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Development of UV spectroscopic method for the estimation of s(-) Amlodipine and Hydrochlorothiazide in dosage forms by simultaneous equation method

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ABSTRACT

S(-) amlodipine is a potent calcium channel blocker used for the treatment of hypertension, congestive heart failure and angina pectoris. S(-) amlodipine avoids the adverse effect of amlodipine in racemic mixtures. Hydrochlorothiazide is a first line diuretic drug of the thiazide class used for the treatment of hypertension, congestive heart failure and symptomatic edema. Literature review indicated that, various methods have been reported for the analysis of S(-) amlodipine and hydrochlorothiazide in formulation by RP-HPLC, HPTLC. But no analytical methods were reported for the estimation of these drugs using UV spectroscopic by simultaneous equation method.

Keywords: Simultaneous equation method, S(-) amlodipine, Hydrochlorothiazide.

INTRODUCTION

S(-) amlodipine is a potent calcium channel blocker used for the treatment of hypertension, congestive heart failure and angina pectoris. S(-) amlodipine avoids the adverse effect of amlodipine in racemic mixtures. Hydrochlorothiazide is a first line diuretic drug of the thiazide class used for the treatment of hypertension, congestive heart failure and symptomatic edema. Literature survey reveals few analytical methods for the determination of amlodipine alone and in combination with other drugs in pharmaceutical preparations and biological fluids including HPLC[1,2], UV spectroscopy[3], LC/MS[4]. Also there are some analytical methods reported for determination of hydrochlorothiazide alone and in combination including HPTLC[5], LC/MS[6]. No methods has been reported for the estimation of S(-) amlodipine and hydrochlorothiazide in combined dosage form. Present work emphasizes on the quantitative

estimation of S(-) amlodipine and hydrochlorothiazide in their combined dosage form by simultaneous equation[7] method using UV spectroscopy.

MATERIALS

JASCO V-530 UV/VIS Spectrophotometer (Shimadzu), Digital electronic balance (BL-220H) (Shimadzu, Japan), S(-) amlodipine (Cipla Pharmaceuticals), Hydrochlorothiazide (Micro Labs).

METHOD

Standard stock solutions of S(-) amlodipine[8] and hydrochlorothiazide were prepared by dissolving 5 and 12.5mg of the drug in methanol and the volume was made up to 100ml in a standard flask. From the stock solution a concentration ranging from 0.25-1.25µg/ml was prepared for S(-) amlodipine. A concentration ranging from 6.0-30.0µg/ml was made from the stock solution for hydrochlorothiazide. The maximum absorbances of S(-) amlodipine and hydrochlorothiazide were obtained at 239nm (λ_1) and 271nm (λ_2) respectively. S(-) amlodipine and hydrochlorothiazide showed linearity with absorbance in the range of 0.25 - 1.25 µg/ml and 6-30µg/ml at their respective maxima. Calibration curves of S(-) amlodipine were plotted using concentration Vs absorbance; at wavelength of 239nm the slope, intercept and correlation coefficient values were found to be 0.0089, 0.0384, and 0.9956 respectively (fig.1). At wavelength 271nm, slope, intercept and correlation coefficient values were found to be 0.1250, 0.1211, and 0.9998 respectively (fig.2). Calibration curves of hydrochlorothiazide were plotted using concentration Vs absorbance; at a wavelength of 239nm, the slope, intercept and correlation coefficient values were found to be 0.0105, 0.0005, and 0.9988 respectively (fig.3). At wavelength 271nm, the slope, intercept and correlation coefficient values were found to be 0.0042, 0.0054, and 0.9971 respectively (fig.4). The scanning solution of S(-) amlodipine and hydrochlorothiazide were carried out in range of 200 and 400 nm against methanol as blank for obtaining the overlain spectra that are used in analysis (fig.5 and 6). Absorbance and absorptivities of series of standard solutions were recorded at selected wavelengths λ_1 and λ_2 .

Twenty tablets each containing 5mg of S(-) amlodipine and 12.5mg of hydrochlorothiazide were weighed and the average weight was calculated. A quantity equivalent to 12.5mg of drug was dissolved in methanol and the volume was made up to get the required concentrations. Absorbances were noted at 239nm and 271nm respectively. The amount of S(-) amlodipine and hydrochlorothiazide was calculated using the simultaneous equation [8] given below

$$\text{At } \lambda_1, A_1 = ax_1bc_x + ay_1bc_y$$

$$\text{At } \lambda_2, A_2 = ax_2bc_x + ay_2bc_y$$

$$C_{amil} = \frac{A_2ay_1 - A_1ay_2}{ax_2ay_1 - ax_1ay_2}$$

$$C_{hct} = \frac{A_1ax_2 - A_2ax_1}{ax_2ay_1 - ax_1ay_2}$$

- A_1 = absorbance of formulation at 239nm.
 A_2 = absorbance of formulation at 271nm.
 ax_1 = absorptivity of S(-) amlodipine at 239nm.
 ax_2 = absorptivity of S(-) amlodipine at 271nm.
 ay_1 = absorptivity of hydrochlorothiazide at 239nm.
 ay_2 = absorptivity of hydrochlorothiazide at 271nm.
 C_{amil} = Concentration of amlodipine.
 C_{het} = Concentration of hydrochlorothiazide.

RESULTS AND DISCUSSION

Literature review indicated that, various methods have been reported for the analysis of S(-) amlodipine and hydrochlorothiazide in formulation by RP-HPLC, HPTLC. But no analytical methods were reported for the estimation of these drugs using UV spectroscopic by simultaneous equation method. The wavelength of S(-) amlodipine and hydrochlorothiazide was found to be 239 and 271 nm respectively. In these wavelengths absorbances of S(-) amlodipine and hydrochlorothiazide and mixture was noted. The results showed an excellent correlation between absorbance and concentration of the drugs. Validation parameters like accuracy, precision and linearity showed low %RSD values which indicates that the method is precise and sensitive. The linearity for S(-) amlodipine and hydrochlorothiazide were found to be 0.9996 and 0.9993 respectively. The accuracy for S(-) amlodipine and hydrochlorothiazide were found to be 98.95% and 99.15% respectively.

Table 1: Absorbance of S (-) amlodipine at selected wavelengths

Concentration ($\mu\text{g/ml}$)	Absorbance at 239nm	Absorbance at 271nm
0.25	0.1549	0.0190
0.50	0.1867	0.0264
0.75	0.2146	0.0396
1.00	0.2462	0.0462
1.25	0.2765	0.0571

Table 2: Absorbance of hydrochlorothiazide at selected wavelengths

Concentration ($\mu\text{g/ml}$)	Absorbance at 239nm	Absorbance at 271nm
6.0	0.0138	0.0379
12.0	0.0161	0.0680
18.0	0.0193	0.0970
24.0	0.0222	0.1395
30.0	0.0256	0.1639

Table 3: Analysis of formulation

Drug	Amount (mg/tab)		% label claim	% RSD*
	Labelled	Found		
s(-) amlodipine	5mg	4.98mg	99.21%	0.106
hydrochlorothiazide	12.5mg	12.41mg	98.76%	0.124

* Mean RSD of six observations

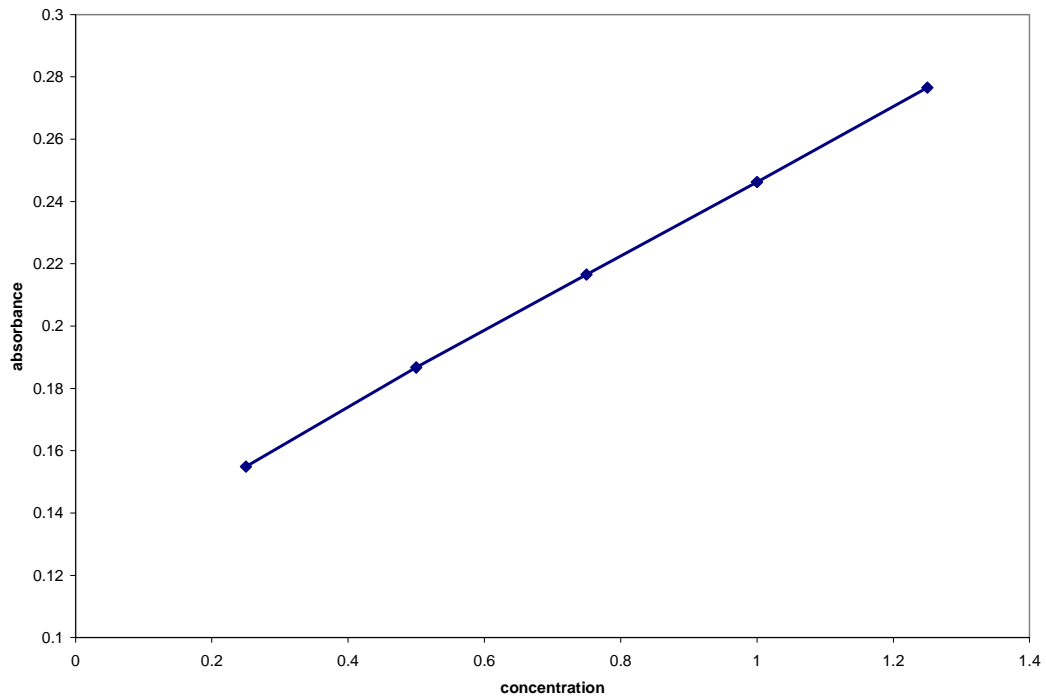


Fig 1: Calibration curve of s (-) amlodipine at 239nm

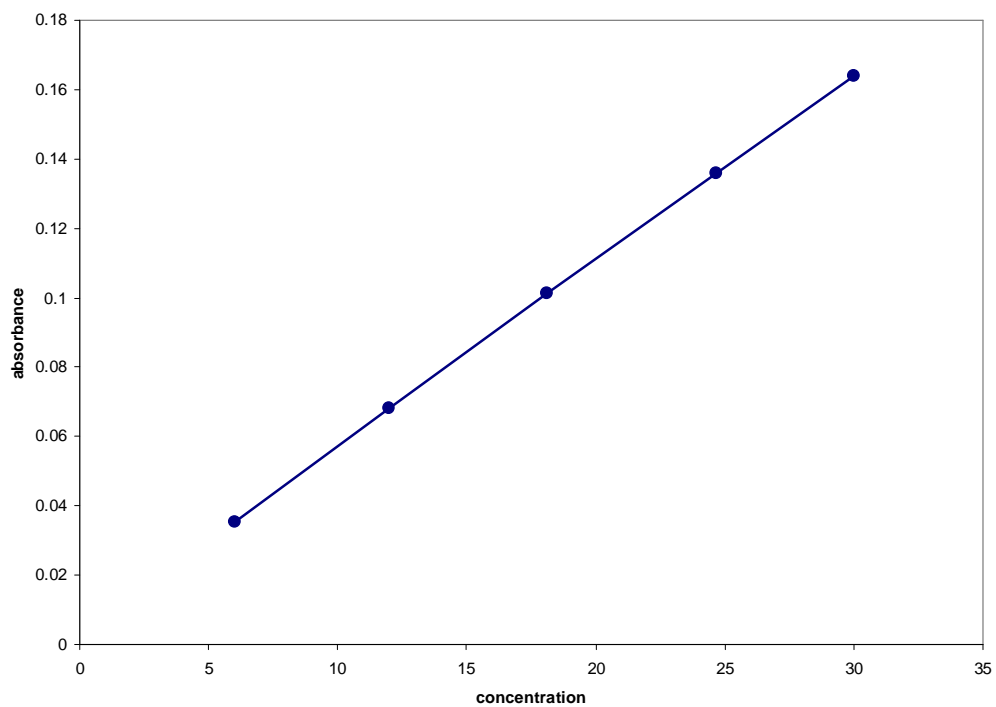


Fig 2: Calibration curve of s (-) amlodipine at 271 nm

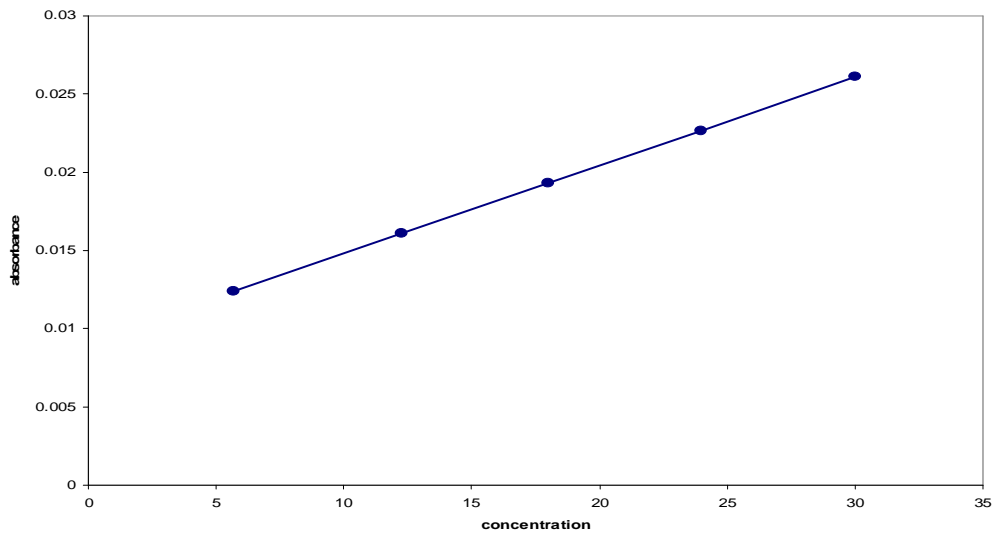


Fig 3: Calibration curve of hydrochlorothiazide at 239nm

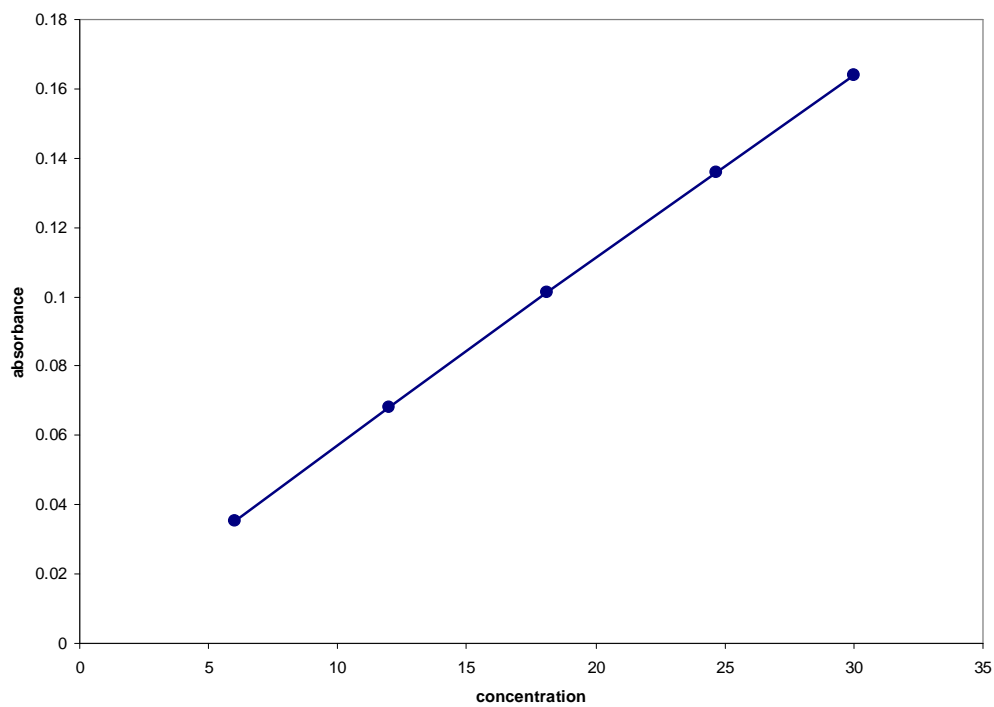


Fig 4: Calibration curve of hydrochlorothiazide at 271nm

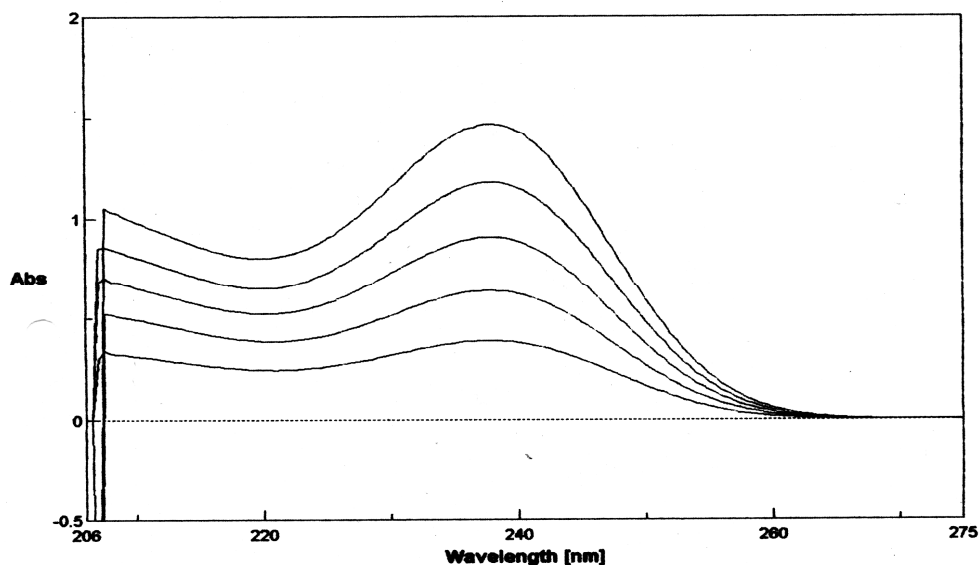


Fig 5: Overlay spectra of standard s (-) amlodipine (0.25-1.25µg/ml)

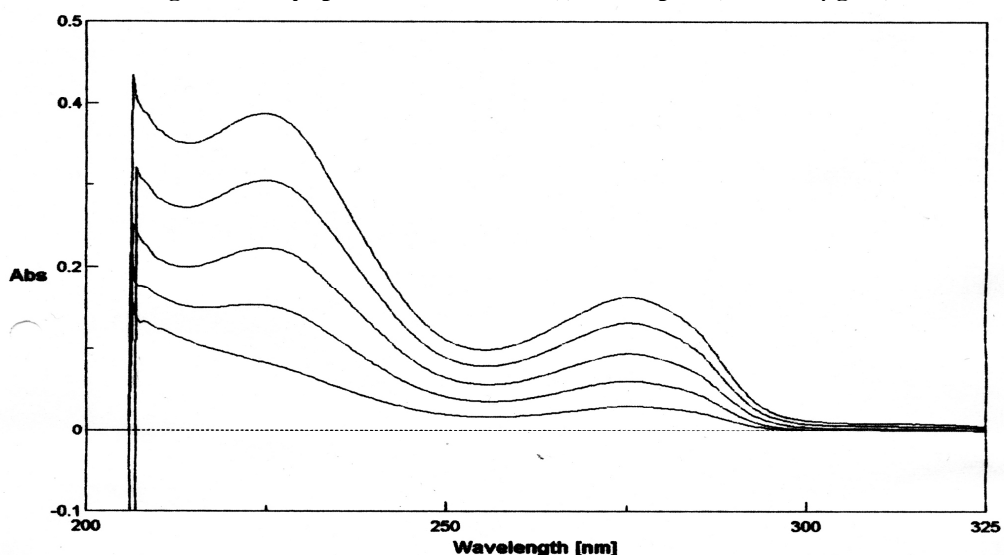


Fig 6: Overlay spectra of standard hydrochlorothiazide (6.0- 30.0µg/ml)

CONCLUSION

The UV spectroscopic estimation of S(-) amlodipine and hydrochlorothiazide in combined dosage form was solved using simultaneous equation method and it was found within the limits. Validation parameters like accuracy, precision and linearity showed low %RSD values which indicates that the method is precise and sensitive. Hence this method can be employed for commercial works.

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