April 2007 Volume 5 Issue 1-6

Trade Science Inc.

Analytical CHEMISTRY

An Indian Journal

Full Paper

ACAIJ, 5(1-6), 2007 [93-96]

Validated Simultaneous Estimation Of Telmisartan And Hydrochlorothiazide In Tablet Dosage Form By RP-HPLC

Corresponding Author

P.Shanmugasundaram
Department of Pharmaceutical Analysis, Vel's College of Pharmacy, Pallavaram, Chennai-600117, Tamilnadu, (INDIA)
E-Mail: samsimahe@yahoo.com

Co-Authors

R.Maheswari, V.Gunasekaran, N.Harikrishnan, M.Vijeyaanandhi, K.S.Syed Ali Abtheen
Department of Pharmaceutical Analysis, Vel's College of Pharmacy, Pallavaram, Chennai-600117, Tamilnadu, (INDIA)

Received: 2nd March, 2007
Accepted: 7th March, 2007

Web Publication Date : 15th April, 2007

ABSTRACT

A validated HPLC method for simultaneous estimation of telmisartan and hydrochlorothiazide in tablet formulations is described. Separation was achieved on a gemini C_{18} column(4.6mm×25 cm), in isocratic mode, with phosphate buffer(pH 2.5), acetonitrile and tetrahydrofuran(6:3:1v/v/v) as mobile phase at a flow rate of 1 ml/minute. Quantitation was carried out by the use of UV detector in absorbance mode at 225nm. The retention times of telmisartan and hydrochlorothiazide were found to be 5.612 and 4.23 minutes, respectively. Linearity of detector response for telmisartan and hydrochlorothiazide were found to be from 0.32 to 0.48mg/ml and 0.10 to 0.15mg/ml, respectively. The amounts of drug estimated in the average weight of the tablet were found to be 40.75 and 13.00mg, respectively. The proposed method was validated and was found to be suitable, precise, accurate and reproducible, and can be adopted for routine analysis of telmisartan and hydrochlorothiazide in tablet formulation.

KEYWORDS

Telmisartan;
Hydrochlorothiazide;
RP-HPLC.

INTRODUCTION

Telmisartan[1] (TELM) is chemically 4'-[(1,4'-dimethyl-2'-propyl [2,6'-bi-1H-benzimidazol]-1'-yl) methyl-[1,1'-biphenyl]-2-carboxylic acid. Telmisartan blocks the vasoconstrictor and aldosterone-secreting effects of angiotensin II by selectively blocking the binding of angiotensin II to the AT_1 receptor. Hydrochlorothiazide[1-2] (HCTZ) is chemically 6-chloro-3,4-dihydro-2H-1, 2,4-benzothiadiazine-7-sulfonamide 1,1-dioxide. Hydrochlorothiazide is a diuretic agent and is indicated in the management of hypertension either as the sole therapeutic agent or to enhance the effectiveness of other antihypertensive drugs in the more severe forms of hypertension. Both the drugs are available in combination as tablet, used

Full Paper

for treating hypertension. Literature survey reveals that telmisartan[3] in tablet dosage form and in biological fluids are estimated by HPLC methods. Hydrochlorothiazide[8] is estimated by HPLC, LC-MS and several other methods in biological fluids. No analytical method has been reported for analysis of these two drugs in combination. In the present work, a successful attempt has been made to estimate both these drugs simultaneously by an accurate, precise and less time consuming HPLC method.

MATERIALS AND METHODS

Instrument

High performance liquid chromatograph, Shimadzu HPLC-LC 2010 CHT with class VP version 6.14 with auto injector with injection volume 20µl, UV-visible detector SPD-10A VP. A gemini C_{18} column (4.6mm×25cm) forms stationary phase.

Chemicals and reagents

Tablet formulations containing 40mg of telmisartan and 12.5mg of hydrochlorothiazide were procured from the market. Acetonitrile HPLC grade, tetrahydrofuran HPLC grade, HPLC grade water, potassium dihydrogen orthophosphate AR grade and orthophosphoric acid AR grade were used.

Preparation of buffer

20 mM potassium dihydrogen phosphate solution is prepared in water. The pH is adjusted to 2.5 with orthophosphoric acid.

Preparation of mobile phase

Mobile phase is prepared by mixing 600ml of buffer, 300ml of acetonitrile and 100ml of tetrahydrofuran. The resulting solution is filtered through 0.45 micron filter.

Standard stock solution

Accurately 80±2.0mg of telmisartan and 25±2.0mg of Hydrochlorothiazide are weighed and transferred to 200ml volumetric flask. Dissolved completely and diluted to the required volume with the mobile phase.

Working standard solution

From the standard stock solution 50ml is pipetted out in to 100ml volumetric flask and made up to volume with the mobile phase.

Sample preparation

Twenty tablets are weighed and crushed. The powdered tablet equivalent to 40mg of telmisartan and 12.5mg of hydrochlorothiazide(average weight of tablet) is weighed and transferred to 100ml volumetric flask, dissolved and made up to volume with mobile phase. The solution is filtered through syringe filter.

Assay

20µl of working standard and sample solutions (n=6) were injected in to an injector of liquid chromatograph and peaks were recorded. From the peak, retention time of telmisartan and hydrochlorothiazide were recorded. And also, the amount of drug in the samples(n=6) were calculated. The values are given in the TABLE 1.

TABLE 1: Assay of telmisartan and hydrochloro thiazide

Tablet sample	Label claim (mg)	Amount present (mg/tablet)	% Label claim
Telmisartan	40	40.75*	101.88*
Hydrochlorothiazide	12.5	13.00*	104.00*

*** Mean of six readings**

Assay results

From the replicate analysis(n=6) of two drugs by the proposed method, the percentage label claims for telmisartan and hydrochlorothiazide were found to be 101.88 and 104.00 respectively.

Validation

Linearity and range

For evaluating the linearity range of telmisartan and hydrochlorothiazide, varying concentrations of standard stock solution were diluted with mobile phase to give minimum of five concentrations in the range of 0.32, 0.36, 0.40, 0.44 and 0.48 mg/ml of telmisartan and 0.1, 0.1125, 0.125, 0.1375 and 0.15 mg/ml for hydrochlorothiazide, respectively. A calibration curve was constructed for each sample by plotting the peak areas obtained against the concentration and there was found to exist, linearity in the concentration range of 0.32 to 0.48mg/ml of telmi-

sartan and 0.1 to 0.15 mg/ml of hydrochlorothiazide. The values of correlation coefficient were 0.9995 and 0.999, respectively.

System suitability

As per USP-24, system suitability tests were carried out on freshly prepared standard stock solutions of telmisartan and hydrochlorothiazide.

20µl of both drugs were injected into the chromatograph under the optimized chromatographic conditions and following parameters were studied to evaluate the suitability of the system.

1. Number of theoretical plates, 2. Resolution, 3. Retention time, 4. Tailing factors, 5. Limit of detection and limit of quantification.

The values of system suitability test were shown in TABLE 2.

System precision

System precision of the method was studied by analysis of multiple samplings of working standard solution and expressed as co-efficient of variance (CV) which was not more than 2%. The results are tabulated in TABLE 3.

Method precision

Precision of the method was studied by analysis of multiple samplings of homogenous sample and expressed as co-efficient of variance(CV), and was not more than 2%. The results are tabulated in TABLE 4.

TABLE 2: System suitability

Parameter	TELM	HCTZ
Resolution	3.89	
Retention time (minutes)	5.612	4.23
Asymmetry factor	1.87	1.68
No. of Theoretical plates	12236.7	13864.84

TABLE 3: System precision

S. NO	Areas	
	TELM	HCTZ
1	29079054	10692011
2	29113525	10684500
3	29228451	10691888
4	29336713	10701996
5	29600064	10729100
6	29758441	10764708
Mean	29352708.00	10710700.50
% R.S.D	0.93	0.29

TABLE 4: Method precision

S.NO	%Label claim	
	TELM	HCTZ
1	100.23	100.8
2	100.30	99.20
3	100.75	102.40
4	101.50	99.60
5	100.38	100.48
Mean	100.63	100.5
%R.S.D	0.522	1.24

TABLE 5

Sample	Amt. of std. added (mg)	Amt. of drug recovered (mg)	% Recovery	Mean % recovery
TELM	2	1.980	99.00	
	2	1.992	99.60	99.30
	4	3.970	99.25	
	4	3.984	99.60	99.43
HCTZ	0.625	0.624	99.84	
	0.625	0.620	99.20	99.52
	1.25	1.245	99.60	
	1.25	1.242	99.36	99.48

Recovery study

To ensure the reliability and accuracy of the method, recovery studies were carried out by mixing a known quantity of standard drug with pre-analyzed sample and content were reanalyzed by the proposed method. The values are shown in TABLE 5.

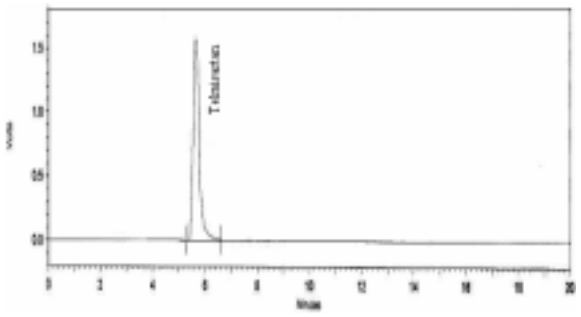
The lower the values of relative standard deviation (RSD) indicate the method is accurate. The mean recoveries of telmisartan and hydrochlorothiazide were 99.37 and 99.50, respectively. Thus shows that there is no positive or negative interference of excipients in tablet.

RESULTS AND DISCUSSION

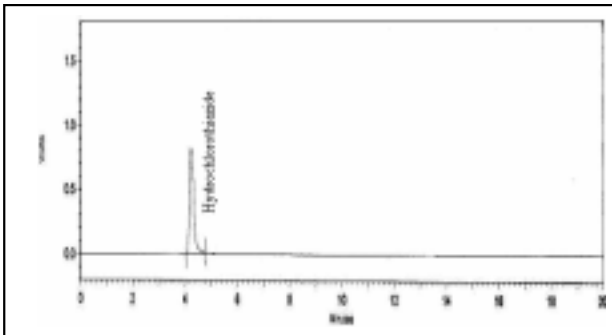
The tables from TABLE 1 to 5 give details of the findings of assay and validation parameters studied.

The mobile phase composing of phosphate buffer of pH 2.5, acetonitrile and tetrahydrofuran in the volume ratio(60:30:10v/v/v) showed good resolution peaks with in a short run time of not more than 8 minutes. As per the current regulatory requirements resolution between the two components should be not less than 3, tailing factor should be not more than 2 and theoretical plates should be more than 2000. It

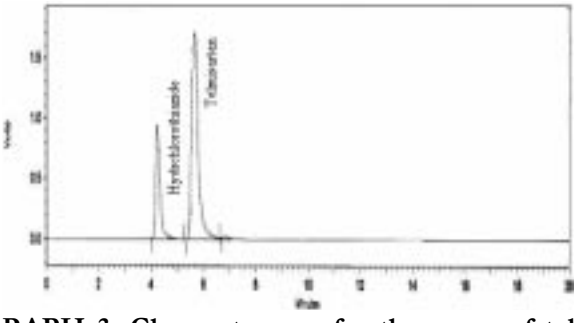
Full Paper



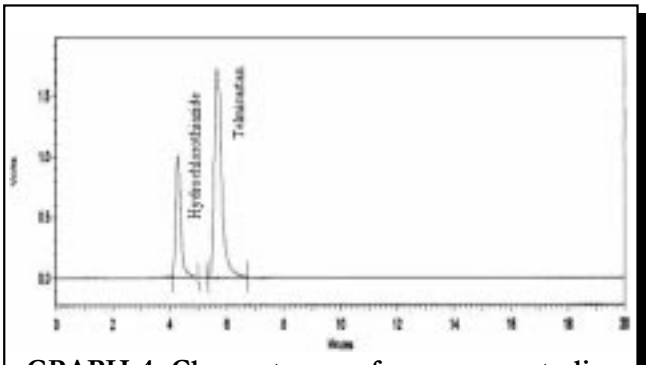
GRAPH 1: Identification chromatogram for telmisartan



GRAPH 2: Identification chromatogram for HCTZ



GRAPH 3: Chromatogram for the assay of telm and HCTZ



GRAPH 4: Chromatogram for recovery studies

is evident from TABLE 2 that the method developed for these drugs in combination is passing the standards of regulatory requirements. The method was found to be precise and accurate with low values of percentage relative standard deviation (<2%).

The TABLE 3 and 4 show results which indicates that the method is precise. The results of recovery study TABLE 5 confirm the accuracy of the method. The proposed RP-HPLC method is accurate, simple, rapid and selective for the simultaneous estimation of telmisartan and hydrochlorothiazide in tablet dosage form. Hence it is conveniently adopted for the routine analysis.

REFERENCES

[1] Anthony C.Moffat, M.David Osselton, Brian Widdop; 'Clarke's Analysis of Drugs and Poisons', Pharmaceutical Press, London, **2**, 1109-1110, 1601-1602 **(2004)**.

[2] Klaus Florey; 'Analytical Profiles of Drug Substances', Academic Press, New York, **10**, 406-435 **(2005)**.

[3] M.S.Palled, P.M.N.Rajesh, M.Chatter, A.R.Bhat; Indian Journal of Pharmaceutical Sciences, **67(1)**, 108-110, **(2005)**.

[4] J.Shen, Z.Jiao, Z.D.Li, X.J.Shi, M.K.Zhong; Pharmazie, **60(6)**, 418-20, **(2005)**.

[5] J.A.Murillo Pulgarin, A.Alanon Molina, G.Perez-Olivares Nieto; Analytica Chimica Acta, **518(1-2)**, 37-43, **(2004)**.

[6] K.Richter, R.Oertel, W.Kirch; Journal of Chromatography A, **729(1-2)**, 293-296 **(1996)**.

[7] Kevin B.Alton, Daniel Desrivieres, James E.Patrick; **374**, 103-110, **(1986)**.

[8] Asbjorg Solberg Christophersen, Knut E. Rasmussen, Bjarne Salvesen; Journal of Chromatography A, **132(1)**, 91-97 **(1977)**.