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A HIGH PERFORMANCE LIQUID CHROMATOGRAPHIC ASSAY FOR LERCANIDIPINE HYDROCHLORIDE

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ABSTRACT

A simple, rapid and reproducible high performance reversed phase liquid chromatographic method has been developed for the estimation of lercanidipine hydrochloride in bulk drug samples and pharmaceutical dosage forms using RPC-18 column. The mobile phase consists of buffer solution and acetonitrile in the ratio 650 : 350, respectively, and was pumped at 1.0 mL/min at 30°C. The detection was carried out at 205 nm and the calibration curve was linear in the range of 0.1 μg/mL to 20 μg/mL. The method was statistically validated for its linearity, precision and accuracy. The intra and inter-day variation was found to be less than 1% showing high precision of the assay method. Due to its simplicity, rapidness, high precision and accuracy, the proposed HPLC method may be used for determining lercanidipine hydrochloride in bulk drug samples or in pharmaceutical formulations.

Key words : Lercanidipine hydrochloride, RP-HPLC.

INTRODUCTION

Lercanidipine hydrochloride[1,2] is chemically (±) – 2 - [(3 – 3 - diphenyl propyl-methylamino] - 1, 1 - dimethylethyl methyl - 1, 4 – dihydrodimethyl – 4 - (m - nitrophenyl) -3, 5 - pyridine dicarboxylate hydrochloride. It is dihydropyridine calcium channel blocker, used alone or with an angiotension converting enzyme inhibitor to treat hypertension, chronic stable angina pectories and prinzmetals variant angina. It is similar to other pheripheral vasodialators. It inhibits the inflex of extra cellular calcium across the mycocardial and vascular smooth muscle, cell membranes[3,4]. It is not official in any pharmacopoeia. Literature survey reveals that no spectrophotometric methods have been reported for its quantitative estimation in bulk drug and pharmaceutical dosage forms. The aim of this study is to develop a simple, rapid, precise and accurate reverse-phase HPLC method for the determination of lercanidipine hydrochloride in bulk drug samples or in pharmaceutical dosage forms.

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EXPERIMENTAL

Instrumentation

Quantitative HPLC was performed on a gradient high pressure liquid chromatograph (Shimadzu HPLC Class VP series) with two LC-10AT VP pumps, variable wavelength progra mmable UV/VIS detector SPD-10A VP, CTO-10AS VP column oven (Shimadzu), SCL-10A VP system controller (Shimadzu), a disposable guard column LC-18 (Pelliguard)^{TM}, LC-18, 2 cm, Supelco, Inc., Bellefonte, PA and RP C-18 column (150 mm x 4.6 mm I.D., particle size 5 μm) was used. The HPLC system was equipped with the software Class – VP series version 6.01 (Shimadzu).

Chemicals and reagents

Pure samples of lercanidipine hydrochloride were obtained as gift sample from Glenmark Pharmaceuticals, Aurangabad, India. Acetonitrile (HPLC grade), sodium dihydrogen phosphate (A.R grade) and phosphoric acid (A.R. grade) were purchased from Merck Ltd. (Mumbai, India). Water (HPLC grade, Qualigens). The co mmercially available lercanidipine hydrochloride tablets claimed to contain 10 mg of drug were procured from local market.

Chromatographic conditions

The contents of the mobile phase were buffer solution and acetonitrile in the ratio of 650 : 350. Buffer was prepared by dissolving 6.0 g of sodium dihydrogen phosphate in 1000 mL of water and pH was adjusted to 3.80 with 10% H_3PO_4. The contents of the mobile phase were filtered before use through 0.45 μm membrane filter, degassed with a helium spurge for 15 min and pumped from the respective solvent reservoirs to the column at a flow rate of 1.0 mL/min, which yielded a column back pressure of 138-140 kg/cm^2. The run time was set at 10 min and the column temperature was maintained at 30°C. The volume of the injection loop was 20 μL. Prior to injection of the drug solutions, the column was equilibrated for at least 30 min with the mobile phase flowing through the systems. The elements were monitored at 205 nm and the data were acquired, stored and analyzed with the software Class-VP series version 6.01 (Shimadzu).

Procedure

About 100 mg of lercanidipine hydrochloride was accurately weighed and dissolved in acetonitrile so as to give 1 mg/mL solution. Subsequent dilutions of this solution were made with mobile phase to get concentrations of 0.1 to 20 μg/mL of

lercanidipine hydrochloride. The standard solutions prepared as above were injected five times into the column at a flow rate of 1.0 mL/min. The peak areas of the drug concentration were calculated. The regression of the drug concentration over the peak areas was obtained. This regression equation was used to estimate the amount of lercanidipine hydrochloride in tablet dosage of forms.

lercanidipine hydrochloride solutions containing 8 μg/mL, 12 μg/mL and 20 μg//mL were subjected to the proposed HPLC analysis for finding out the intra-and inter-day variations. The recovery studies were carried out by adding known amounts of lercanidipine hydrochloride to the preanalyzed samples and subjecting them to the proposed HPLC method.

Assay

Fifty tablets each containing 10 mg were weighed and powdered. An accurately weighed portion of the powder equivalent to 100 mg of lercanidipine hydrochloride was transferred to a 100 mL volumetric flask containing 50 mL of acetonitrile. The contents of the flask were sonicated for 15 min. to dissolve lercanidipine and made upto volume with mobile phase and the resulting mixture was filtered through a 0.45 μm filter. One millitre of this solution was added to a 100 mL volumetric flask and made upto the volume with mobile phase. This solution (20 μL) was injected five times into the column. The mean values of peak areas of five such determinations were calculated and the drug content in the tablet was quantified using the regression equation obtained above. The same procedure was followed for the estimation of lercanidipine in other co mmercially available tablet dosage forms.

RESULTS AND DISCUSSION

The present study was carried out to develop a sensitive, precise and accurate HLPC method for the analysis of lercanidipine hydrochloride in bulk samples or pharmaceutical dosage forms. The column pressure varied from 138 to 140 kg/cm^2. The retention time for lercanidipine hydrochloride was 2.292 min. for a run period of 15 min. Each of the samples was injected 5 times and the same retention times were observed in all cases. The peak area of different concentrations set up as above were observed in all cases. The peak area of different concentrations set up as above were calculated and the average values for 5 such determinations are shown in Table 1. The peak area for drug solution was reproducible as indicated by low coefficient of variation (1.96%). A good linear relationship (r = 0.9991) was observed between the concentrations of lercanidipine

hydrochloride and the respective peak areas. The calibration graph was found to be Y = – 0.01269 + 0.62609 X, where Y is the peak area and X is the concentration of lercanidipine in the range of 0.1 to 20 μg/mL. When lercanidipine solutions containing 8 μg/mL, 12 μg/mL and 20 μg/mL were analyzed by the proposed reversed phase HPLC method for finding out intra-and inter-day variations, a low coefficient of variation was observed (Table 2). This shows that the present HPLC method is highly precise. The amount of lercanidipine from the preanalyzed sample containing known amounts of the drug are shown in Table 3. About 107.3% lercanidipine could be recovered from the preanalyzed sample indicating the high accuracy of the proposed HPLC method.

Table 1. Calibration of HPLC method for the estimation of lercanidipine hydrochloride

Concentration of lercanidipine (μg/mL)	Peak area*	C.V. (%)
0.1	180162	0.970
0.2	360324	1.250
0.4	720647	1.390
0.8	1441297	1.070
1.0	1801625	1.960
2.0	360325	0.208
4.0	7206481	0.042
8.0	14412961	1.038
12.0	2161945	1.259
20.0	3603241	0.095

* mean of six determinations

The drug content in the tablets was quantified using the proposed analytical method. The mean content of lercanidipine hydrochloride in two different brands of tablet dosage form is shown in Table 4.

The absence of additional peaks indicates no interference of the excipients used in the tablet. The tablets were found to contain 100.6% to 107.3% of the labelled amount. The low 1% CV indicates the reproducibility of the assay of lercanidipine hydrochloride in

the tablet dosage form. The proposed reversed phase HPLC method was found to be simple, precise, highly accurate, specific and less time consuming.

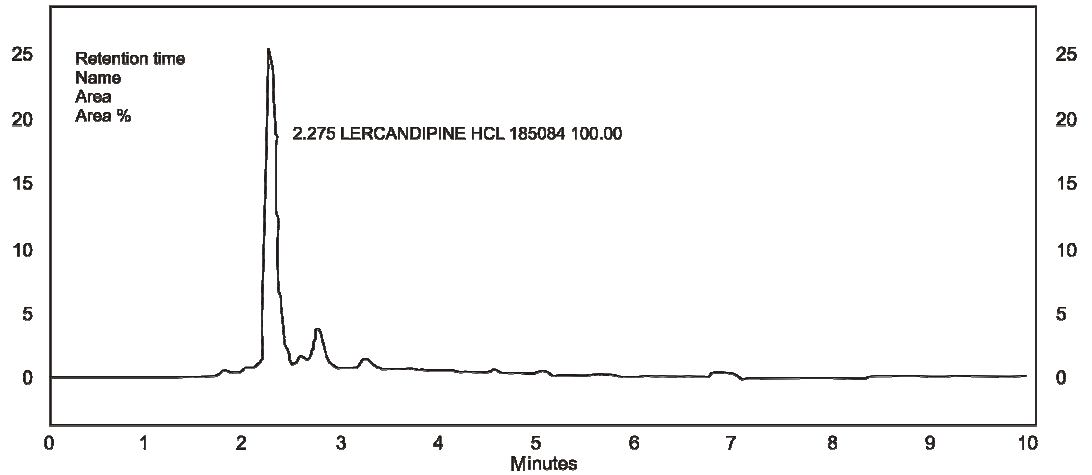

Fig. 1 : A typical chromatogram for lercanidipine

Table 2. Inter and intra-day precision for lercanidipine assay in pharmaceutical dosage forms by the proposed HPLC method

Lercanidipine concentration (μg/mL)	Concentration of lercanidipine found on			
	Intra-day		Inter-day	
	Mean n = 5	C. V. (%)	Mean n = 5	C.V. (%)
8	7.94	0.95	8.00	0.88
12	11.99	0.31	12.01	1.40
20	20.00	1.39	19.95	0.71

Table 3. Recovery of lercanidipine using proposed HPLC method

Amount of drug added (μg/mL)	Mean (± SD) amount found (μg) (n = 5)	Mean (± SD) % of recovery (n = 5)
4	3.95 ± 0.34	99.77 ± 1.22
8	8.00 ± 0.62	100.10 ± 1.01
12	11.99 ± 0.09	99.99 ± 0.88

Table 4. Mean (± SD) amount of lercanidipine in tablet dosage forms by proposed HPLC method

Tablets *	Labelled amount of drugs (mg)	Mean (± s.d) amount found (mg) (n = 5)	Mean (± s.d.) purity
T_1	10	10.73	107.3%
T_2	10	10.06	100.6%

*T_1 = Lerez, Glenmark Pharmaceuticals; *T_2 = *Lerka,* Nicholas Pharmaceuticals are tablets from manufacturers.

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REFERENCES

1. Seam C. Sweetman (Ed.), "Martindale; The Complete Drug Reference, Pharmaceutical Press, London, U.K., 33rd Edn., (2002) p. 920.
2. The Merck Index, Merck and Co. Inc., White House Station, N. J., 13th Edn., (2001) p. 973
3. G. Bianchi, Pharmacol. Res., **21**, 193 (1989).
4. E. Rimoldi, Acta. Therap., **20**, 23 (1994).

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