Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Pharmaceutical and Biomedical Analysis 37 (2005) 627–630
JOURNAL OF PHARMACEUTICAL AND BIOMEDICAL ANALYSIS
www.elsevier.com/locate/jpba

Short communication

Quantitative analysis of enalapril by ^{1}H NMR spectroscopy in tablets

Ariana Zoppi, Marcela Linares, Marcela Longhi*

Departamento de Farmacia, Facultad de Ciencias Químicas, Universidad Nacional de Córdoba, Ciudad Universitaria, 5000 Córdoba, Argentina

Received 12 November 2004; accepted 12 November 2004
Available online 25 December 2004

Abstract

A simple, rapid, accurate and selective ^{1}H NMR method was developed for quantitative determination of enalapril maleate in pharmaceutical preparations. Spectra were determined in D_2O, using L-leucine as internal standard. Both synthetic mixtures and commercial dosage forms were assayed, and the results were compared to those obtained using the USP XXIV procedure and were both in close agreement.

Keywords: Enalapril; ^{1}H NMR spectroscopy

1. Introduction

Enalapril is an angiotensin-converting enzyme inhibitor widely used in the treatment of hypertension and heart failure. Enalapril is a pro-drug. Following oral administration, it is bioactivated by hydrolysis of the ethyl ester to enalaprilat, which is the active drug. Enalapril is used in oral dosage forms since its oral absorption is superior to that of enalaprilat [1]. Methods use for the assay of enalapril in pharmaceutical samples include spectrophotometry [2–4], potentiometry [5,6] and HPLC [7–9]. In the USP 24, the assay of the pure drug and its tablets also relies on HPLC determination [10]. The procedure here proposed uses ^{1}H NMR spectroscopy. No isolation is required, and good quantitative results were also obtained for the determination of enalapril in the presence of enalaprilat, its major degradation product in solid state [1]. The applicability of the method was checked on several commercial tablets, and the results were compared to those obtained by the USP procedure.

* Corresponding author. Tel.: +54 351 433 4163; fax: +54 351 433 4127x115.
E-mail address: mrlcor@dqo.fcq.unc.edu.ar (M. Longhi).

0731-7085/$ – see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.jpba.2004.11.027

2. Experimental

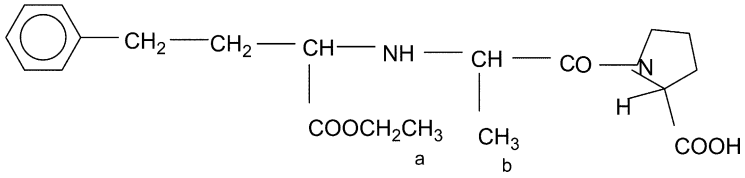
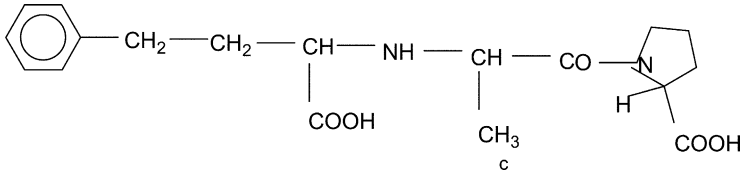
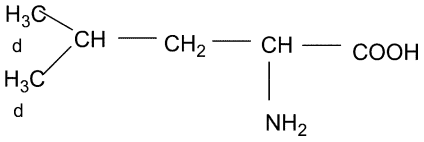
2.1. Materials

- Enalapril maleate was obtained from Parafarm (Argentina) and was used after purification by recrystallization. Internal standard L-leucine and deuterated water, D_2O (99.96%) were obtained from Aldrich.
- Enalaprilate was obtained by hydrolysis of enalapril in a 10N NaOH solution and was characterized by IR and ^{1}H NMR spectrocopies.
- Commercial tablets of enalapril maleate were purchased from different local firms.

2.2. Instrumentation

- NMR; Bruker AC 200; 200.13 MHz.
- HPLC; Spectra System P2000 liquid chromatograph including 7725 rheodyne injector (20 μl loop), peak simple chromatography data system, peak system for Windows 1.85, UV–vis detector. A 5 μm packing C-18 column (250 mm × 4.6 mm) from Supelco was used for the separations.

Table 1
^{1}H NMR resonance assignments and chemical shifts of enalapril, enalaprilat, and L-leucine in D_2O

Component	Structure and peaks integrated for analysis	Number of protons involved in integration	Chemical shifts, δ (ppm)	Multiplicity
Enalapril		$a = 3$, $b = 3$	1.38, 1.59	Triplet, d-doublet
Enalaprilat		$c = 3$	1.49	d-Doublet
L-Leucine		$d = 6$	1.02	Doublet

2.3. Assay preparation for NMR

Twenty tablets of each commercial product were weighed and finely powdered. A portion of well-mixed powder equivalent to 10 mg of enalapril maleate was weighed accurately and transferred to a glass stoppered tube, and about 0.7–1.0 ml of D_2O were added. The solution was mixed by means of a vortex mixer and centrifuged. Using an automatic pipette, about 0.5 ml of the clean supernatant solution was transferred to an analytical NMR tube containing about 10 mg of accurately weighed L-leucine, and the spectrum was recorded.

3. Calculations

The amount of enalapril per unit dose was calculated from the following equation:

$$W_E = \frac{W_L}{W_{st}} \frac{E_E}{E_L} \frac{A_E}{A_L} T$$

where A_E is the integral value of enalapril, A_L the integral value of L-leucine, the internal standard, E_E the formula weight of enalapril divided by the number of absorbing protons, E_L the formula weight of L-leucine divided by the number of absorbing protons, W_L the weight of L-leucine taken for the assay (mg), W_{st} the quantity of the tablet sample taken for the assay (mg), T the average tablet weight (mg), and W_E is the weight of enalapril in the tablet sample.

4. Results and discussion

Fig. 1 shows the 200 MHz ^{1}H NMR spectra of enalapril, enalaprilat, L-leucine, and their mixture in D_2O. Spectral

Table 2
Recovery of enalapril by ^{1}H NMR spectroscopy

Sample number	L-Leucine (mg)	Enalaprilat (mg)	Enalapril		Recovery (%)
			Added (mg)	Found (mg)	
1	10.2	9.82	10.0	10.1	101
2	10.1	10.0	5.53	5.39	97.5
3	10.2	10.3	15.3	15.3	100
4	15.5	10.1	10.2	10.3	101
5	15.3	9.91	5.31	5.25	98.9
6	15.1	10.2	15.2	15.2	100
7	10.4	15.1	10.5	10.4	99.1
8	10.1	14.9	5.04	4.88	96.8
9	10	15.4	14.8	14.9	101
Average (%)					99.5
S.D.					1.54

Table 3
Determination and statistical results of the developed ^{1}H NMR and USP XXIV methods

Formulation	Enalapril labelled strength (mg)	Statistical parameters ($n = 5$)					
		NMR method			USP XXIV method		
		Mean	%	S.D.	Mean	%	S.D.
Eritril (Northia)	10.0	9.88	98.8	1.70	9.98	99.8	1.01
Gadopril (Gador)	10.0	9.92	99.2	1.03	9.97	99.7	0.96
Nalapril (Klonal)	10.0	9.83	98.3	2.15	10.4	104	1.81

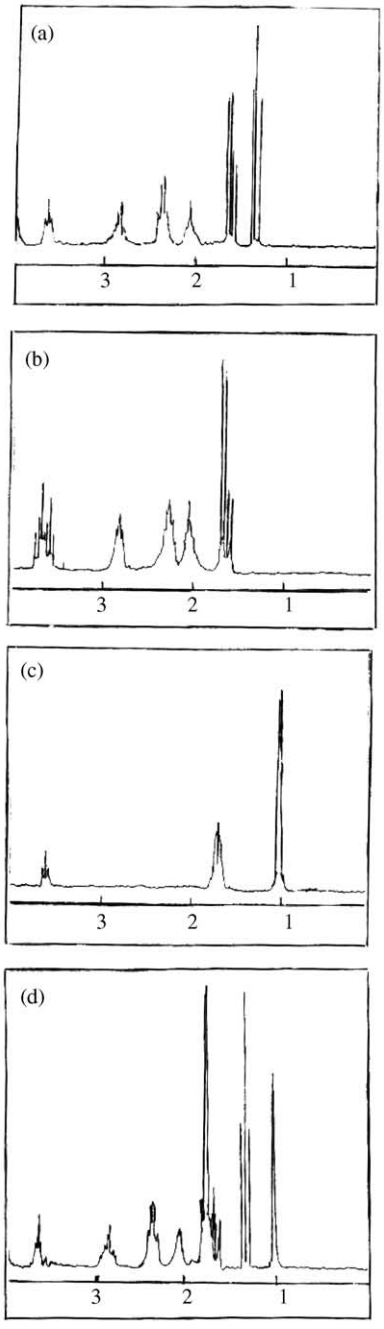


Fig. 1. ^{1}H NMR spectra of enalapril (a), enalaprilat (b), L-leucine (c), and the mixture of the three of them (d) in D_2O.

assignments and chemical shifts are summarized in Table 1. Among the various resonances, those for the methyl protons of the ethyl ester of enalapril at δ 1.38, and the methyl protons of L-leucine at δ 1.02 were selected as analytical signals for quantitative purposes.

The method was standardized by analyzing nine mixtures of known composition of enalapril with enalaprilat and L-leucine. The amounts used are shown in Table 2, and analyzed by the proposed NMR method. The average recovery of enalapril was 99.5%, with standard deviation of 1.54. These results indicated that the relative proportions of enalaprilat and internal standard have no influence on method accuracy since enalapril was recovered quantitatively.

By the proposed NMR method, the assay of commercial products, consisting of tablets from three companies, yielded the results presented in Table 3. The enalapril content ranged from 98.3 to 99.2% (mean 98.8%) of the amount declared. The efficacy of the proposed method was checked by comparing the results with those obtained by the USP XXIV HPLC method and were both in close agreement (Table 3).

5. Conclusion

Enalapril maleate content of a pharmaceutical dosage form can be determined by ^{1}H NMR with the use of L-leucine as the internal standard. The method is selective, simple and rapid compared to previously reported methods.

Acknowledgements

The authors thank the Secretaría de Ciencia y Técnica de la Universidad Nacional de Córdoba (SECyT), and the Agencia Córdoba Ciencia for financial support.

References

[1] P.I. Dominic, S.B. Gerald, in: K. Florey (Ed.), Analytical Profiles of Drug Substances, vol. 16, Academic Press, New York, 1987, pp. 207–243.
[2] O. Abdel Razak, S.F. Belal, M.M. Bedair, N.S. Barakat, R.S. Haggag, J. Pharm. Biomed. Anal. 31 (2003) 701–711.
[3] M.M. Ayad, A.A. Shalaby, H.E. Abdellatef, M.M. Hosny, J. Pharm. Biomed. Anal. 28 (2002) 311–321.

[4] S.M. Blaih, H.H. Abdine, F.A. El-Yazbi, R.A. Shaalan, Spectrosc. Lett. 33 (2000) 91–102.
[5] H.Y. Aboul-Enein, A.A. Bunaciu, C. Bala, S. Fleschin, Anal. Lett. 30 (1997) 1999–2008.
[6] H.Y. Aboul-Enein, R.I. Stefan, J.F. van Staten, Analysis 27 (1999) 53–56.
[7] H.L. Rau, N. Udupa, A.R. Aroor, Indian Drugs 29 (1991) 46–48.
[8] R.T. Sane, A.J. Vaidya, J.K. Ghadge, A.B. Jani, A.K. Kotwal, Indian Drugs 29 (1992) 244–245.
[9] E. Anzenbacherova, P. Anzabacher, K. Macek, J. Kventina, J. Pharm. Biomed. Anal. 24 (2001) 1151–1156.
[10] The United States Pharmacopeia, XXIV ed., United States Pharmacopeial Convention Inc., Rockville, MD, USA, 2000.