International Journal of ChemTech Research
CODEN(USA): IJCRGG ISSN : 0974-4290
Vol.4, No.3, pp 1146-1150, July-Sept 2012

Development and Validation of Spectrophotometric Method for Simultaneous Estimation of Atenolol and Hydrochlorothiazide in Pure and Tablet Dosage Form

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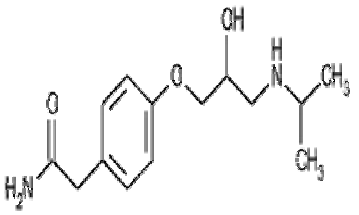
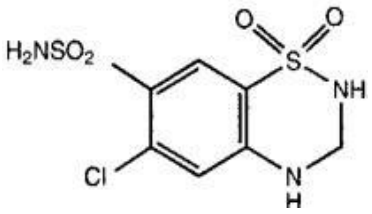
Abstract: The method for the simultaneous estimation of atenolol and hydrochlorothiazide in combined tablet dosage form have been developed, based on absorbance ratio method at two selected wavelengths 232.0nm (Iso-absorptive point) and 272.0nm (λmax of hydrochlorothiazide). The linearity was obtained in the concentration range of 5-30µg/ml and 5-30µg/ml for atenolol and hydrochlorothiazide, respectively. These methods are simple, accurate and results of analysis have been validated statistically and by recovery studies.
Key words: Atenolol, Hydrochlorothiazide, Absorbance ratio method.

INTRODUCTION:

Atenolol (ATN) chemically, 4-(2-.hydroxy-3-isopropyl aminopropoxy)-phenyl acetamide is a β-adrenoreceptor blocking agent, primarily used in hypertension, angina pectoris and myocardial infraction. It mainly acts by inhibition of rennin release and angiotensin-2 & aldosterone production. It is reported to lack intrinsic sympathomimetic activity and membrane-stabilizing properties. The Indian pharmacopeia describes non-aqueous titration method for assay of atenolol. Hydrochlorothiazide (HCTZ), 6-chloro-3, 4-dihydro-2H-1, 2, 4-benzothiadiazine-7-sulfonamide 1, 1-dioxide, which is widely used in antihypertensive pharmaceutical preparations, reduces active sodium reabsorption and peripheral vascular resistance. The review of the literature revealed that no method is yet reported for the simultaneous estimation of both the drugs in combined dosage forms. Present work describes two simple, accurate, reproducible, rapid and economical methods for simultaneous estimation of ATN and HCTZ in tablet formulation.



Atenolol



Hydrochlorothiazide

EXPERIMENTAL:

INSTRUMENTATION:

A double-beam Jasco UV- 2075; UV Visible spectrophotometer, spectral bandwidth of 2nm, wavelength accuracy ±0.5nm and a pair of 1-cm matched quartz cells was used to measure absorbance of the resulting solution.

MATERIALS:

Standard sample of atenolol and hydrochlorothiazide were taken. Combined dose atenolol and hydrochlorothiazide tablets (ATEN-H, 50mg atenolol and 12.5mg hydrochlorothiazide; manufactured by Zydus Cadila) were taken.

SOLVENT

Distilled water selected as common solvent for developing spectral characteristics of drug. The selection was made after assessing the solubility of both the drugs in different solvents.

PREPARATION OF STANDARD STOCK SOLUTIONS:

Atenolol and hydrochlorothiazide (100mg each) were accurately weighed and dissolved separately in100ml of distilled water to give stock (100µg/ml).From the standard stock solution, 1ml each of ATN and HCTZ was taken in 10ml volumetric flask. Volume was made up to mark with distilled water. Aliquot portion was appropriately diluted with distilled water to get final concentration of 5-30µg/ml (HCTZ) and 5-30 µg/ml (ATN) prepared respectively to give final concentrations and scanned between 200-400nm.

METHOD (ABSORBANCE RATIO METHOD):

In the absorbance ratio method, from the overlain spectra of both drugs (fig-3), wavelengths 232.0nm (Iso-absorptive point) and 272.0nm (λmax of hydrochlorothiazide) were selected for analysis. The calibration curves for atenolol and hydro – chlorothiazide were plotted in the concentration range of 5-30µg/ml and 5-30µg/ml at both the wavelengths respectively. The absorbtivities values were determined for both the drugs at both the wavelengths. From the following set of equations the concentration of each component in sample was calculated,

$C\times = Qm-Qy/Qx-Qy \ .A_1/ax_1$ …… (1) and

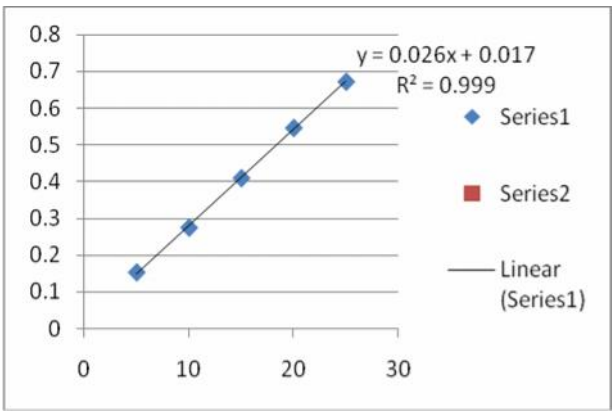
$Cy = Qm-Qx/Qy-Qx \ .A_1/ay_1$ …….(2)

Where

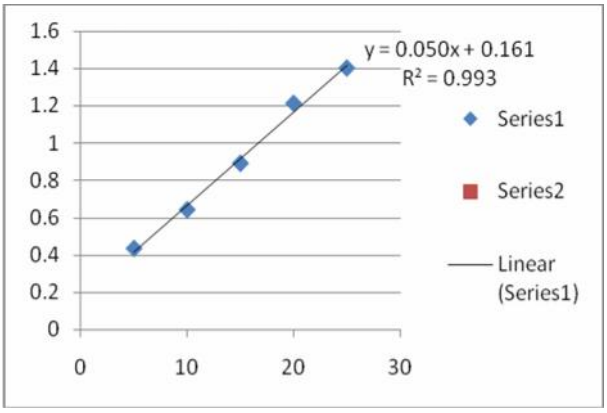
Cx=concentration of atenolol, Cy=concentration of hydrochlorothiazide, A_1=absorbance of sample at iso-absorptive wavelength232.0nm, ax_1= absorbtivity of atenolol at 232.0nm,ay_1=absorbtivity of hydrochlorothiazide at 232.0nm,Qm=ratio of absorbance of sample solution at 232.0nm and 272.0nm, Qx=ratio of absorbtivities of atenolol at 272.0nm and 232.0nm and Qy=ratio of absorbtivities of hydrochlorothiazide at272.0nm and 232.0nm.

APPLICATION OF THE PROPOSED METHOD FOR THE DETERMINATION OF ATN AND HCTZ IN TABLET DOSAGE FORM:

Twenty tablets were weighed and average weight was calculated. The tablets were crushed into fine powder. Tablet powder equivalent to 10mg of ATN was transferred to 100ml volumetric flask and ultrasonicated for 10min .The volume was made up to the mark with distilled water. The resulting solution was then filtered through a whatmann filter paper (No. 41).Aliquot portion was appropriately diluted with distilled water to get final concentration of 15µg/ml . The concentration of both ATN and HCTZ were determined by measuring absorbance of sample at 232.0nm, 272.0nm in spectrum mode and values were substituted in respective formulae to obtain the concentration.



Fig(1): Linearity of Atenolol



Fig(2) Linearity of Hydrochlorothiazide

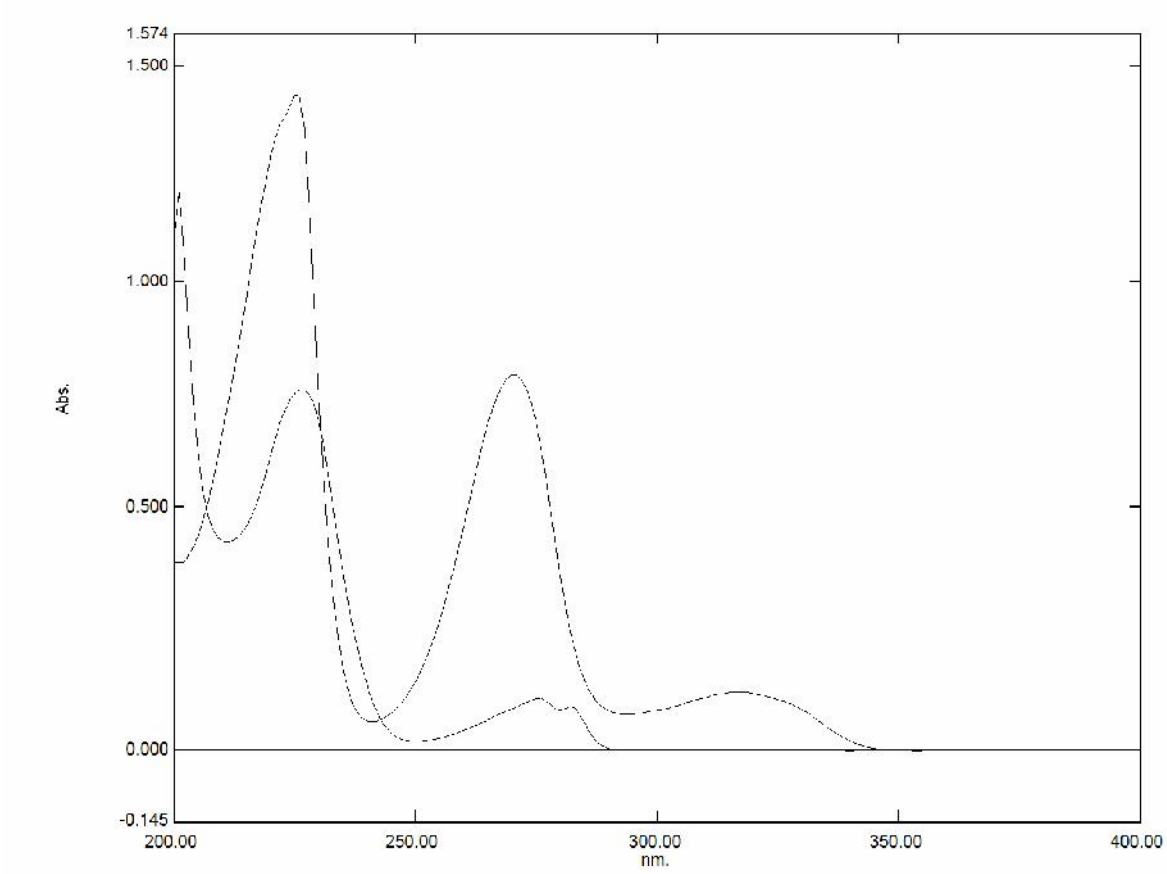


Fig : (3)Absorbance spectra of ACT and HCTZ at wavelength range of 200-400nm

VALIDATION PARAMETER

LINEARITY:
The linearity was obtained in the concentration range of 5-30µg/ml and 5-30µg/ml for atenolol hydrochlorothiazide and respectively in both methods which obeys Beer-Lambert's law. The results of the same are shown in Table (1).

ACCURACY:
To ascertain the accuracy of the proposed methods, recovery studies were carried out by standard addition method at Table (2).

LIMIT OF DETECTION (LOD) AND LIMIT OF QUANTITATION (LOQ)
The LOD and LOQ of paracetamol and meloxicam by proposed methods were determined using calibration standards. LOD and LOQ were calculated as 3.3s/S and 10s/S, respectively, where S is the slope of the calibration curve and s is the standard deviation of response. The results of the same are shown in Table (2).

RESULTS AND DISCUSSION:

From the proposed method, it was found that atenolol and hydrochlorothiazide obeys linearity within the concentration range 5-30µg/ml and 5-30µg/ml respectively. Percentage label claim for ATN and HCTZ in tablet, by both the methods was found in the range of 96.24% to 99%. For Coefficient of variation (CV) were calculated, which was found to be less than 2% indicating the method has good reproducibility. Accuracy of proposed methods was ascertained by recovery studies and results are expressed as %recovery. Percent recovery for ATN and HCTZ was found in range of 96.24% to 99%, values of standard deviation, standard error and coefficient of variation were in range of 0.8 to 0.115, 0.462 to 0.066 and 0.82 to 0.127 respectively indicating the accuracy of proposed method.

CONCLUSION:
Based on the results obtained, it is found that the proposed methods are accurate, precise, reproducible and economical and can be employed for routine quality control of atenolol and hydrochlorothiazide in combined dose tablet formulation.

TABLE NO:1 DATA CHARACTERISTICS OF ATN AND HCTZ

Parameters	Values			
	ATN	**HCTZ**	**ATN at isobestic point**	**HCTZ at isobestic point**
Working λ	224nm	272nm	232nm	232nm
Beer's Law Limit(µg/ml)	5-30	5-30	5-30	5-30
Absorbtivity Value	0.0278	0.0643	0.02295	0.04321
Correlation coefficient	0.993	0.999	0.996	0.995
Intercept	0.161	0.017	0.031	0.169
Slope	0.05021	0.02621	0.020	0.027

ATN-ATENOLOL**, HCTZ**-HYDROCHLOROTHAIZIDE

TABLE NO: 2 RESULTS OF ANALYSIS OF TABLET FORMULATION

Drug	Label Claim (µg/ml)	Amount Found (mg/tab)	Amount Taken		% Recovery	S.D	S.E	C.V	LOD (µg/ml)	LOQ (µg/ml)
			%	mg/ml						
ATN	50mg/tab	49.5	100	50	99.00	0.8	0.42	0.82	0.481	1.59
			80	40	98.90					
HCTZ	12.5mg/ tab	12.03	100	12.5	96.24	0.115	0.066	0.127	0.144	0.438
			80	10	97.20					

S.D : Standard Deviation**, S.E:** Standard Error **,C.V:** Coefficient Variation

ACKNOWLEDGEMENTS:

The author is thankful to Padmashree Dr.Vithalrao Vikhe Patil College Of Pharmacy, Ahmednagar (MH) for providing necessary facilities to carry out research work.

REFERENCES:

1. Veerasekaran V, Katakdhond SJ, Kadam SS, Rao JR, Simultaneous determination of hydrochlorthiazide and metoprolol tartrate from combined dosage form, Indian Drugs, 38,2001,187–189,
2. Barary M. H., Determination of Hydrochloro thiazide and Alprenolol Hydrochloride in combinations, Indian J. Pharm. Sci.,46, 1984, 224.
3. Ja in SK, Jain D, Tiwari M, Chaturvedi SC, Simultaneous spectrophotometric estimation of Propanolol and Hydrochlorothiazide in formulation, Indian J Pharm Sci, 64,2002, 267-276.
4. Suhagia BN, Shah RR, Pate DM, Development of RP-HPLC method for Losartan potassium and Hydrochlorothiazide, Indian J Pharm Sci, 67,2005, 37-42.
5. Sachan A, Jain DK, Trivedi P, Simultaneous estimation of Captopril and hydrochlorothiazide in two components tablets by ultraviolet absorption spectrophotometry, Indian Journal of Pharmaceutical sciences, 59(1),1997, 29-31.
6. Bonazzi D, Gotti R, Andrisano V, Cavrini V,Derivative UV spectrophotometric determination of atenolol and metoprolol in single and multicomponent pharmaceutical dosage forms,Farmaco,51,1996,733.
7. Erram S. V., Tipnis H. P., Simplespectro photometric analysis of Acebutanol hydrochloride and Atenolol in combined pharmaceutical dosages with Hydrochlorothiazide Indian Drugs,30, 1993, 462
8. Prasad CVN, Parihar C, Sunil K, Parimoo P,Simultaneous determination of amiloride HCl, hydrochlorothiazide and atenolol in combined formulations by derivative spectroscopy, J Pharm Biomed Anal, 17,1998, 877-84.
9. Indian Pharmacopoeia, Government of India,Ministry of Health and Family Welfare, Delhi,1996, p. 72.
10. Tripathi K.D., Essential of Medical Pharmacology,5th Ed., Jaypee Brothers Medical Publishers (P)Ltd., New Delhi, 2003.
11. Williams A., Foye's Principles of Medicinal Chemistry, 5th Ed., published by B.I. Publications Pvt. Ltd., 2004.
12. Beckett A.H. and Stenlake J.B., pratical pharmaceutical chemistry, 4th.Edn.The press of university of London, New Delhi, 199, 281.

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