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Simultaneous estimation of metoprolol tartrate and amlodipine besylate by absorbance ratio method

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A simple and cost-effective spectrophotometric method is developed for the determination of metoprolol tartrate and amlodipine besylate in bulk drug and tablets. In the present case the method developed based on absorbance ratio method (Q method). Absorbance ratio method uses the ratio of absorbances at two selected wavelengths, one which is an isoabsorptive point (λ_2) 270 nm and other being the absorption maxima (λ_1) 229 nm of one of the two components. The drugs obeys Beer' law in the concentration range 10–100 μg mL^{-1}. Results of analysis were validated statistically and by recovery studies. The method was found to be suitable for routine determination of metoprolol tartrate and amlodipine besylate in bulk drug and tablets.

Keywords: Metoprolol, amlodipine, spectrophotometric, Q method, β-blockers.

Introduction

Metoprolol tartrate (MT) is chemically (*RS*)-1-isopropylamine-3-*p*-(2-methoxyethyl)phenoxypropan-2-ol(2*R*,3*R*)-tartrate (Fig. 1). It is a cardio-selective β-blocking agent with an intrinsic sympathomimetic activity which slows the heart rate[1,2]. It is one of the potent drugs as its small quantity is enough to aggravate the desired pharmacological action[3]. MT is prepared by chemical synthesis, and the racemate is used clinically[4]. MT is administered orally or intravenously[5].

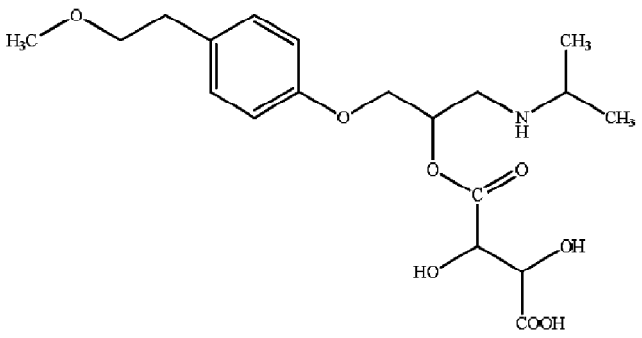

Fig. 1. Structure of metoprolol tartrate.

Amlodipine besylate (AB) is chemically 3-ethyl 5-methyl-(4-*RS*)-2-[(2-aminoethoxy)methyl]-4-(2-chlorophenyl)-6-methyl-1,4-dihydropyridine-3,5-dicarboxlate benzene sulfonate (Fig. 2) a long-acting calcium channel blocker. It is used as

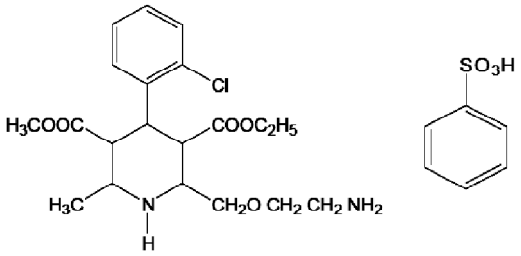

Fig. 2. Structure of amlodipine besylate.

an anti-hypertensive and in the treatment of angina. Amlodipine acts by relaxing the smooth muscle in the arterial wall, decreasing total peripheral resistance and hence reducing blood pressure; in angina, it increases blood flow to the heart muscle[6,7]. The therapeutic importance of these drugs has prompted the development of many methods for its assay. The literature survey revealed titrimetric and spectrophotometric methods[8–15]. Besides, several ways have been reported for quantification in plasma using high-performance liquid chromatography with UV or fluorescence de-

tection[16–22]. Colorimetric determination of some β-blocking drugs has been reported[23–34]. The purpose of this investigation was to develop a simple and sensitive simultaneous spectrophotometric method for the quantitation of MT and AB in pure drug and pharmaceutical formulations. This method uses the well known absorbance ratio method, which is the modification of the simultaneous equation procedure. It depends on the property that, for a substance which obeys Beer's law at all wavelength, the ratio of absorbance at any two wavelengths is a constant value independent of concentration path length.

Results and discussion

The calibration curve of MT and AB individually at 270 nm and 229 nm were plotted. The relationship between the absorbance and the concentration of MT and AB was found to be linear in the range of 10–100 g/mL at both wavelengths 229 nm and 270 nm. The representative linear equations were calculated by the least squares method and the correlation coefficients have indicated very good linearity. Good accuracy of the proposed method was proved by good percent recovery in the standard addition method. It ranged between 99.82 and 102.36%. In the present case the method developed (based on absorbance ratio method (Q method)), wavelengths selected were 229 nm and 270 nm. Two equations are constructed for a method of simultaneous equation. There treatment is somewhat different, however and use the relationship $ax_1 = ay_1$ at λ_1. Assume b = 1 cm.

The concentration of two drugs in the mixture can be calculated using derived simultaenous eqs. (1) and (2).

Using these equations the concentrations of metoprolol tartrate and amlodipine besylate were estimated in commercial formulations. The results of the analysis showed lower values of standard deviation, standard error of mean, coefficient of variation and percentage range of error (within 95% confidence limit) and thus showed the precision of methods. The overlain UV absorption was found to be simple, accurate, precise and economic. Hence the method can be employed for the routine analysis of these two drugs in combined form.

Experimental

Instrumentation:

UV-1800 UV-Vis spectrophotometer, Shimadzu, Japan, equipped with 10 mm matched quartz cells was used in the present investigation. A Citizen scale, Japan analytical balance was used.

Materials and reagent:

Drug procured from IPCA Laboratories Limited Sajavata, Ratlam, Madhya Pradesh. Regd. off. 48, Kandivali Ind. Estate, Mumbai. All the chemicals used during the experiment were of analytical grade.

Preparation of stock solutions:

Metoprolol tartrate 100 mg (accurately weight) was dissolved in 100 mL of ethanol in a volumetric (1000 μg/mL) (S_{MT}) and amlodipine besylate 100 mg (accurately weighed) was separately dissolved in 100 mL of ethanol in a separate volumetric flask (1000 μg/mL) (S_{AB}).

Steps of method development:

Selection of working wavelength:

From stock solution of metoprolol tartrate (MT) and amlodipine besylate (AB). Working standard solution of metoprolol tartrate (100 μg/mL) and amlodipine besylate (100 μg/mL) were prepared by appropriate dilution of the stock solution with ethanol. Aliquots of 40 μg/mL were prepared from the working standard solutions. Both were scanned separately in the range of 200–400 nm. An overlain spectrum of these scans was then recorded.

Absorbance ratio method uses the ratio of absorbances at two selected wavelengths, one which is an isoabsorptive point and other being the absorption maxima of one of the two components. From the overlay spectra of two drugs, it is evident that MT and AB show an isoabsorptive point at 270 nm. The second wavelength used is 229 nm, which is the absorption maximum of MT.

Linearity determination:

Various dilutions of metoprolol tartrate and amlodipine besylate from standard solution were prepared and absorbances were measured at the selected wavelength. Linear-

ity curve for concentration versus absorbance was plotted.

Absorption coefficient of metoprolol tartrate:

Aliquots (1.0, 2.0, 3.0....10 mL) of working standard solution (100 µg/mL) were transferred into a series of 10 mL volumetric flasks. The volumes were made with ethanol. The absorbances were measured at 229 nm and 270 nm. The absorption coefficients were calculated for these concentrations are 0.0196 (a_{X_2}) and 0.0034 (a_{X_1}) (Figs. 3 and 4).

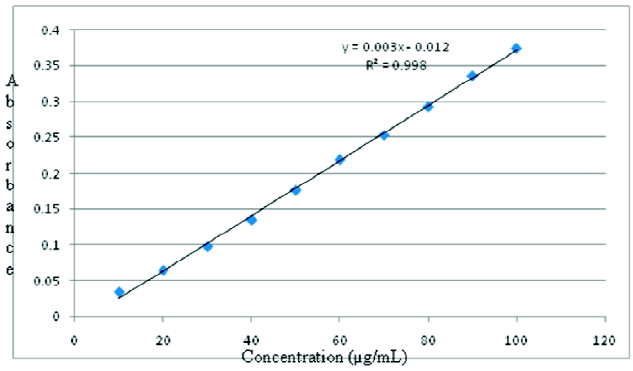

Fig. 3. Calibration curve of metoprolol tartrate at 229 nm.

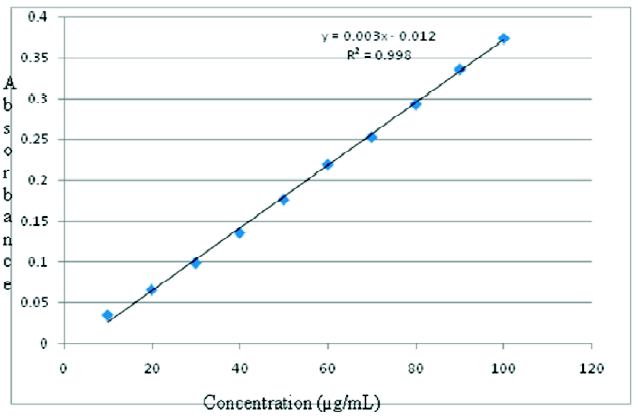

Fig. 4. Calibration curve of metoprolol tartrate at 270 nm.

Absorption coefficient of amlodipine besylate:

Aliquots (1.0, 2.0, 3.0....10 mL) of working standard solution of amlodipine besylate (100 µg/mL) was transferred into a series of 10 mL volumetric flasks. The volumes were made up with ethanol. The absorbances measured at 229 and 270 nm. The absorption coefficients were calculated for these concentrations are 0.0189 (a_{Y_2}) and 0.0034 (a_{Y_1}) (Figs. 5 and 6).

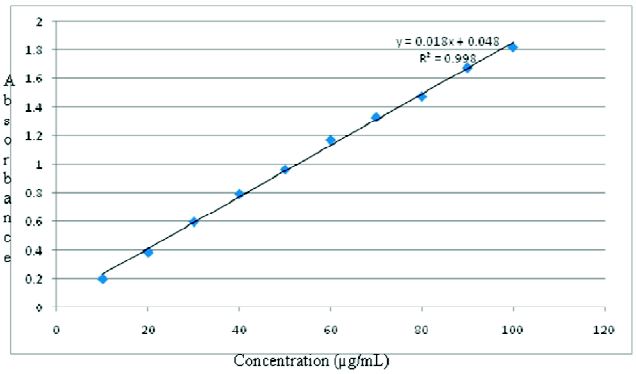

Fig. 5. Calibration curve of amlodipine besylate at 229 nm.

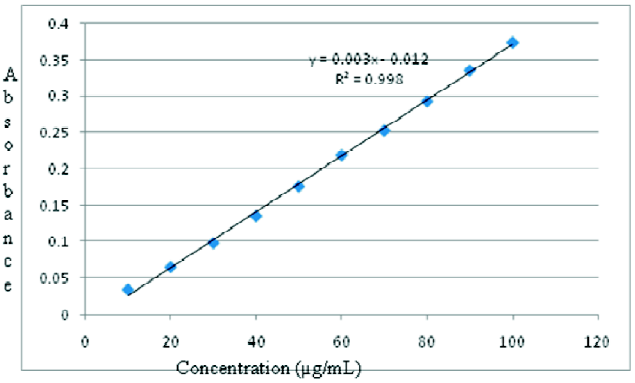

Fig. 6. Calibration curve of amlodipine besylate at 270 nm.

Development of method:

In the quantitative assay of two components in admixture by the absorbance ratio method, absorbance measured at two component (λ_1) and the other being a wavelength of equal absorptivity of the two component (λ_2). The simultaneous equations developed for this method. The value of absorptivities of MT and AB at 229 nm was found to be the same. So $ax_2 = ay_2$ at λ_2, where b = 1 cm.

The concentration of two drugs in the mixture can be calculated using the following equations.

$$CX = [(Q_M - Qy)/(Q_X - Q_Y)] \times A_1/ax_1 \quad (1)$$

$$CY = [(Q_M - Qx)/(Q_Y - Q_X)] \times A_1/a_{Y1} \quad (2)$$

where A_1 and A_2 are absorbances of the mixture at 270 and

Table 1. Compilation of results of statistical analysis of commercial formulation

Tablet brand	Tablet components	Label claim[a] (mg/tab)	Amount found[a] (mg/tab)	SD[a]	CV[a]	SE[a]
AMTAS-M	Metoprolol tartrate	50 mg	49.911±0.3962	0.159	0.318	0.064
	Amlodipine besylate	5 mg	5.118±0.3512	0.123	0.201	0.050

[a]Average of six determinations: S.D.: Standard deviation, C.V.: Coefficient of variation, S.E.: Standard error.

229 nm; ax_1 and a_{Y1} are absorptivities of MT and AB at 270 nm; ax_2 and a_{Y2} are absorptivities of MT and AB at 229 nm;

$Q_M = A_2/A_1$, $Q_X = ax_2/ax_1$ and $Q_Y = a_{Y2}/a_{Y1}$

Estimation from tablets:

The average weight of twenty tablets was determined [AMTAS-M (Intas), 0.1112 g]. These tablets were then finely powdered and triturated well. A quantity of tablet powder equivalent to 100 mg metoprolol tartrate and 10 mg of amlodipine besylate (each tablet contain 50 mg metoprolol tartrate and 5 mg amlodipine besylate) extracted quantitatively with (4×20) mL ethanol. The ethanol extract was filtered into a 100 mL volumetric flask and the volume made up.

Aliquots of a definite concentration were taken in six 10 mL volumetric flasks which were in the Beer's law limit. Absorbance was noted at 229 nm and 270 nm. From these readings and using eqs. (1) and (2). The drug content in tablet (amount found), standard deviation, coefficient of variation, standard error of the mean, percentage range of error (within 95% confidence limits) were calculated.

Recovery study:

Recovery experiments were performed by adding the known amount (10 mg of metoprolol tartrate and 5 mg of amlodipine besylate) of the pure drug to the previously analyzed sample and reanalyzing the mixture by the proposed method. Results are incorporated in Table 1. All results of statistical analysis and recovery studies are incorporated in Table 2.

Table 2. Compilation of results of drug recovery study

Tablet brand	Percentage recovery ± SD[a]	
	Metoprolol tartarate	Amlodipine besylate
AMTAS-M	99.82±0.159	102.36±0.201

[a]Average of six determinations: S.D.: Standard deviation.

Conclusion

The reproducibility, repeatability, and accuracy of this method were found to be good, which is evident by low standard deviation values. The percent recovery experiment values obtained indicates non-interference from the excipients used in the formulations. The percentage recovery was close to 100% for these methods. Thus, it can be concluded that the method developed was simple, accurate, sensitive and precise. Hence, the above method can be applied successfully in simultaneous estimation of metoprolol tartrate and amlodipine besylate in marketed formulations.

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References

1. C. G. Regårdh, *Acta Pharmacol. Toxicol. (Copenh)*, 1975, **37(1)**, 1.
2. D. L. Geffner and J. M. Hershman, *Am. J. Med.*, 1992, **93(1)**, 61.
3. S. Chen, A. C. Fu, R. Jain and H. Tan, *Am. Health Drug Benefits*, 2015, **8(2)**, 71.
4. H. U. Shetty and W. L. Nelson, *J. Med. Chem.,* 1988, **31(1)**, 55.
5. W. Bensinger, R. T. Maziarz, S. Jagannath, A. Spencer, S. Durrant, P. S. Becker, B. Ewald, S. Bilic, J. Rediske, J. Baeck and E. A. Stadtmauer*, Br. J. Haematol.,* 2012, **159(1)**, 58.
6. B. J. Kim, S. U. Kwon, D. Wajsbrot, J. Koo, J. M. Park and B. W. Jeffers, *J. Am. Heart Assoc.*, 2018, **7(24)**.
7. F. Takahashi, M. Kobayashi, A. Kobayashi, K. Kobayashi and H. Asamura, *Molecules*, 2018, **7(23)**.
8. S. S. M. Hassan, M. M. Abou-Sekkina, M. A. El-Ries and A. A. Wassel, *J. Pharm. Biomed. Anal.,* 2003, **32**, 175.
9. S. Khalil and M. M. El-Rabiehi, *J. Pharm. Biomed. Anal.,* 2000, **22**, 7.

10. A. Golcu, D. Tarinc and M. Cesme, *Pharmaceuticals,* 2011, **4**, 964.
11. M. I. Walash, A. M. El-Brashy, M. E. Metwally and A. Abdelal, *Farmaco,* 2004, **59**, 493.
12. K. Gohil, P. Trivedi and K. I. Molvi, *Indian J. Pharma. Sci.,* 2005, **67**, 376.
13. G. Ragno, A. Garofalo and C. Vetuschi, *J. Pharm. Biomed. Anal.*, 2002, **27**, 19.
14. K. Sridhar, C. S. P. Sastry and M. N. Reddy, *Anal. Lett*., 1997, **30**, 121.
15. S. A. Khopade and N. K. Jain, *Indian Drugs*, 2000, **37**, 351.
16. V. G. Dongre, S. B. Shah, P. P. Karmuse, M. Phadke and V. K. Jadhav, *J. Pharm. Biomed. Anal.,* 2008, **46**, 583.
17. P. Mishra, A. Gupta and K. Shah, *Indian J. Pharma. Sci.,* 2007, **69**, 831.
18. P. Mishra, A. Gupta and K. Shah, *International Journal of Pharmacy and Pharmaceutical Sciences,* 2009, **1**, 55.
19. M. Aqil, A. Ali, A. Ahad, Y. Sultana, A. K. Najmi and N. Saha, *Acta Chromatogr.*, 2007, **19**, 130.
20. B. K. Durga, N. I. Mounika, S. Shajhan, S. N. Rao and R. Kuchi, *International Journal of Science Innovations and Discoveries*, 2011, **1**, 151.
21. V. G. Dongrea, S. B. Shaha, P. P. Karmusea, M. Phadkeb and V. K. Jadhav, *J. Pharm. Biomed. Anal.*, 2008, **46**, 583.
22. A. K. Sarkar, D. Ghosh, A. Das, P. S. Selvan, K. V. Gowda, U. Mandal, A. Bose, S. Agarwal, U. Bhaumik and T. K. Pal, *J. Chromatogr. B: Analyt. Technol. Biomed. Life Sci*., 2008, **873**, 77.
23. B. R. Simmons and J. T. Stewart, *J. Liq. Chromatogr.,* 1994, **17**, 2675.
24. P. Hubert, P. Chiap, M. Moors, B. Bourguignon, D. L. Massart and J. Crommen, *J. Chromatogr. A*, 1994, **665**, 87.
25. B. Mistry, J. Lesile and N. E. Eddington, *J. Pharm. Biomed. Anal.,* 1998, **16**, 1041.
26. V. L. Herring, T. L. Bastian and R. L. Lalonde, *J. Chromatogr.*, 1991, **567**, 221.
27. R. J. Straka, K. A. Johnson, P. S. Marshall and R. P. Remmel, *J. Chromatogr.*, 1990, **530**, 83.
28. A. F. A. Nabil and S. M. Eman, *International Journal of Chemical Studies*, 2015, **3**, 24.
29. S. A. Pagar, D. M. Shinkar and R. B. Saudagar, *Int. J. Pharm. Bio. Sci.,* 2013, **3**, 224.
30. C. A. R. Salamanca Neto, A. P. P. Eisele, V. G. Resta, J. Scremin and E. R. Sartori, *Sens. Actuators B: Chem.*, 2016, **230**, 630.
31. D. K. Sharma and P. Raj, *Int. J. Pharm. Sci. Res.,* 2017, **8**, 1000.
32. I. Baranowska, W. Adolf and S. Magiera*, J. Chromatogr. B: Anal. Technol. Biomed. Life Sci.*, 2015, **1004**, 79.
33. F. Sharma, H. Jain, V. Kanzariya and U. Upadhyay, *Asian J. Pharm. Clin. Res.*, 2014, **7**, 38.
34. D. K. Sharma, J. Singh and P. Raj, *International Journal of Pharmacy and Pharmaceutical Sciences*, 2018, **10**, 107.