

Simultaneous Spectrophotometric Estimation of Aceclofenac and Paracetamol

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Four accurate, sensitive and economical procedures for simultaneous estimation of paracetamol and aceclofenac in tablet dosage form have been developed. The methods employed are simultaneous equation method, first order derivative method, area under curve method and absorbance ratio method for simultaneous estimation of paracetamol and aceclofenac. The first method employs the formation and solving of simultaneous equation using 244 and 273 nm as the two wavelengths for forming equation. The second method employs first order derivative spectroscopy to eliminate spectral interference. The third method employs selection of area under curve in wavelength region of 242-246 and 271-275 nm and solving the equation. The fourth method employed 266 nm as λ_1 (isobestic point) and 244 nm as λ_2 , which is the λ_{\max} of paracetamol. The drugs obey Beer's law in the concentration range 10-100 $\mu\text{g/mL}$ for aceclofenac and 10-50 $\mu\text{g/mL}$ for paracetamol. The results of analysis have been validated statistically and by recovery studies.

Key Words: UV-Visible spectrophotometry, Paracetamol, Aceclofenac.

INTRODUCTION

Aceclofenac is *o*-(2,6-dichloroanilino)phenyl}acetate glycolic acid ester with antiinflammatory and analgesic properties¹. Few analytical methods are reported which includes titrimetric², colorimetric³⁻⁵, spectrofluorimetric⁴ and spectrophotometric⁶ estimation of aceclofenac in bulk and pharmaceutical dosage formulations. Stability indicating methods are also reported which include RP-HPLC⁵, densitometry^{5,7} and spectrophotometric⁷ estimation of aceclofenac in presence of degradation products. Paracetamol (Para) chemically is 4-hydroxy acetanilide⁸. Several titrimetric⁹, spectrophotometric¹⁰⁻¹⁸, HPLC¹⁹⁻²³ and HPTLC²⁴ methods have been reported for estimation of paracetamol as single component as well as in combination with other drugs. Both the drug find place in official books which suggest different methods for estimation^{1-3,8,9}. Extensive literature survey revealed that no single method available for estimation of aceclofenac

and paracetamol in combined dosage form. The object of present work was to develop simple, rapid, economical and reproducible methods for simultaneous estimation of binary drug formulation.

EXPERIMENTAL

The instrument used in the present study was Jasco double beam UV-Vis spectrophotometer (Model V-530) with fixed slit width 2 nm connected to a computer with spectra manager software. All weighing were done on electronic balance (Model Shimadzu AY 120). All reagents used were of Analytical grade.

Commercial tablets of paracetamol (500 mg) and aceclofenac (100 mg) ACEROC-P (WOCKHRD LTD.) were procured from the local market.

Preparation of standard stock solution: Standard stock solution was prepared by dissolving 50 mg of each drug in 50 mL of methanol to get concentration of 1 mg/mL for both drugs separately. 10 mL of stock solutions were further diluted to 100 mL with phosphate buffer (pH-7, prepared as per I.P.) to get a working solution of concentration 100 µg/mL of paracetamol and aceclofenac, respectively. The solutions were further diluted with phosphate buffer and scanned in the wavelength range 200-400 nm. Both the drugs obey Beer's law within the concentration range of 10-100 µg/mL for aceclofenac and 10-50 µg/mL for paracetamol.

Preparation of sample stock solution: 20 Tablets were weighed accurately and powdered. Powder equivalent to 50 mg of paracetamol was weighed and transferred to 50 mL volumetric flask. In the same flask 40 mg of pure aceclofenac drug was added and dissolved in methanol by shaking the flask for 10 min. The solution was filtered through Whatman filter paper no. 41 and first few mL were rejected. 10 mL of this filtrate was further diluted to 100 mL with phosphate buffer pH -7. Further dilutions were made from this sample stock solution to get required concentration.

Method for preparation of phosphate buffer⁹: Place 50.0 mL of 0.2 M potassium dihydrogen phosphate in a 200 mL volumetric flask, add the 29.1 mL of 0.2 M sodium hydroxide and then add water to make up the volume. The pH of the solution is 7.

Simultaneous equation method (method I): The overlain zero order spectra of paracetamol and aceclofenac showed an absorption maximum of paracetamol is at 244 nm while aceclofenac has absorption maxima at 273 nm. Spectrum of standard solution of paracetamol and aceclofenac having concentration of 20 µg/mL was recorded separately in the range of 200-400 nm. Absorption was determined at wavelengths 244 nm and 273 nm. The absorptivity coefficients at these wavelengths were calculated. The absorptivity of paracetamol is 49.967 (ax_1) at 244 nm (λ_1) and 11.057 (ax_2) at 273 nm (λ_2), while absorptivity of aceclofenac is 13.875 (ay_1) at

244 nm (λ_1) and 19.120 (a_{y_2}) at 273 nm (λ_2). A_1 and A_2 are absorbances of diluted sample at 244 and 273 nm, respectively.

Procedure for analysis of tablet formulation: An aliquot of sample stock solution (2 mL) was transferred to 10 mL standard volumetric flask and volume was made up to mark with phosphate buffer pH 7. The solution was scanned in the range of 200-400 nm against blank. Absorbances were recorded at wavelength 244 and 273 nm. The concentration of drug was then calculated by using equation given below:

$$\text{Concentration of paracetamol} = (A_2a_{y_1} - A_1a_{y_2}) / (ax_2ay_1 - ax_1ay_2) \quad (1)$$

$$\text{Concentration of aceclofenac} = (A_1ax_2 - A_2ax_1) / (ax_2ay_1 - ax_1ay_2) \quad (2)$$

First order derivative spectroscopy (method II): The second method is based on first order derivative spectroscopy to overcome spectral interference from other drug. It was observed that paracetamol showed $dA/d\lambda$ zero at 243.6 nm in contrast to aceclofenac that has considerable $dA/d\lambda$ at this wavelength. Further, aceclofenac has zero $dA/d\lambda$ at 273.4 nm while at this wavelength paracetamol has significant $dA/d\lambda$. Therefore these two wavelengths can be employed for the estimation of paracetamol and aceclofenac without any interference. The calibration curves were plotted at these two wavelengths of concentrations against $dA/d\lambda$ within the range mentioned above. The equations obtained to determine concentration of paracetamol and aceclofenac are as follows:

$$\text{Conc. of aceclofenac} = dA/d\lambda \text{ at } 243.6 - 0.002/0.0058 \quad (3)$$

$$\text{Conc. of paracetamol} = dA/d\lambda \text{ at } 273.4 - 0.0005/0.0048 \quad (4)$$

Procedure for analysis of tablet formulation: An aliquot of sample stock solution (2 mL) was transferred to 10 mL standard volumetric flask and volume was made up to mark with phosphate buffer pH - 7. This solution was scanned in the range 200-400 nm and the spectra were derivatized to first order derivative ($n = 6$), $dA/d\lambda$ were measured at wavelengths 243.6 and 273.4 nm. Using the above equation of line the concentration of each drug was calculated.

Area under curve method (method III): This method involves area calculation of integrated value of absorbance with respect to wavelength in suggested region of wavelength.

Spectra of both the drugs were recorded and area under curve was calculated employing data processing mode. The optimum region was selected on the basis of repeated observation, which showed linearity in given concentration range of that drug. This method involves area calculation in region of 242-246 and 271-275 nm.

$$X = \frac{\text{AUC of component between selected wavelengths}}{\text{Concentration in g/L}}$$

The X values were calculated at these wavelength regions. The X value of paracetamol is 191.375 at 242-246 and 44.4308 at 271-275 nm and X value of aceclofenac is 55.6875 at 242-246 and 76.3585 at 271-275 nm.

Procedure for analysis of tablet formulation: The procedure used same as method I (AUC calculated instead of absorbances). The concentration of drug was then calculated by using following equation:

$$C_{\text{paracetamol}} = \frac{55.6875 \times \text{AUC}_{271-275} - 76.3585 \times \text{AUC}_{242-246}}{55.6875 \times 44.4308 - 76.3585 \times 191.375} \quad (5)$$

$$C_{\text{aceclofenac}} = \frac{44.4308 \times \text{AUC}_{242-246} - 191.375 \times \text{AUC}_{271-275}}{55.6875 \times 44.4308 - 76.3585 \times 191.375} \quad (6)$$

Absorbance ratio method (method IV): In the fourth method, the isoabsorptive point for both the drugs has been determined from the standard stock solution, which is found to be 265.6 nm used as λ_1 and 244 nm as λ_2 , which is the λ_{max} for paracetamol.

The Q-values and absorptivities for both drugs were calculated as follows:

$$Q_{\text{paracetamol}} = \frac{\text{Absorbance of paracetamol at 244.0 nm}}{\text{Absorbance of paracetamol at 265.6 nm}}$$

$$Q_{\text{aceclofenac}} = \frac{\text{Absorbance of aceclofenac at 244.0 nm}}{\text{Absorbance of aceclofenac at 265.6 nm}}$$

$$a_{\text{paracetamol}} = \frac{\text{Absorbance of paracetamol at 265.6 nm}}{\text{Concentration of paracetamol in g/L}}$$

$$a_{\text{aceclofenac}} = \frac{\text{Absorbance of aceclofenac at 265.6 nm}}{\text{Concentration of aceclofenac in g/L}}$$

$$Q_0 = \frac{\text{Absorbance of mixture/sample at 244.0 nm}}{\text{Absorbance of mixture/sample at 265.6 nm}}$$

The equations obtained for the estimation of concentration are:

$$\text{Conc. aceclofenac} = \frac{Q_0 - 2.7064}{0.7867 - 2.7064} \times \frac{A}{20.73} \quad (7)$$

$$\text{Conc. paracetamol} = \frac{Q_0 - 0.7867}{2.7064 - 0.7867} \times \frac{A}{20.73} \quad (8)$$

Procedure for analysis of tablet formulation: An aliquot of sample stock solution (2 mL) was transferred to 10 mL standard volumetric flask and volume was made up to mark. This solution was scanned in the range

200-400 nm. Absorbances were recorded at wavelength 244 nm and 265.6 nm. The concentration of each drug was then calculated by using above eqns. 7 and 8.

RESULTS AND DISCUSSION

Results of sample analysis for all methods are given in Table-1. To determine the accuracy and precision of method, recovery experiments were performed using proposed methods. Different concentration of standard solution was added to fixed volume of preanalyzed sample solution. The total amount of drug was then determined by these methods and the amount of added drug was then determined by difference. The results of recovery study are given in Table-2.

TABLE-1
RESULT OF COMMERCIAL SAMPLE ANALYSIS

Method	Label claim drug (mg/tab)	% Label claim estimated* (Mean \pm SD)		RSD (%)	SE
I	Paracetamol 500	99.825	0.14437	0.14450	0.05893
	Aceclofenac 100	99.891	0.16857	0.16860	0.06882
II	Paracetamol 500	99.7750	0.38826	0.38910	0.15850
	Aceclofenac 100	99.8516	0.19651	0.19680	0.08023
III	Paracetamol 500	100.000	0.17320	0.17320	0.07071
	Aceclofenac 100	100.033	0.23380	0.23370	0.09545
IV	Paracetamol 500	100.208	0.42102	0.42015	0.17188
	Aceclofenac 100	99.775	0.25050	0.25106	0.10226

*Mean of 6 determinations; SE = Standard error, RSD = Relative standard deviation.

TABLE-2
RECOVERY STUDY OF COMMERCIAL SAMPLE

Drug	Conc. of drug added		% Recovery (Methods)*			
	$\mu\text{g/mL}$	%	I	\pm SD	II	\pm SD
Aceclofenac	0	0	99.916	0.2516	99.916	0.1258
	10	50	99.606	0.7678	99.730	0.4334
	20	100	99.890	0.0173	98.986	0.4509
Paracetamol	0	0	99.650	0.3883	99.883	0.5251
	5	25	100.010	0.1665	99.266	0.5220
	10	50	99.870	0.0360	99.536	0.4954
Aceclofenac	0	0	99.883	0.2516	99.383	0.4193
	10	50	100.210	0.1905	100.33	0.7211
	20	100	99.940	0.1153	99.016	1.0066
Paracetamol	0	0	100.010	0.1755	100.45	0.4924
	5	25	100.010	0.1803	100.010	0.1973
	10	50	99.876	0.0472	99.796	0.0650

*Average of 3 determinations; SD = Standard deviation.

Values of standard deviation are satisfactorily low showing accuracy of proposed methods. All the four methods were found to be accurate, simple and rapid for routine simultaneous analysis of drugs from the formulations. Percentage recovery values ranged from 99 to 101 %.

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