

Simultaneous Spectrophotometric Estimation of Lercanidipine Hydrochloride and Atenolol in Tablet Dosage Form

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Abstract

Two accurate, precise, sensitive and economical spectrophotometric methods were developed and validated for simultaneous estimation of Lercanidipine hydrochloride and Atenolol in tablet dosage form. These methods were developed based on the simultaneous estimation of drugs in a binary mixture without previous separation. The methods employed were Absorbance Ratio Method (Q-Analysis) (I) and Simultaneous Equation Method (Vierodt's Method) (II). The first method employs 261nm as λ_1 (Isobestic point) and 273 nm as λ_2 (λ_{\max} of Atenolol) for formation of equations. The second method employs estimation of a drug concentration by selecting λ_{\max} where the absorbances of these drugs were maximum. So λ_{\max} for Lercanidipine hydrochloride and Atenolol is 242 nm and 273 nm respectively. Lercanidipine hydrochloride and Atenolol obey Beer's law in the concentration range 10-50 $\mu\text{g mL}^{-1}$ ($r^2=0.9999$) and 50-250 $\mu\text{g mL}^{-1}$ ($r^2=0.9999$) in 0.1 N HCL. The mean recovery for Lercanidipine hydrochloride and Atenolol were found to be 98.07 \pm 0.21% and 100 \pm 0.14% from method I and 98.60 \pm 0.36% and 100 \pm 0.05% from method II. The developed methods were validated according to ICH guidelines and values of accuracy, precision and other statistical analysis were found to be in good accordance with the prescribed values. Thus the proposed methods were successfully applied for simultaneous determination of Lercanidipine hydrochloride and Atenolol in routine industrial work.

Keywords:

Lercanidipine hydrochloride; atenolol; absorbance ratio method; simultaneous equation method; spectrophotometric

1. Introduction

Lercanidipine hydrochloride, (LER) chemically, 1,4-Dihydro-2,6-dimethyl-4-(3-nitrophenyl)-3,5-pyridinedicarboxylic acid 2-[(3,3-diphenylpropyl)methyl amino]-1,1-dimethylethyl methyl ester (Fig.1A) is used in the treatment of mild to moderate hypertension. Atenolol, (ATN) chemically, 4-(2-hydroxy-3-isopropylaminopropoxy) phenyl acetamide (Fig.1B) is an antihypertensive, antianginal and antiarrhythmic drug [1, 2]. Literature survey revealed that many analytical method [3, 4] spectrophotometric methods [5-7] and a HPLC [8-11] and UPLC [12] method has been reported for determination of lercanidipine hydrochloride in bulk and in biological fluids. Several analytical methods reported for the quantitative determination of atenolol individually in pharmaceutical formulations or in biological fluids, are HPLC [13-16], capillary zone electrophoresis [17] and spectrophotometry [18-20]. Extensive literature survey reveals that no method is reported

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for simultaneous determination of LER and ATN in tablet dosage form. Aim of present work was to develop simple, precise, accurate and economical spectrophotometric methods and validate as per ICH guidelines [21] for simultaneous determination in binary drug formulation.

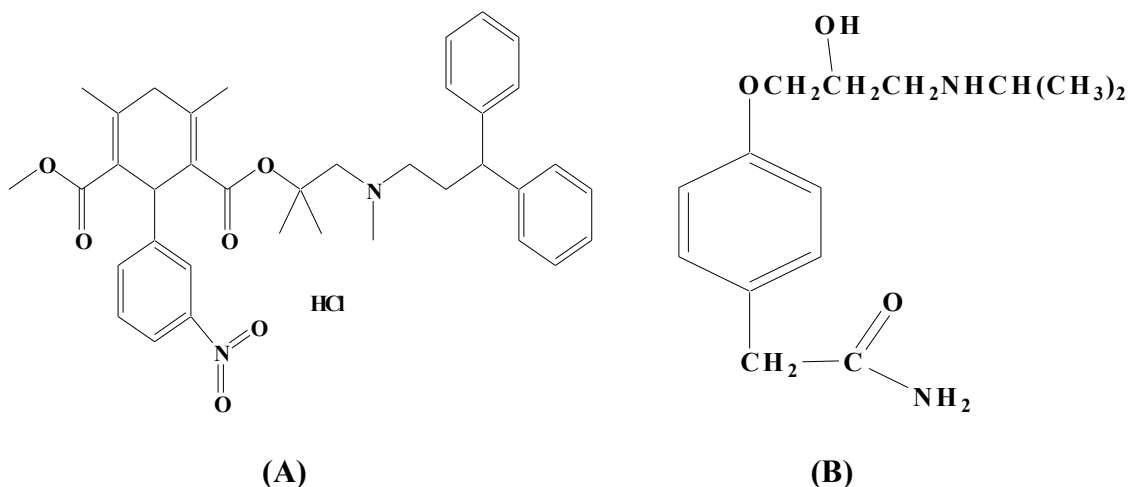


Fig.1. Chemical structures of (A) Lericandipine hydrochloride and (B) Atenolol

2. Experimental

2.1. Instrumentation

The proposed work was carried out on a shimadzu UV-visible spectrophotometer (model UV-1700 series), which possesses a double beam double detector configuration with a 1 cm quartz matched cell. All weighing was done on electronic balance (Citizen).

2.2. Reagents and Chemicals

Analytically pure sample of LER and ATN was kindly supplied by Glenmark Pharmaceuticals Ltd. (Nashik, India). The pharmaceutical dosage form used in this study was a Lotensyl AT (Sun Pharmaceuticals Industries Ltd. Mumbai) tablets containing 50 mg atenolol and 10 mg lercanidipine hydrochloride were obtained from the local drug market.

2.3. Theory

2.3.1. Absorbance Ratio Method (Method I)

In this method, the isoabsorptive points for both the drugs were determined from the spectra of standard drug solutions, The wavelengths selected were 261 nm as λ_1 (Isoabsorptive point) and 273 nm (λ_{\max} for ATN) as λ_2 for formation of equations as shown in Eqn. 1, 2. The concentration of individual components calculated by mathematical treatment of the simultaneous equations

$$C_{\text{LER}} = \frac{Q_m - 1.885 / 0.7593 - 1.885}{0.0108} \times A_1 \quad (1)$$

$$C_{\text{ATN}} = \frac{Q_m - 0.7593 / 1.885 - 0.7593}{0.0022} \times A_1 \quad (2)$$

where $Q_m = A_2 / A_1$, A_1 is absorbance of sample at isoabsorptive point, A_2 is absorbance of sample at λ_{\max} of LER, $Q_x (0.7593) = a_{x2} / a_{x1}$, $Q_y (1.885) = a_{y2} / a_{y1}$, a_{x1} and a_{x2} represent absorptivities of LER at λ_1 (261 nm, isoabsorptive point) and λ_2 (273nm, λ_{\max} of ATN) and a_{y1} and a_{y2} denote absorptivities of ATN at λ_1 (261 nm, isoabsorptive point) and λ_2 (273nm, λ_{\max} of ATN) respectively; C_{LER} and C_{ATN} be the concentration of LER [should lie outside the range of (0.1-0.2)] and ATN, component by the mechanism of the absorbance respectively.

2.3.2. Vierordt's Simultaneous Equation Method (Method II)

This method of analysis is based on the absorption of drugs (X and Y) at the wavelength maximum of the other. The quantification analyses of LER and ATN in a binary mixture were performed with the following equations:

$$C_{\text{LER}} = (A_2 a_{y1} - A_1 a_{y2}) / (a_{x2} a_{y1} - a_{x1} a_{y2}) \quad (3)$$

$$C_{\text{ATN}} = (A_1 a_{x2} - A_2 a_{x1}) / (a_{x2} a_{y1} - a_{x1} a_{y2}) \quad (4)$$

where C_{LER} and C_{ATN} are the concentrations of LER and ATN respectively in the diluted sample, a_{x1} and a_{x2} are absorptivities of LER at λ_1 (242nm, λ_{max} of LER) and λ_2 (273nm, λ_{max} of ATN), a_{y1} and a_{y2} are absorptivities of ATN at λ_1 (242nm, λ_{max} of LER) and λ_2 (273nm, λ_{max} of ATN). The absorbance of the diluted samples at 242nm and 273nm are A_1 ($A_1 = a_{x1} b c_x + a_{y1} b c_y$) and A_2 ($A_2 = a_{x2} b c_x + a_{y2} b c_y$) respectively.

2.4. Preparation of Standard Stock Solutions

Standard stock solutions were prepared by dissolving separately 100 mg of each drug in 100 mL of 0.1 N HCL to get concentration of $1000 \mu\text{g mL}^{-1}$. The standard solution ($1000 \mu\text{g mL}^{-1}$) was further diluted with 0.1 N HCL to obtain concentration range 10, 20, 30, 40 and $50 \mu\text{g mL}^{-1}$ for LER and 50, 100, 150, 200, $250 \mu\text{g mL}^{-1}$ for ATN. Working standard solution of concentration $10 \mu\text{g mL}^{-1}$ of LER and $50 \mu\text{g mL}^{-1}$ of ATN were scanned in the wavelength range of 200-400 nm against 0.1 N HCL as blank, the overlay spectra of the two were recorded (Fig 2). The overlay spectra exhibit major absorbance maxima at 242 nm and 273 nm for LER and ATN respectively and at 261 nm as isoabsorptive point which revealed that the peaks are well satisfying the criteria for obtaining maximum precision based on LER and ATN, respectively.

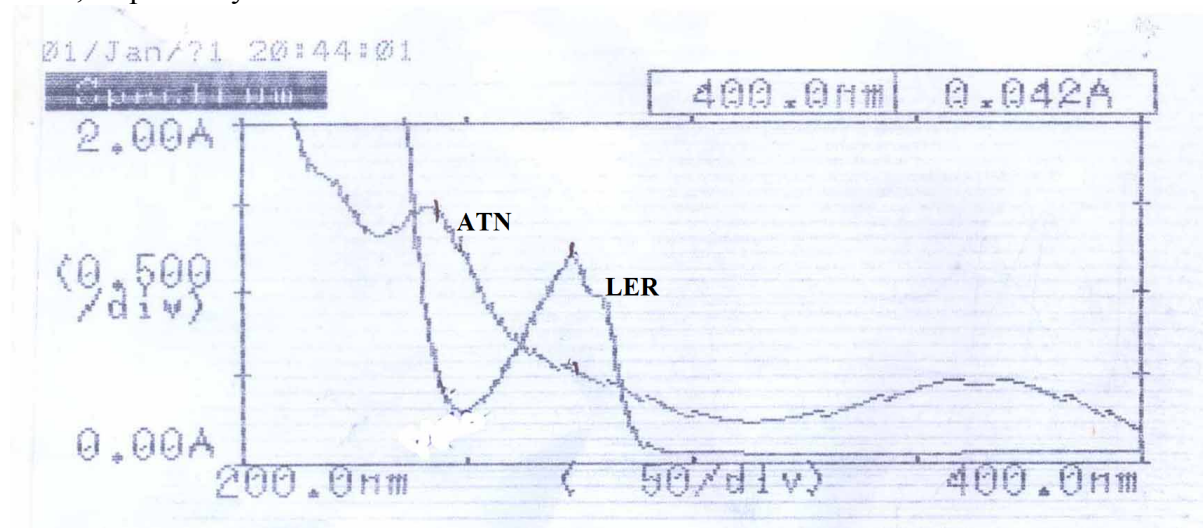


Fig.2. Overlay spectra of Atenolol and Lericandipine hydrochloride

2.5. Preparation of Analysis of Tablet sample

Twenty tablets (Lotensyl AT) were weighed and ground to a fine powder. An accurately weighed powder sample equivalent to 10 mg of LER and 50 mg ATN were transferred to 100 mL of volumetric flask containing 0.1 N HCL solution. 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 HCL and was analyzed on UV spectrophotometer. The absorbance at 242 (λ_{\max} of LER), 273 (λ_{\max} of ATN) and 261 (Isobestic point) as in Fig.2 were recorded. Drug content of tablet formulation were calculated by using the equations mentioned above in method I and method II and value are reported in Table 1.

Table 1. Results of commercial tablet analysis

Method	Amount of drug claimed (mg/TAB)		% Label Claim estimated* (Mean \pm S.D)		% R. S. D.		Standard error of standard deviation (SE σ)	
	LER- 10	ATN-50	LER	ATN	LER	ATN	LER	ATN
I	LER- 10	ATN-50	98.50 \pm 0.37	100.13 \pm 0.16	0.373	0.156	0.150	0.065
II	LER- 10	ATN-50	98.63 \pm 0.721	99.33 \pm 0.526	0.731	0.530	0.295	0.215

* Mean of six determinations, R.S.D. is relative standard deviation

2.6. Recovery Studies

The recovery studies were performed by analyzing the definite amount of added drug to pre-analyzed tablet solution. For this study, pre-analyzed tablet solution ranging from 10-30 $\mu\text{g mL}^{-1}$ of LER and 50-150 $\mu\text{g mL}^{-1}$ of ATN was taken. Bulk drug samples of 10 and 50 $\mu\text{g mL}^{-1}$ of LER and ATN respectively was added as spiked concentrations. This was repeated three times with three concentrations to emphasize validation. The drug contents were determined by the proposed analytical methods and Results of recovery studies are reported in Table 2.

Table 2. Result of recovery studies of tablet formulation with statically evaluation

Method	Theoretical conc. ($\mu\text{g mL}^{-1}$)		Amount added ($\mu\text{g mL}^{-1}$)		Percentage recovery Mean \pm S.D. (n=6)		Coefficient of variation, %		* Standard error of standard deviation (SE σ)	
	LER	ATN	LER	ATN	LER	ATN	LER	ATN	LER	ATN
I	10	50	10	50	98.3 \pm 0.32	100.04 \pm 0.29	0.33	0.29	0.08	0.07
	20	100	10	50	97.9 \pm 0.89	100.12 \pm 0.47	0.91	0.47	0.21	0.11
	30	150	10	50	98.0 \pm 0.78	99.84 \pm 0.92	0.80	0.92	0.18	0.22
II	10	50	10	50	98.7 \pm 0.59	100.02 \pm 0.31	0.60	0.31	0.14	0.07
	20	100	10	50	98.2 \pm 0.73	99.94 \pm 0.27	0.74	0.27	0.17	0.06
	30	150	10	50	98.9 \pm 0.98	100.04 \pm 0.81	0.99	0.81	0.23	0.19

* Mean of nine determinations (3 replicates at 3 concentration level)

2.7. Precision Studies

To evaluate precision at different parameter like repeatability, intermediate precision, five dilutions in three replicates were analyzed in same day, in two different days and by two analysts for day to day and analyst to analyst variation and results were shown in Table 3.

3. Results and Discussion

In the present work, two methods, namely graphical absorbance ratio method (Q-Analysis) and simultaneous equation (Vierordt's method) were developed for the simultaneous spectroscopic estimation of LER and ATN in commercially available tablet dosage form using 0.1 N HCL. From the overlay spectra of the two drugs the wavelengths used for graphical absorbance ratio method is 261 nm (isoabsorptive point) and 273 nm (λ_{\max}

of ATN) and for simultaneous equation method 242 nm (λ_{\max} of LER) and 273 nm (λ_{\max} of ATN) were selected to give optimum accuracy, precision, time, economy and sensitivity. The mean percent label claims in tablet were found to be $98.50 \pm 0.37\%$, $100.13 \pm 0.16\%$ by the proposed methods-I and $98.63 \pm 0.72\%$, $99.33 \pm 0.53\%$ by Method-II for LER and ATN respectively (Tables 1). In order to demonstrate the validity and applicability, recovery studies were performed by spiking of bulk drugs in pre analyzed tablets and the percentage recoveries for LER and ATN were found to be ranging from 97.9-98.9%, 99.94-100.12% by methods I and Method II respectively as shown in Tables 2. The values of percent relative standard deviation for the validation parameters of LER and ATN were found to be less than 2 in both methods (Table 3), indicating good accuracy, precision and repeatability of the proposed methods.

Table 3. Result of Precision

Method	Validation Parameter	Percentage Mean \pm S.D*		Percentage RSD*	
		LER	ATN	LER	ATN
I	Repeatability	99.21 \pm 0.14	99.83 \pm 0.01	0.58	0.004
	Intermediate precision				
	Day to Day	99.70 \pm 0.010	99.70 \pm 0.01	0.06	0.07
	Analyst to Analyst	99.14 \pm 0.01	99.33 \pm 0.02	0.07	0.08
II	Repeatability	99.50 \pm 0.05	100.06 \pm 0.05	0.16	0.03
	Intermediate precision				
	Day to Day	99.56 \pm 0.01	100.12 \pm 0.02	0.04	0.02
	Analyst to Analyst	99.73 \pm 0.03	100.13 \pm 0.01	0.14	0.01

* Mean of fifteen determinations (3 replicates at 5 concentration level)

4. Conclusion

The validated spectrophotometric methods employed here proved to be simple, economical, rapid, precise and accurate. Thus these can be used for routine simultaneous determination of LER and ATN in tablet dosage form instead of processing and analyzing each drug separately.

References

1. Oneil M J, Smith A and Heckelman P E (2001) The Merck Index, 13th edition, Merck Research Laboratories, Whitehouse Station: New Jersey, p. 5465, 863.
2. Sweetman S C (2002) Martindale: The complete drug reference, 33rd edition, The Pharmaceutical Press, London, p. 897-3, 825-3.
3. Christians T, Diewald D, Wessler C, Otte Y, Lehmann J and Holzgrabe U (1999) The voltametric polarographic method to determine lercanidipine. J Chromatogr A 853: 455-60.
4. Alvarez L A, Nunez Vergara L J, Pujol S and Squella A (2002) Voltametric behavior of lercanidipine and its differential pulse polarographic determination in tablets. Electroanalysis 14: 1098-1104.
5. Mubeen G, Damanjit S G and Kadri U (2008) Spectrophotometric method for estimation of Lercanidipine in tablets. J Biomed Pharmacol 1(2):479-80.

6. Eswar G M, Shyam S B and Devala R G (2004) Spectrophotometric methods for the determination of lercanidipine in pharmaceutical formulations. *The Indian Pharmacist* (29):77-78.
7. Abu El-Enin M A, El-Wasseef D R, El-Sherbiny D T and El-Ashry S M (2009) Spectrophotometric determination of labetalol and lercanidipine in pure form and in pharmaceutical preparations using ferric-1,10-phenanthroline. *Int J Biomed Sci* 5(3): 261-266.
8. Salem I I, Idrees J, Tamimi J A T and Farina P (2004) Selective and rapid liquid chromatography mass spectrometry method for determination of lercanidipine in human plasma. *J Chromatogr A* 803: 201-207.
9. Jessica F, Roberto G, Carlo B and Vanni C (2006) Photochemical stability of lercanidipine and its determination in tablets by HPLC-UV and LC-ESI-MS/MS. *J Pharm Biomed Anal* 41:176-181.
10. Alvarez L A, Pujol S, Squella J A and Nunez Vergara L J (2003) A selective HPLC method for determination of lercanidipine HCl in tablet. *J Pharm Biomed Anal* 31: 1-9.
11. Vijaya M K and Muley P R (2004) Reverse phase high performance liquid chromatographic method for determination of lercanidipine in bulk drug and solid dosage forms. *Indian Drugs* 41 (1):24-27.
12. Kalovidouris M, Michalea S, Robola N, Koutsopoulou M and Panderi I (2006) Ultra-performance liquid chromatography/tandem mass spectrometry method for the determination of lercanidipine in human plasma. *R Comm M Spectra* 20 (19): 2939–2946.
13. Sivakumar T, Venkatesan P, Manavalan R and K Valliappan (2007) Development of a HPLC method for the simultaneous determination of losartan potassium and atenolol in tablets. *Indian J Pharma Sci* 69 (1):154-157.
14. Ravi Sankar S, Nanjan M J, Vasudevan M, Shaat N, Suresh B and Sankar S R (1997) Simultaneous estimation of atenolol and amlodipine in formulations by reversed Phase-HPLC. *Indian J Pharma Sci* 59(4):171-173
15. Shah S A, Rathod I S, Shishoo C J, Savale S S, Satia M C and Bhat K M.(2000) A sensitive high performance liquid chromatography method for determination of bioequivalence of atenolol formulations. *Indian J Pharma Sci* 62(3): 187-192
16. Lamprecht G, Kraushofer T, Stoschitzky K and Lindner W (2000) Enantioselective analysis of (R)- and (S)-atenolol in urine samples by a high-performance liquid chromatography column-switching setup. *J Chromatogr B* 740: 219.
17. Shafaati A and Clark B J (1996) Development and validation of a capillary zone electrophoresis for determination of atenolol in presence of its related substances in bulk and tablet dosage form. *J Pharm Biomed Anal* 14: 1547.
18. Prasad U V, Rao G V and Sastry C S (2002) New spectrophotometric analysis of atenolol in pure and pharmaceutical formulations. *Indian J Pharma Sci* 64 (4):129-132.
19. Kasture A V and Madhuri R (2006) Simultaneous UV-spectrophotometric method for the estimation of atenolol and amlodipine Besylate in combined dosage form. *Indian J Pharma Sci* 68(3):394-396
20. Prasad C V N, Parihar C, Sunil K and Parimoo P (1998) Simultaneous determination of amiloride HCl hydrochlorothiazide and atenolol in combined formulations by derivative spectroscopy. *J Pharm Biomed Anal*.17:877-84.

21. International conference on harmonization of technical requirements for registration of pharmaceuticals for human use. 2005 Validation of analytical procedures: text and methodology Q2 (R1). <http://www.ich.org/LOB/media/MEDIA417.pdf>.