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Research Article

New Spectrophotometric Methods for Simultaneous Determination of Amlodipine besylate and Benazepril in Tablet Dosage Forms

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ABSTRACT

Two simple, accurate, precise, reproducible, requiring no prior separation and economical procedures for simultaneous estimation of Amlodipine besylate (AML) and Benazepril (BEN) in tablet dosage form have been developed. The first method is simultaneous equation method; in this method 360.0 nm and 240.0 nm were selected to measure the absorbance of drugs at both wavelengths. The second method is Q-value analysis based on measurement of absorptivity at 227.50 nm (as an iso-absorptive point) and 360.0 nm at maximum wavelength of AML, 360.0 nm and at isoabsorptive point 227.50 nm both AML and BEN shows linearity in a concentration range of 5-40 µg/mL. Recovery studies range from >99.41% for AMD and >98.93% for BEN in case of simultaneous equation method and >100% for AMD and >99.55% for BEN in case of Q-analysis method confirming the accuracy of the proposed method. The proposed methods are recommended for routine analysis since it is rapid, simple, accurate and also sensitive and specific (no heating and no organic solvent extraction is required).

Keywords: Benazepril (BEN), Amlodipine besylate (AML), Methanol, UV Spectrophotometer.

1. INTRODUCTION

Amlodipine besylate[1], chemically (R, S) 2-[(2-Aminoethoxy) methyl]-4 -(2-chlorophenyl)-1,4-dihydro-6-methyl-3,5-pyridinedicarboxylic acid 3-ethyl 3-methyl ester benzene sulphonate, is a Ca-antagonist which blocks the calcium entry by preventing opening of voltage gated L – type and T – type Ca –channels. It mainly affects heart and smooth muscles inhibiting calcium entry caused by depolarization in these tissues. They also dilate coronary vessels, which is important in variant angina.

Benazepril[2], 3-[[1-(ethoxycarbonyl)-3-phenyl-(1S)propyl]amino]-2,3,4,5-tetrahydro-2-oxo-1H-1-(3S)-benzazepine-1-acetic acid the active metabolite of benazepril, is a nonsulfhydryl angiotensin-converting enzyme (ACE) inhibitors class that is primarily used in treatment of hypertension, congestive heart failure, and heart attacks, and also in preventing renal and retinal complications of diabetes.

Commercially fixed combination of AML (10 mg) and BEN (20 mg) is available in the market as tablet and capsule formulation.

Both the drugs are not official in BP and USP and Literature survey reveals several methods such as HPLC[8], U.V. spectrosctropy[9,10,11], Colorimetric[12] have been reported for individual drugs as well as in combination with other drugs in formulation. However there is no UV- spectrophotometric method reported so far for the simultaneous estimation of these two drugs from the pharmaceutical formulations. A successful attempt has been made to estimate them simultaneously by spectrophotometric analysis.

2. MATERIALS & METHODS

EXPERIMENTAL

A Shimadzu UV/Visible spectrophotometer (Model: UV1700) was employed with spectral bandwidth of 2nm and wavelength accuracy of ± 0.5 nm with automatic wavelength correction with a pair of 10mm quartz cells.

3. MATERIALS AND REAGENTS

Amlodipine besylate (Glenmark Pharmaceuticals Ltd.) Benazepril (Sun Pharmaceuticals) and Methanol – AR grade (Qualigens Fine Chemicals, Mumbai) were used in the study.

4. EXPERIMENTAL PROCEDURE

Method I

Two wavelengths selected for the method are 360.0 nm and 248.0 nm that are absorption maxima of AMD and BEN respectively in methanol. The stock solutions of both the drugs were further diluted separately with methanol to get a series of standard solutions of 5-40 µg /mL concentrations. The absorbances were measured at the selected wavelengths and absorptivities (A 1%, 1 cm) for both the drugs were determined as mean of six independent determinations. Concentrations in the sample were obtained by using following equations.

$$Cx = (A_2ay_1 - A_1ay_2)/(ax_2ay_1 - ax_1ay_2)$$
$$Cy = (A_1ax_2 - A_2ax_1)/(ax_2ay_1 - ax_1ay_2)$$

Where A_1 and A_2 are absorbances of mixture at 360.0 nm and 248.0 nm respectively, ax_1 and ax_2 are absorptivities of AML at λ_1 and λ_2 respectively and ay_1 and ay_2 are absorptivities of BEN at at λ_1 and λ_2 respectively. Cx and Cy are concentration of AML and BEN respectively.

Analysis of tablet formulation

Tablet sample solution was made as per the method described in Method – I and solution was diluted to get a final concentration equivalent to 10µg/ml of AML and 20µg/ml of BEN (n=6) and from the overlain spectra the absorbances were measured at 360.0 nm for AML and 248.0 nm for BEN in spectrophotometric mode of an instrument. Amount of drug present in the sample solution was obtained from the simultaneous equation.

The results of analysis and statistical validation for the marketed tablet formulation are reported in Table-1 and Table-2 respectively. The results of recovery studies conducted by the addition of different amounts of pure drugs at 80%, 100% and 120% levels to a tablet solution were found to be satisfactory and are given in the Table-3.

Method II

Absorption ratio method uses ratio of absorbances at two selected wavelengths, one of which is an 'Isoabsorptive point' and other being the λ_{max} of one of the two components. From the overlain spectra (fig 1) of the two drugs it is evident that AML and BEN shows iso absorptive point at 227.50 nm and the λ_{max} of AML is at 360.5 nm. Hence the two wavelengths selected are 227.50 nm and 360.5 nm.

Six standard solutions of each drug having concentrations 5, 10, 20,30, 40, 50µg/ml were prepared separately in methanol and absorbances at 227.50 nm and 360.5 nm were measured and absorptivity coefficients were calculated using calibration curve. Mixed standards containing 10µg/ml for AML and 20µg/ml (n=6) for BEN were prepared and absorbances at 227.50 nm and 360.5 nm were measured. From the absorbance values the concentration of drug in the pure mixed standard was determined using following formula,

For AML,

$$C_1 = Q_0 - Q_2 / Q_1 - Q_2 \times A/a_1$$

For BEN,

$$C_2 = Q_0 - Q_1 / Q_2 - Q_1 \times A/a_2$$

Where,

Q_0 = Absorbance of mixed standard at 227.50 nm/absorbance of mixed standard at360.5nm

Q_1= Absorbance of AML 227.50 nm / Absorbance of AML 360.5

Q_2= Absorbance of BEN 227.50 nm / Absorbance of BEN at 360.5nm

A = Absorbance of mixed standard at isoabsorptive point

a_2 and a_2 = Absorptivities of AML and BEN respectively.

Analysis of tablet formulation:

Twenty tablets were weighed and ground to a fine power. An accurately weighed powder sample equivalent to 10 mg of AML and 20 mg of BEN was transferred to a 100 ml volumetric flask, dissolved in methanol and volume was made up to the mark with methanol. The solution was kept for the sonication for 20 minutes, filtered through Whatmann filter paper No. 41. Aliquot of this solution was diluted to produce the concentration of 10µg/ml for AML and 20µg/ml for BEN (n=6). The absorbances of sample solutions at 300.00 nm and 360.5 nm were measured and amount of drug present in the sample solution was calculated in the same manner as that of pure mixed standard solution.

The results of analysis and statistical validation for the marketed tablet formulation are reported in Table-

1 and Table-2 respectively. The results of recovery studies conducted by the addition of different amounts of pure drugs at 80%, 100% and 120% levels to a tablet solution were found to be satisfactory and are given in the Table-3.

5. RESULT AND DISCCUSION

The absorption ratio method, also called as Q – analysis, employs the absorption ratio at two selected wavelengths and can be employed for the routine analysis of the two drugs in the combined dosage forms using simple instrument unlike the second method, which requires more accuracy. Second method is used to eliminate the spectral interference from one of the two drugs as the wavelength for estimation of the other drug. This method requires spectral data processing and hence can be performed only on recording spectrophotometers with such facilities.

The amount found from the proposed methods was in good agreement with the label claim of the formulation. Also the value of standard deviation and coefficient of variation calculated were satisfactorily low, indicating the suitability of the proposed methods for the routine estimation of tablet dosage forms.

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Table1: Analysis of tablet formulation

Method	Label claim (mg/tab)				Amount Found* (mg/tab)				Label claim (%)			
	T1		T2		T1		T2		T1		T2	
	AML	BEN	AML	BEN	AML	BEN	AML	BEN	AML	BEN	AML	BEN
I	10	20	10	20	9.59	19.65	10.21	19.59	99.66	99.54	101.12	99.87
II	10	20	10	20	9.88	20.08	9.91	20.06	99.97	100.11	100.33	100.09

T1 = Lotrel (Novartis India Ltd)
T2 = AMACE-BP (Systopic Pvt. Ltd)

Table 2: Statistical validation of tablet formulation

Method	Standard Deviation				% coefficient of variation				Standard error			
	T1		T2		T1		T2		T1		T2	
	AML	BEN	AML	BEN	AML	BEN	AML	BEN	AML	BEN	AML	BEN
I	0.1245	0.1980	0.0569	0.0621	0.55	0.26	0.96	0.85	0.0210	0.0256	0.01957	0.0265
II	0.0965	0.0854	0.5715	0.0841	0.39	0.41	0.57	0.55	0.01585	0.0854	0.02333	0.0244

Table 3: Statistical validation of recovery studies

Method	Type of Recovery in %	Mean ±S D*				Coefficient of variation*				Standard Error*			
		T1		T2		T1		T2		T1		T2	
I		AML	BEN	AML	BEN	AML	BEN	AML	BE N	AML	BEN	AML	BEN
	80	99.23 ±0.13	99.54 ±0.13	100.38 ±0.83	99.50 ±0.43	99.26 ±0.21	99.54 ±0.25	100.83 ±0.52	99.50 ±0.43	99.13± 0.62	99.86 ±0.43	99.36 ±0.14	99.25 ±0.41
	100	99.87 ±0.21	99.80 ±0.15	101.46 ±0.77	99.97 ±0.37	99.82 ±0.19	100.52 ±0.25	99.76± 0.31	99.97 ±0.37	99.26± 0.21	99.65 ±0.37	99.86 ±0.17	99.17 ±0.17
	120	100.10 ±0.21	100.07 ±0.21	100.61 ±1.15	100.09 ±0.43	100.2 1±0.21	100.06 ±0.19	99.16 ±0.27	100.10 ±0.12	99.36 ±0.27	100.10 ±0.2179	100.08 ±0.21	100.09 ±0.11
II	80	100.00 ±0.52	100.05 ±0.12	100.06 ±0.23	99.65 ±0.58	99.69 ±0.51	100.20 ±0.12	99.23 ±0.19	100.09 ±0.52	99.65 ±0.19	100.00 ±0.5292	99.87 ±0.29	99.65 ±0.57
	100	100.26 ±0.32	100.09 ±0230	99.96 ±0.20	100.08 ±0.24	99.68 ±0.56	100.26 ±0.35	100.20 ±0.32	100.26 ±0.30	100.09 ±0.21	100.26 ±0.3055	100.26 ±0.30	99.56 ±0.26
	120	99.87 ±0.30	99.87 ±0.30	100.10 ±0.17	99.68 ±0.35	100.9 1±0.18	99.58 ±0.51	99.09 ±0.21	99.87 ±0.30	100.23 ±0.25	99.87 ±0.3055	100.23 ±0.55	99.68 ±0.39

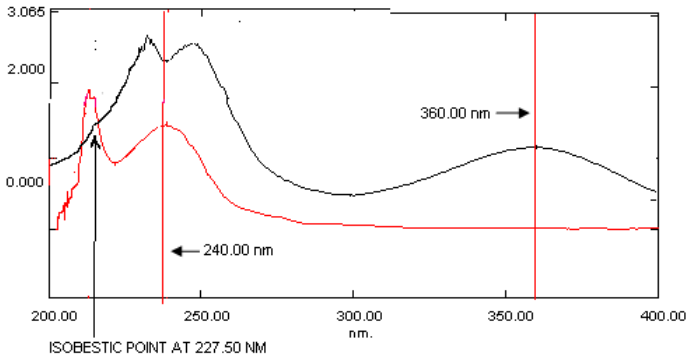

Fig. 1: Overlay spectra of BEN and AML

REFERENCES

1. Beckett AH and Stenlake JB. Practical Pharmaceutical Chemistry. 4th ed. CBS Publishers and distributors, 2002:281-300.
2. A Wolters Kluwer. Drugs Facts and Comparisons. 57th edition. 2003:519, 625.
3. Devid A. Willians and Thomas L. Lemke. Foye's Principles of Medicinal Chemistry. 5th ed. 2006:507.
4. Rang HP, Dale MM, Ritter JM and Moore PK. Pharmacology. 5th ed. 282-283.
5. Willard, Merritt, Dean and Settle. Instrumental Methods of Analysis. 7th ed. CBS Publisher and distributors, 169-178.
6. Goodman and Gilman. The pharmacological basis of therapeutics. 10th ed. 984.
7. Remington, The science and practice of pharmacy. 19th ed. MACK Publishing Company. 1995:593-598.
8. Raja Rajeswari K, Sankar GG, Rao AL and Seshagirirao JVLN. RP-HPLC method for the simultaneous determination of Atorvastatin and Amlodipine in tablet dosage form. Indian Journal of Pharma Science and Research. 2006;68:275-277.
9. Shah DA, Bhatt KK and Mehta RS. Stability indicating RP-HPLC estimation of atorvastatin calcium and amlodipine besylate in pharmaceutical formulations. Indian Journal of Pharma Sci and Res. 2008;70: 754-760.
10. Wankhede SB, Wadkar SB, Raka KC and Chitlange KD. Simultaneous estimation of amlodipine besilate and olmesartan medoxomil in pharmaceutical dosage form. Ind J of Pharm Sci and Res. 2009;71:563-567.
11. Sohan S Chitlange, Mohammed Imran and Dinesh M Sakarkar. RP-HPLC method for Simultaneous estimation of Amlodipine and Metoprolol in tablet formulation. Asian Journal of Pharmaceutics. 2008;2: 232-234.
12. Garg G, Saraf S and Saraf S. Development and validation of simultaneous estimation of Enalapril maleate and Amlodipine besylate in combined dosage forms. Trends Applied Sci Res. 2008;3:278-284.
13. Belal F, Al-Zaagi IA and Abounassif MA. Spectrophotometric determination of benazepril in tablets. Farmaco. 2000l;55(6-7):425-32.
14. Fawzy A El-Yazbi, Heba H Abdine and Rasha A Shaalan. Spectrophotometric methods for the determination of benazepril hydrochloride in its single and multi-component dosage forms. Journal of Pharmaceutical and Biomedical Analysis. 1999;20(1-2):343-350.
15. Budawari, S. The Merck Index.13th Ed. Merck & Co. Whitehouse Station, NJ, 2001:488.