containing 0.1 N NaOH solutions. The flask was sonicated for about 10 min to solubilize the drug and the volume was made up to mark. The solution was filtered through Whatmann filter paper No 41. The filtrate was diluted appropriately with 0.1 N NaOH and was analyzed on UV spectrophotometer.

Results and Discussion

The stability of working solutions of Lornoxicam and Paracetamol was studied by recording their absorption spectra. At first these spectra were measured. No changes in the spectra were observed for at least 48 hours when the solutions were stored at room Temperature in the dark.

The aliquot portions of standard stock solutions of Lornoxicam and Paracetamol were diluted appropriately with 0.1 N NaOH to obtain a concentration 8 μg mL-1 of Lornoxicam and 50 μg mL-1of Paracetamol. They were scanned in the wavelength range of 400−200 nm and the overlain spectrum was obtained (Fig 1). Two wavelengths 286nm and 230.5nm (for estimation of Lornoxicam) and 257nm and 282nm (for estimation of Paracetamol) were selected for the formation Dual wavelength mehod. Since studied, making it difficult to resolve by a standard addition method may prove useful. In t his procedure, a mixture is taken, and the absorbance of the solute under investigation is determined. To a known volume of the original unknown mixture, a measured quantity of pure solute is added and another absorbance of the mixture is taken. The original concentration of the solute may be determined from the increase in the absorbance of the solute. The final expression for C_0, the concentration of the solute in the original solution, is given below:

$$Co = \frac{A_o I_2 C_s V_s}{[A_f I_1 (V_s - V_s - A_0 I_2 V_0)]}$$

Where A_o and A_f are the original and final absorbance of the solute under investigation, Cs is the concentration of the solute in the pure solute solution, I_1 and I_2 are the volumes of the sample for the first and second absorbance, Vo and Vs are the volumes of the original solution and the volume of the pure solute added to the original solution. The effects of matrix interactions are greatly reduced or eliminated by this procedure.

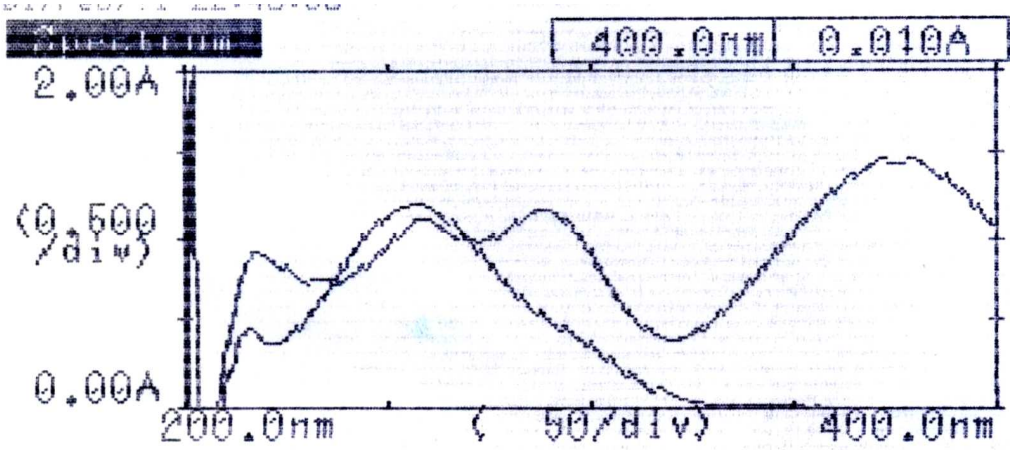

Fig: 1 Overlay spectra of Lornoxicam and Paracetamol

Under the described experimental conditions, the graphs were obtained by absorbance of each drug in this mixture versus concentration, in the range stated in Table 1, show linear relationship. Evaluation of the proposed method was performed by statistical analysis of data, where slopes, intercepts and correlation coefficients were shown in Table 1.

Table 1. Statistical analysis of calibration graph

Parameters	Lornoxicam	Paracetamol
Range (µg/ml)	08 -40	10-50
Regression equation(y)a		
Slope (m)	1.01	1.00
Intercept (c)	0.000015	0.001089
Correlation coefficient (r^2)	0.9997	0.9998

y = mx + c where x is concentration in µg mL-1 and y in absorbance unit.

Intra-day precision was performed by using same procedure as under tablet formulation analysis and absorbance recorded at 2 hours interval within day. Inter-day precision was assessed by analyzing the same samples on different days. The data obtained were within 2% RSD indicating reasonable repeatability of the proposed method which is shown in Table 2.Thus, it was concluded that there was no significant difference on the assay, which was tested on an intra – day and inter – day basis.

Table 2. Intra- and inter-assay precision data

	Lornoxicam		Paracetamol	
Parameters	Mean* ± S.D.	% R.S.D.	Mean* ± S.D.	% R.S.D.

Intraday Precision	97.59 ± 0.318	0.320	95.63 ± 0.267	0.2677
Interday Precision	100.4 ± 0.098	0.0976	99.8 ± 0.0761	0.0762

- Results are mean of three replicates

Recovery studies were performed to a pre-analyzed sample solution, a definite conc. of standard drug was added and then the recovery was studied. Different amount of pure drug solutions were added to final dilution (lornoxicam & paracetamol) sample, and then the solutions were analysed. The experiments were repeated for five times to emphasize validation the result of recovery studies and statistical data in Table 3.

Table 3. Data for recovery studies

Level of recovery	Amount added (μg mL-1)	Amount found (μg mL-1)	*Recovery (%)	Mean ± % R.S.D.
Lornoxicam				
50	8	7.60	95.00	98.33±0.607
100	16	16.09	100.5	
150	24	23.88	99.50	
Paracetamol				
50	10	9.82	98.20	99.50±0.312
100	20	19.89	99.45	
150	30	30.32	101	

* Recovery is mean of three estimations.

The validated method was applied to the determination of lornoxicam & paracetamol in tablet dosage from. The results are summarized in Table 4. The results of assay indicate that the method is selective for the analysis of both Lornoxicam & Paracetamol without interference from the excipients used to formulate and produce these tablets

Table 4. Result of Analysis of Tablet Formulation

Parameters	Lornoxicam		Paracetamol	
	Mean* ± S.D.	% R.S.D.	Mean* ± S.D.	% R.S.D.
	94.78 ± 0.366	0.386	95.40 ± 0.346	0.3624

- S.D. is standard deviation and R.S.D. is relative standard deviation

Conclusion

A simple, rapid, accurate and precise spectrophotometric method has been developed and validated for the routine analysis of Lornoxicam and Paracetamol in API and tablet dosage forms. The developed method is recommended for routine and quality control analysis of the investigated drugs in two component pharmaceutical preparations.

Acknowledgements

The authors are thankful to Lupin Laboratories Mumbai for providing drug samples and RGCP, Bhopal, for providing facilities to carry out this work.

References

becket, a.k., Stenlase, J.B., spectroscopy in: Practical pharmaceutical Chemistry, CBS publication, 4th Ed., Part II, 275-299.

Journal of Chromatography B: Biomedical Sciences and Applications Volume 707, Issues 1-2, 10 April 1998, Pages 151-159.

Kalsi, P.S. Ultraviolet (UV) and Visible Spectroscopy; In: Spectrosocopy of Organic Compounds, 4th Ed.; New Age International Publishers; New Delhi, 2001;7-58.

Kasturi, A.V, Mahadik V R., Principle of spectroscopy In: Pharmaceutical analysis, Nirali Prakashan 9th Ed, 2003, 156-159.

Khandpur, R.S. Colorimeters and Spectrophotometers (Visible-Ultraviolet). In: Hand Book of Analytical instruments, 1st Ed.; Tata McGraw Hill Publishing Company Limited: Delhi, 2000; 92-146.

Silverstein, R.M.; Morril, TY.C.; Bassler, G.C. Ultaviolet spectometer, In: Spectrometric Identification of Organic Compounds, 5th Ed.; John wiley; New York, 1997; 289-315.

Para and ibu *Asian J. Research Chem. 2(1): Jan.-March, 2009, 37* ISSN 0974-4169 www.ajrconline.org.

Para and tinidazole ISSN: 0973-4945; CODEN ECJHAOE-Journal of Chemistry http://www.e-journals.net 2010, 7(1), 260-26.

Validation of Analytical Procedures- Definition and Terminology, FDA's Centre for Veterinary Medicine Guidance Document 63, 1999.