Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Pharmaceutical and Biomedical Analysis
34 (2004) 35–44
JOURNAL OF PHARMACEUTICAL AND BIOMEDICAL ANALYSIS
www.elsevier.com/locate/jpba

Determination of lisinopril in dosage forms and spiked human plasma through derivatization with 7-chloro-4-nitrobenzo-2-oxa-1,3-diazole (NBD-Cl) followed by spectrophotometry or HPLC with fluorimetric detection

Ali A. El-Emam [a,*], Steen Honoré Hansen [b], Mohamed A. Moustafa [c], Saadia M. El-Ashry [d], Dina T. El-Sherbiny [d]

[a] *Department of Pharmaceutical Chemistry, College of Pharmacy, King Saud University, P.O. Box 2457, Riyadh 11451, Saudi Arabia*
[b] *Department of Analytical Chemistry, Royal Danish School of Pharmacy, Copenhagen, Denmark*
[c] *Department of Pharmaceutical Chemistry, Faculty of Pharmacy, King Abdulaziz University, Jeddah, Saudi Arabia*
[d] *Department of Medicinal Chemistry, Faculty of Pharmacy, Mansoura University, Mansoura, Egypt*

Received 28 March 2003; received in revised form 21 August 2003; accepted 24 August 2003

Abstract

Two sensitive, simple and specific methods based on spectrophotometry and reversed-phase HPLC with fluorimetric detection are described for the determination of lisinopril in dosage forms as well as in spiked human plasma using solid phase extraction (SPE) procedures. Both methods are based on the derivatization of lisinopril with 7-chloro-4-nitrobenzo-2-oxa-1,3-diazole (NBD-Cl) in borate buffer of pH 9 to yield a yellow, fluorescent product. The spectrophotometric method depends on measuring the formed yellow color at 470 nm after optimization of the reaction conditions. The HPLC method is based on measurement of the derivatized product using fluorescence detection at 540 nm (excitation at 470 nm). The separation of the derivatized drug, the excess reagent and the internal standard (bumetanide) was performed on a reversed-phase ODS column using isocratic elution with methanol–0.02 M sodium dihydrogen phosphate, pH 3.0 (55:45, v/v) at a flow rate of 1.0 ml/min. The calibration graphs were linear over the concentration ranges 2–20 or 0.02–3.2 μg/ml of lisinopril with minimum detectability of 0.3 and 0.008 μg/ml (6.1×10^{-7} and 1.7×10^{-8} M) for the spectrophotometric and the HPLC methods, respectively. The proposed methods were applied without any interference from the tablet excipients for the determination of lisinopril in dosage forms, either alone or co-formulated with hydrochlorothiazide. Furthermore, the use of the HPLC method was extended to the in vitro determination of the drug in spiked human plasma. Interference from endogenous amino acids has been overcomed by using the solid phase extraction technique, the percentage recovery ($n = 6$) was 101.6 ± 3.35.

Keywords: Lisinopril; NBD-Cl; Spectrophotometry; HPLC; Fluorimetric detection; Dosage forms; Plasma; SPE

* Corresponding author. Tel.: +966-1-467-7350; fax: +966-1-467-6220.
E-mail address: elemam5@hotmail.com (A.A. El-Emam).

0731-7085/$ – see front matter © 2003 Elsevier B.V. All rights reserved.
doi:10.1016/j.japna.2003.08.021

1. Introduction

Lisinopril, 1-[*N*-[(*S*)-1-carboxy–3-phenylpropyl]-L-lysyl]-L-proline dihydrate, is an ACE inhibitor used in the treatment of hypertension and heart failure, in prophylactic treatment after myocardial infarction and in diabetic nephropathy [1] Lisinopril is a subject of monograph in the United States Pharmacopoeia (USP) [2], which recommends an HPLC method for both the raw material and the formulations.

The literature is enriched with a variety of analytical methods for determination of lisinopril in pharmaceutical preparations, such as HPLC [3,4], gas chromatography (GC) [5], and capillary electrophoresis [6–9]. Lisinopril has been frequently determined in dosage forms using spectrophotometry [4,10–13] and spectrofluorometry using *o*-phthalaldehyde [4] and fluorescamine [14] as derivatizing agents and via its reaction with acetyl acetone and formaldehyde [12]. UV derivative spectrophotometry has been utilized for lisinopril determination either alone [15] or in binary mixture with amlodipine [16,17] or hydrochlorothiazide [18,19].

Few methods have been described for determination of lisinopril in biological fluids, including; an HPLC method for its determination in urine [20], gas chromatographic methods [21,22] and fluoroimmunoassay [23] for its determination in serum. Recently, Tsakalof et al. [24] described a liquid chromatography–mass spectrometry (LC–MS) method for monitoring lisinopril in serum, the limit of detection was 6 ng/ml. Lisinopril is weakly absorbing light in the UV region [25] and is thus subject to interference from excipients and/or impurities. The problem is more aggravated if it is required to determine the drug in biological fluids. Therefore, the aim of this work is to develop simple, sensitive and specific methods for the determination of lisinopril in dosage forms as well as in spiked human plasma samples. The two methods are based on the derivatization of lisinopril with 7-chloro-4-nitrobenzo-2-oxa-1,3-diazole (NBD-Cl) in alkaline medium. However, a similar method was described for the determination of ramipril (ACE inhibitor) in tablets and human plasma using 7-fluoro-4-nitro-2-oxa-1,3-diazole (NBD-F) which is similar and more reactive reagent but it is more expensive [26]. The proposed HPLC method is comparable to the previously reported chromatographic methods regarding sensitivity and limit of detection.

2. Experimental

2.1. Instrumentation

- A Merck Hitachi chromatograph model L-7100 equipped with a Rheodyne injector valve with a 20 μl loop, a L-7400 UV detector and a L-7485 fluorescence detector (Darmstadt, Germany). Retention times, peak areas, fluorescence spectra were recorded on a Merck Hitachi D-7500 integrator. Mobile phases were degassed using Merck solvent L-7612 degasser. The flow rate was 1.0 ml/min.
- A Hibar®, Lichrosorb® RP-18 (5 μm) pre-packed column 250 mm × 4 mm combined with a guard column (Merck) was used. All chromatographic solutions were filtered through 0.45 μm HA membrane filter (Millipore, Ireland).
- A Shimadzu UV 1601 PC spectrophotometer was used for the spectrophotometric measurement.
- A RF-1501 Shimadzu spectrofluorimeter with a xenon arc lamp was used to obtain the excitation and emission spectra of the derivatized drug.

2.2. Materials and reagents

All materials and reagents used were of analytical reagent grade. Lisinopril dihydrate was kindly provided by Sedico Pharmaceuticals, Egypt. Zestril tablets from Sedico Pharmaceuticals under license of Zeneca Ltd., UK containing 10 and 20 mg lisinopril anhydrous per tablet, and Zestrotic tablets from Sedico Pharmaceuticals under license of Zeneca Ltd. containing lisinopril dihydrate equivalent to 20 mg lisinopril dihydrate and 12.5 mg hydrochlorothiazide per tablet were purchased from local pharmacy. A reagent solution of 0.3% 7-chloro-4-nitrobenzo-2-oxa-1,3-diazole (Sigma, St. Louis, USA) solution was prepared in methanol. Bumetanide of Ph. Eur. quality was used as internal standard. A 0.2 M borate buffer of pH 9 was used. Methanol HPLC grade was obtained from Merck. A 0.02 M sodium dihydrogen phosphate from Prolabo (Paris, France) was prepared. The mobile phase was a mixture of methanol–0.02 M sodium dihydrogen phosphate adjusted to pH 3.0 by phosphoric

acid (55:45, v/v). Sep-Pak® C18 solid phase extraction (SPE) cartridges were from Waters Corporation, Milford, MA, USA. All chromatographic solutions were filtered through 0.45 μm membrane filter.

2.3. Preparation of standard solutions

A stock solution of lisinopril dihydrate containing 0.1 mg/ml was prepared in methanol and was further diluted with methanol as appropriate.

2.4. Preparation of calibration standard

2.4.1. For the spectrophotometric method

To a set of 10 ml volumetric flasks, increasing volumes from the stock solution of the drug were quantitatively transferred so as to contain the drug within the concentration range 2–20 μg/ml. To each flask 4 ml of borate buffer of pH 9 followed by 1.6±0.2 ml of NBD-Cl solution (0.3%) was added. The solutions were heated for 30 min at 70 °C. The reaction was quenched by cooling under tap water, then 0.2 ml of concentrated HCl was added and each flask was completed to volume with methanol. The absorbance was measured at 470 nm against a reagent blank. The calibration curve was constructed by plotting the absorbance against the final concentration of the drug. Alternatively, the corresponding regression equation was derived.

2.4.2. For the HPLC method

Transfer 1.0 ml of the previously derivatized solutions into a set of 10 ml volumetric flasks so as to contain the drug within the concentration range 0.02–3.6 μg/ml, then add 1 ml of the internal standard (IS) bumetanide solution 1.0 mg/ml. Complete to the mark with the mobile phase. The separation was affected at ambient temperature (25 °C). Twenty microliters aliquots were injected (triplicate) and the calibration curve was constructed by plotting the peak area ratios (derivatized drug peak area/internal standard peak area) against the final concentration of the drug. Alternatively, the corresponding regression equation was derived.

2.5. Sample preparation

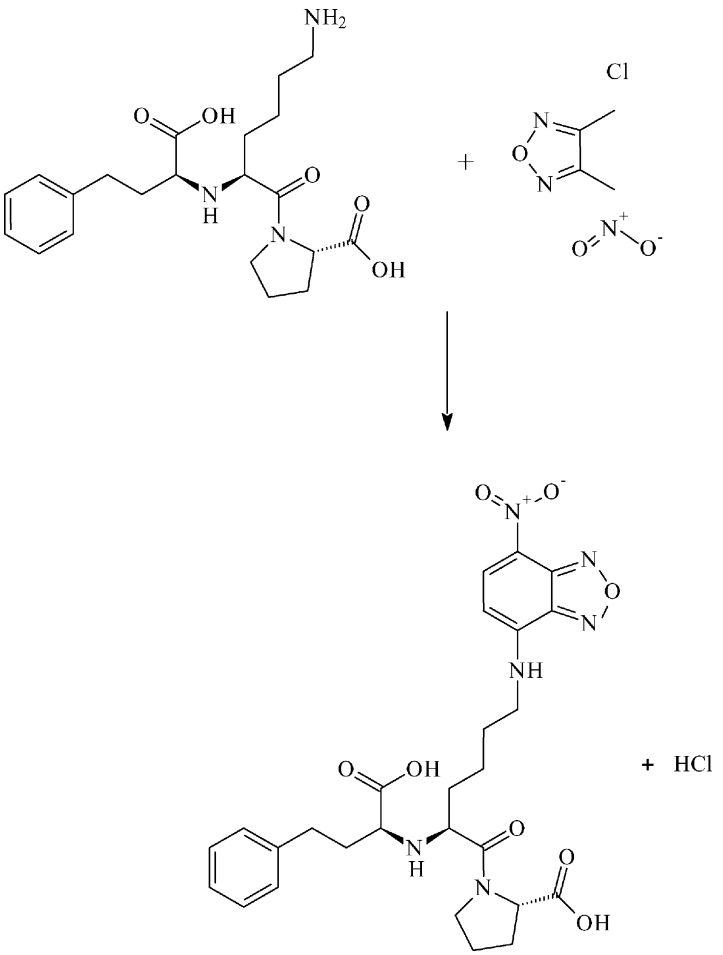
An accurately weighed amount of the powdered tablets equivalent to 10 or 20 mg lisinopril was transferred into a small flask with 50 ml methanol and stirred for 45 min. The extract was filtered into 100 ml volumetric flask and completed to volume with methanol. Further dilution of the filtrate were made as appropriate with methanol then proceed as described under preparation of calibration curve either for the spectrophotometric or the HPLC method. The nominal contents of the tablets were calculated using either the calibration graph or the corresponding regression equation.

2.6. Assay of plasma

The method of solid phase extraction described by Leis et al. [21,22] has been adopted. One milliliter of the spiked plasma was mixed with 2 ml of 0.1 M HCl, then applied to a preconditioned Sep-Pak® C18 SPE cartridges (200 mg), Preconditioning of each cartridge was performed with methanol (6 ml) and then with 0.1 M HCl (3 ml). After application of the acidified sample the cartridge was washed with three times with 3 ml of 0.1 M HCl and two times with 2 ml CH_2Cl_2 and the analyte was then eluted with 2 ml of methanol into a set of 10 ml volumetric flasks. Proceed as described in Section 2.4.2.

3. Results and discussion

Lisinopril exhibits a very low UV absorption with A 1%, 1 cm at 258 nm = 5.1 and 4.5 in 0.1 M sodium hydroxide and 0.1 M hydrochloric acid, respectively [25]. As a consequence, poor sensitivity is achieved by direct spectrophotometric measurement. Determination of the drug in biological fluids using this technique will not be possible due to many interferences. Lisinopril contains both primary and secondary amino groups which are known to react with NBD-Cl in alkaline medium to yield a fluorescent derivative [27]. By analogy to previous reports [28,29], the reaction is proposed to proceed as shown in Scheme 1. Due to interfering signals from the reagent and side reactions it was decided to apply HPLC with fluorimetric detection to get higher sensitivity and complete resolution of the peaks corresponding to the derivatized drug and the peaks corresponding to the reagent and side reactions was achieved. This was of major importance when the method was applied to the determina-



Scheme 1. A proposal of the reaction pathway between lisinopril and NBD-Cl.

tion of lisinopril in plasma where some endogenous amino acids might interfere.

3.1. Optimization of the reaction conditions

The reaction between lisinopril and NBD-Cl in borate buffer of pH 9.0 produces a yellow colored product with maximum absorbance at 470 nm. The different experimental parameters affecting the intensity of the color produced were studied and optimized to obtain maximum color intensity.

The pH was varied over the pH range of 7–10 using borate buffer where the maximum absorbance was obtained at pH 9.0 as shown in Fig. 1, NBD-Cl is hydrolyzed in alkaline medium to give NBD-OH which has a maximum absorbance at 462 nm. Therefore, it was necessary to acidify the reaction mixture to pH 2 (by adding 0.2 ml concentrated HCl) before the measurement was carried out. At this acidic pH, the reagent blank did not show any significant absorption peak between 420 and 500 nm [29].

The effect of temperature on the color intensity was studied in the range from 50 to 100 °C for different periods of time. It was found that heating at relatively lower temperature (70 °C) for longer period of time (30 min) gave more reproducible results as illustrated in Table 1, which is in agreement with previous studies [28,29].

The effect of concentration of NBD-Cl on the color intensity was studied over the range 0.1–0.4%. It was

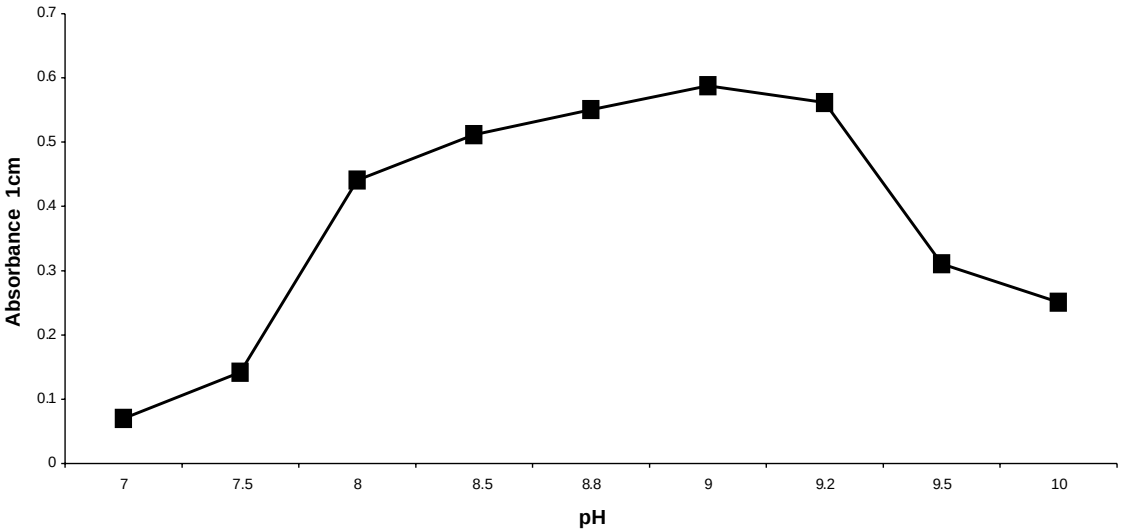


Fig. 1. Effect of pH on the development of the reaction product of lisinopril (15 μg/ml) with NBD-Cl.

Table 1
Factors affecting the formation of the reaction product of lisinopril (10 μg/ml) with NBD-Cl

Heating temperature (°C)	Absorbance
Effect of heating temperature	
50	0.221
60	0.439
70	0.445
80	0.512
100	Precipitation
Heating time (min)	
Effect of heating time at 70 °C	
5	0.086
10	0.167
15	0.242
20	0.337
25	0.443
30	0.443
40	0.442
50	0.446
60	0.441
Concentration of NBD-Cl (%)	
Effect of NBD-Cl concentration (using 1 ml)	
0.10	0.089
0.20	0.312
0.25	0.345
0.30	0.421
0.40	0.444
0.3% NBD-Cl (ml)	
Effect of volume of NBD-Cl	
0.4	0.126
0.8	0.240
1.0	0.411
1.4	0.443
1.8	0.446
2.0	0.445
2.5	0.444
3.0	0.442

found that 1.6 ± 0.2 ml of 0.3% NBD-Cl solution was the most suitable volume of the reagent as illustrated in Table 1. These optimum conditions have been used for the spectrophotometric method and the precolumn derivatization for the HPLC method. Regarding the stability of the produced derivative, it was found to be stable for at least 3 h.

3.2. Stoichiometry of the reaction

The stoichiometry of the reaction was studied adopting the limiting logarithmic method [30]. The two straight lines obtained upon using increasing concentrations of the drug while keeping the concentration of the reagent constant (Fig. 2A) and upon using increasing concentrations of the reagents while keeping

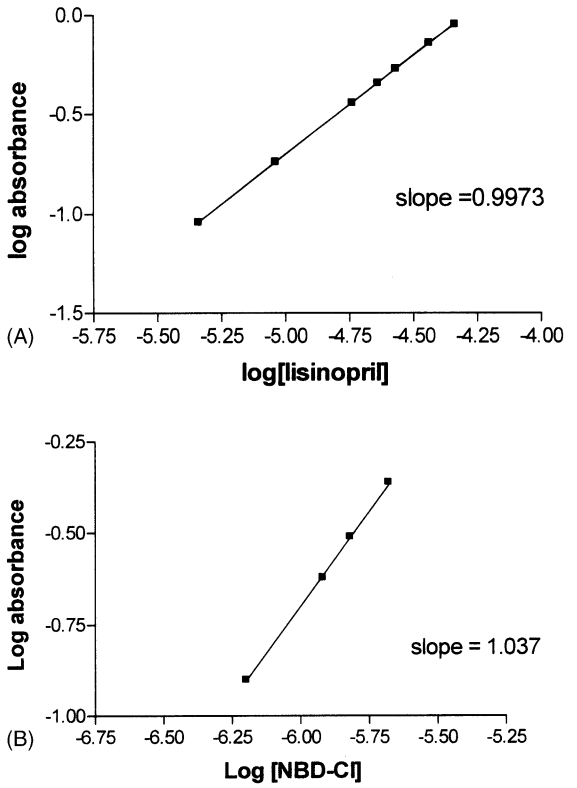


Fig. 2. Limiting logarithmic plots for the molar reactivity of lisinopril with NBD-Cl: (A) log(absorbance) vs. log[lisinopril] with [NBD-Cl] kept at 2.26×10^{-6} M; (B) log(absorbance) vs. log[NBD-Cl] with [lisinopril] kept at 2.26×10^{-5} M.

the concentration of the drug constant (Fig. 2B). The two lines gave two slopes with the values of 0.9973 and 1.037, respectively, therefore the molar reactivity of the reaction is 0.9973:1.037, i.e. 1:1. Hence the reaction pathway in Scheme 1 was proposed. Only the primary amino group of the drug reacts with the active chloride of NBD-Cl with formation of benzofurazon by analogy to previous reports [28,29], while the reaction of the secondary amino group is retarded by the steric hindrance effect.

3.3. HPLC method development

A simple and reliable HPLC method was developed for the determination of lisinopril in single- and multi-component dosage forms and in spiked human plasma. The separation and resolution of the peaks could be achieved upon using a mixture of 0.02 M sodium dihydrogen phosphate and methanol (45:55, v/v). The effect of pH on the retention of the solutes was investigated by changing the pH of sodium phosphate buffer over the range from 3.0 to 7.0. The optimum separation was accomplished upon using solution of pH 3.0 ± 0.2.

3.4. Characteristics of the chromatographic peak

According to the conditions described, the retention times were about 3.60, 5.40 and 14.60 min for NBD-Cl, derivatized drug and IS (bumetanide), respectively. Typical chromatogram of lisinopril after derivatization is shown in Fig. 3.

3.5. Validation

3.5.1. Linearity

Both methods were tested for linearity by using the above mentioned procedures. Linear regression equations were obtained over the ranges given in Table 2. The regression plots showed that there is a linear dependence of the analytical response both for the UV absorbance and for the peak area ratios on the concentration for the spectrophotometric and the HPLC methods, respectively. The table also shows the detection limits as well as the slopes, the intercepts and the correlation coefficients obtained by the linear least square treatment of the results along with standard deviation of slopes (S_b) and intercept (S_a) on the ordinate and the standard deviation of the residuals ($S_{y/x}$) [31].

3.5.2. Accuracy and precision

The inter- and intra-day accuracy and precision for both methods are summarized in Table 3. The inter- and intra-day precision were examined by analysis of authentic samples of lisinopril with the concentrations 10 and 16 μg/ml for the spectrophotometric method and 0.2 and 0.8 μg/ml for the HPLC method (each $n = 5$) for three consecutive days. As shown in Table 3, the repeatability and reproducibility of both methods is fairly good as indicated by the low values of S.D. and R.S.D.%, respectively. The specificity of both methods was proved by observing no interference encountered from the excipients of the tablets, or from other co-formulated drugs such as hydrochlorothiazide. The robustness of the two methods is demonstrated by the

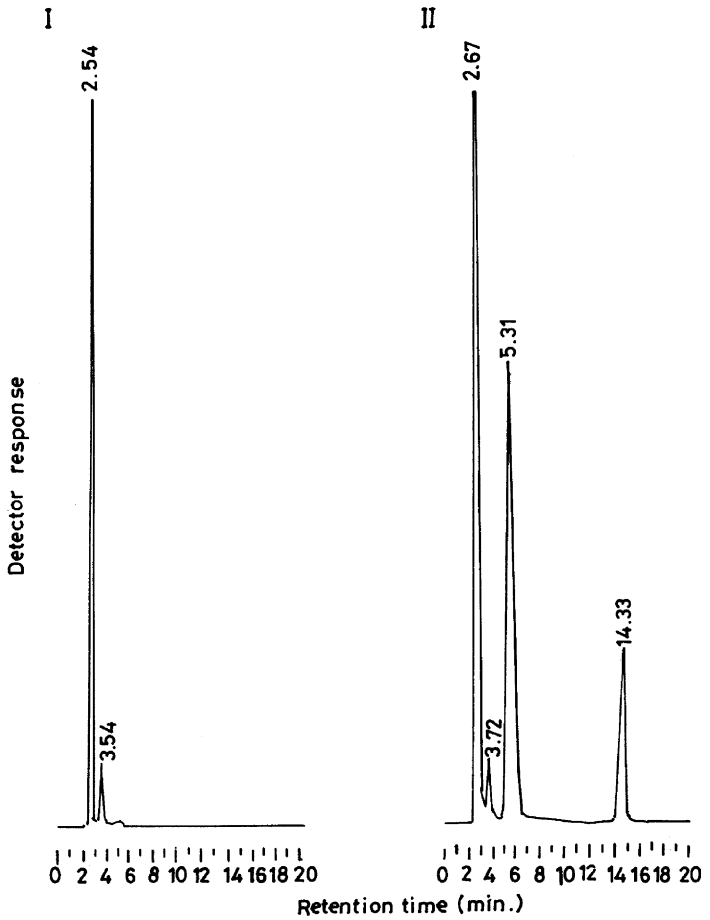


Fig. 3. Typical chromatograms of: (I) blank reagent; (II) derivatized drug (0.8 μg/ml, 5.31 min) and internal standard bumetanide (100 μg/ml, 14.33 min). Flow rate, 1 ml/min; fluorescence detection: excitation, 470 nm and emission, 540 nm; column temperature, ambient.

Table 2
Analytical parameters for the assay of lisinopril via derivatization with NBD-Cl adopting the proposed spectrophotometric and HPLC methods

Parameter	Spectrophotometric method	HPLC method
Wavelength (Λ_{max}, nm)	470	470/540
Concentration range (μg/ml)	2−20	0.02–3.6
Regression equation		
Intercept (a)	1.54×10^{-3}	0.0139
Slope (b)	0.045	4.15
Correlation coefficient (r)	0.9999	0.9999
$S_{y/x}$	0.00352	0.00594
S_a	2.68×10^{-3}	2.43×10^{-3}
S_b	2.26×10^{-4}	1.976×10^{-3}
Detection limits (μg/ml)	0.27 (6.12×10^{-7} M)	0.0077 (1.73×10^{-8} M)

Table 3
Evaluation of the accuracy and precision of the proposed methods for the determination of lisinopril via derivatization with NBD-Cl

Concentration added (μg/ml)	Concentration found (μg/ml)			
	Mean ± S.D.[a]	Recovery (%)	R.S.D.%[b]	Error (%)[c]
HPLC				
Intra-day				
0.200	0.20 ± 0.002	100.9 ± 0.86	0.85	0.38
0.800	0.80 ± 0.006	100.2 ± 0.72	0.72	0.32
Inter-day				
0.200	0.20 ± 0.001	100.3 ± 0.62	0.62	0.36
0.800	0.81 ± 0.005	101.2 ± 0.60	0.59	0.34
Spectrophotometry				
Intra-day				
10.000	10.05 ± 0.044	100.5 ± 0.44	0.43	0.19
16.000	15.98 ± 0.054	99.9 ± 0.34	0.34	0.15
Inter-day				
10.000	10.03 ± 0.037	100.3 ± 0.37	0.36	0.21
16.000	15.98 ± 0.030	99.9 ± 0.19	0.19	0.11

[a] Mean ± S.D. ($n = 5$ for intra-day, $n = 3$ for inter-day).
[b] Relative S.D.
[c] Percentage relative error.

versatility of the experimental factors that affect the absorbance intensity for the derivatization reaction.

3.6. Applications

3.6.1. Assay of dosage forms

The applicability of both methods was tested by determination of lisinopril in its dosage forms either single or as binary mixture with hydrochlorothiazide. The results obtained are accurate and precise as indicated by the excellent percentage recovery and S.D. < 2 (Table 4).Common excipients in tablets, such as talc, lactose, starch, avisil, gelatin or magnesium stearate did not interfere with the assay. Statistical analysis of the results obtained by both methods and by those given by the reference UV derivative spectrophotometric method [22,32] was performed using the Student's *t*-test and the Variance

Table 4
Application of the proposed method to the determination of the studied compounds in dosage forms via derivatization with NBD-Cl

Pharmaceutical preparations	Recovery[a] ± S.D. (%)		
	Proposed method		Reference method [15,32]
	Spectrophotometry	HPLC	
Zestril tablets[b] 10 mg (lisinopril anhydrous 10 mg per tablet)	99.9 ± 0.57 ($t = 1.62$, $F = 1.04$)	100.0 ± 0.38 ($t = 1.9$,$F = 2.33$)	100.8 ± 0.58
Zestril tablets[b] 20 mg (lisinopril anhydrous 20 mg per tablet).	100.3 ± 0.42 ($t = 0.21$, $F = 2.94$)	100.5 ± 0.46 ($t = 0.63$, $F = 2.48$)	100.2 ± 0.72
Zestrotic tablets[b] (lisinopril dihydrate equivalent to 20 mg anhydrous lisinopril and 12.5 mg hydrochlorothiazide)	98.9 ± 0.27 ($t = 2.06$, $F = 1.30$)	98.8 ± 0.38 ($t = 2.39$, $F = 1.16$)	98.0 ± 0.41

Theoretical values of t and F at $P = 0.05$ are 2.776 and 19.00, respectively.
[a] Mean recovery of three separate determinations.
[b] Product of Sedico Pharmaceuticals, under license of Zeneca Ltd.

ratio *F*-test. The calculated values did not exceed the theoretical ones, indicating no significance difference between the compared methods regarding accuracy and precision, respectively (Table 4).

3.6.2. Application to spiked plasma

Calibration graphs prepared from data obtained from the analysis of spiked plasma were linear over the range 0.05–1.6 μg/ml. Linear regression analysis of the data gave the following regression equation:

$$Y = -0.066 + 3.27C, \qquad r = 0.9999$$

where Y is the peak area ratio of drug to IS and C the concentration of drug in μg/ml.

The percentage recovery of lisinopril was 101.63 ($n = 6$) with a coefficient of variation of 3.35%, with a limit of detection of 0.017 μg/ml.

The high sensitivity of the proposed HPLC method allowed the determination of the studied compound in spiked human plasma. The oral dose of lisinopril ranges from 5 to 40 mg daily (single or multiple dosages) [33]. The usual maintenance dose is 10–20 mg given once daily [1]. Utilizing the radioimmunoassay (RIA) procedures [25] for serum, the peak

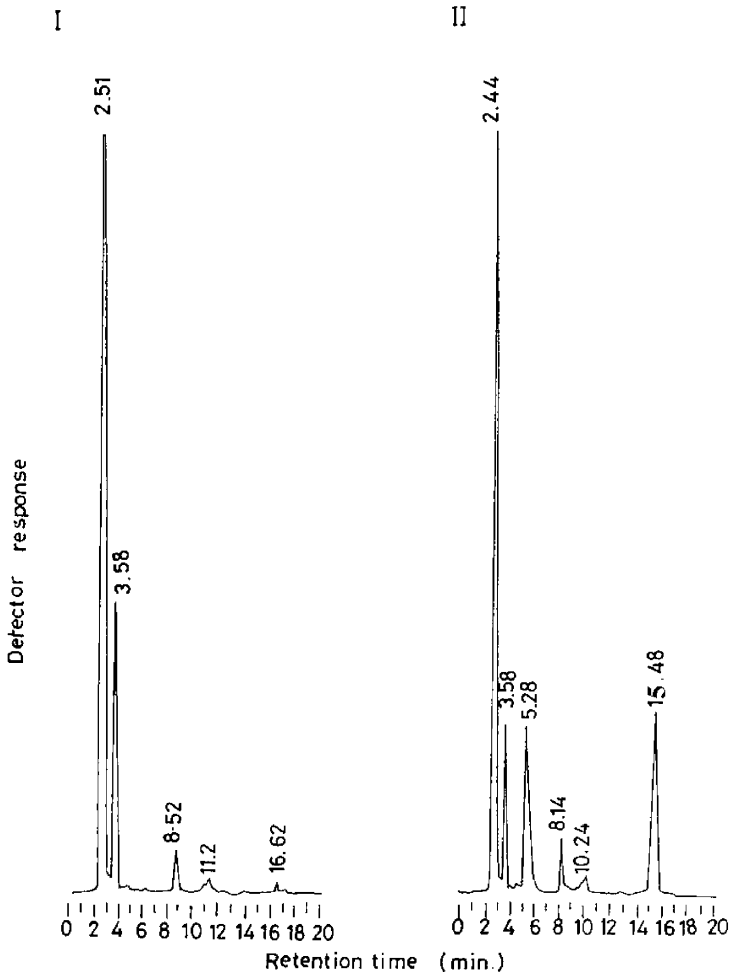


Fig. 4. Typical chromatograms of: (I) blank plasma; (II) derivatized drug (0.4 μg/ml, 5.28 min) and internal standard bumetanide (100 μg/ml, 15.48 min) extracted from spiked plasma. Flow rate, 1 ml/min; fluorescence detection: excitation, 470 nm and emission, 540 nm; column temperature, ambient.

Table 5
Application of the proposed HPLC method for the determination of lisinopril in spiked human plasma via derivatization with NBD-Cl

Concentration added (μg)	Concentration found (μg)	Percentage found[a]
0.0500	0.0541	108.20
0.1000	0.1037	103.70
0.2000	0.1992	99.60
0.4000	0.3954	98.85
0.8000	0.7932	99.15
1.6000	1.6040	100.25
$\bar{X} \pm$ S.D.		101.63 ± 3.35

[a] Each result is the average of three separate determinations.

serum concentration following oral administration of 10 mg capsule of lisinopril was determined within 0–72 h and it was found to be 0.75 ± 0.36 μg/ml. This means that the drug level in plasma is above the limit of quantitation of the proposed HPLC method. The interference arising from the endogenous amino acids has been overcome by the efficient separation using the reported SPE procedure [21]. Fig. 4 represents a typical chromatogram of the derivatized lisinopril in spiked plasma sample. The results of analysis of spiked human plasma samples are shown in Table 5.

4. Conclusion

The two described methods are simple, reproducible and sensitive. The HPLC method exhibits the highest sensitivity that makes it possible to measure concentrations down to 0.02 μg/ml with detection limit of 0.008 μg/ml. This value is comparable to other GC and HPLC methods reported in the literature.

References

[1] K. Parfitt, The Complete Drug Reference, 32nd ed., The Pharmaceutical Press, Martindalle, MA, 1999, 898 pp.
[2] The United States Pharmacopeia XXIV, National Formulary 19, The Pharmaceutical Convention, Rockville, 2001.
[3] R.T. Sane, G.R. Valiyare, U.M. Deshmukh, S.R. Singh, R. Sodhi, Indian Drugs 29 (1992) 558–560.
[4] A. El-Gindy, A. Ashour, L. Abdel-Fattah, M.M. Shabana, J. Pharm. Biomed. Anal. 25 (2001) 913–922.
[5] A.B. Avadhamulu, A.R.R. Pantulu, Indian Drugs 30 (1993) 646–649.
[6] R. Gotti, V. Andrisano, V. Cavrini, C. Bertucci, S. Fulanetto, J. Pharm. Biomed. Anal. 22 (2000) 423–431.
[7] S. Hillaert, W. van-den-Bossche, J. Chromatogr. A 895 (2000) 33–42.
[8] X.Z. Qin, D.S.T. Nguyen, D.P. Ip, J. Liq. Chromatogr. 16 (1993) 3713–3734.
[9] D. Bonazzi, R. Gotti, V. Andrisano, V. Cavrini, J. Pharm. Biomed. Anal. 16 (1997) 431–438.
[10] G. Iskender, B. Yarenci, Acta Pharm. Turc. 37 (1995) 5–8.
[11] S. Atmaca, S. Tartar, G. Iskender, Acta Pharm. Turc. 36 (1994) 13–16.
[12] F.A. El-Yazbi, H.H. Abdine, R.A. Shaalan, J. Pharm. Biomed. Anal. 19 (1999) 819–827.
[13] G. Paraskivas, J. Atta-Politou, M. Koupparis, J. Pharm. Biomed. Anal. 29 (2002) 865–872.
[14] G. Iskender, B. Yarenci, Acta Pharm. Turc. 38 (1996) 65–68.
[15] D. Ozer, H. Senel, J. Pharm. Biomed. Anal. 21 (1999) 692–695.
[16] C.V.N. Prasad, R.N. Saha, P. Parimoo, Pharm. Pharmacol. Commun. 5 (1999) 383–388.
[17] H.K. Jain, R.K. Agrawal, Indian Drugs 37 (2000) 196–199.
[18] P.D. Panzade, L.R. Mahadik, Indian Drugs 36 (1999) 321–323.
[19] N. Erk, Spectrosc. Lett. 31 (1999) 633–645.
[20] Y. Wong, B.G. Charles, J. Chromatogr. B 75 (1986) 512–516.
[21] H.J. Leis, G. Fauler, G. Raspotnig, W. Windischhofer, Rapid Commun. Mass Spectrom. 12 (1998) 1591–1594.
[22] H.J. Leis, G. Fauler, G. Raspotnig, W. Windischhofer, Rapid Commun. Mass Spectrom. 13 (1999) 650–653.
[23] A.S. Yuan, J.D. Gilberti, J. Pharm. Biomed. Anal. 14 (1996) 773–781.
[24] A. Tsakalof, K. Pairachtari, M. Georgarakis, J. Chromatogr. B 784 (2003) 425–432.
[25] D.P. Ip, J.D. De Marco, M.A. Brooks, in: H.G. Britain (Ed.), Analytical Profile of Drug Substances, vol. 21, Academic Press, London, 1992, p. 234.
[26] A.A. Almajed, G. Al-Zehouri, Il Farmaco 65 (2001) 291–296.
[27] M. Pesez, J. Batros, Colorimetric and Fluorimetric Analysis of Organic Compounds and Drugs, Marcel Dekker, New York, 1974, p. 170.
[28] L. De-La-Pena, A. Gomez-Hens, D. Perez-Bendito, J. Pharm. Biomed. Anal. 13 (1995) 199–203.
[29] H.M. Saleh, S.M. Al-Ghanam, Alex. J. Pharm. Sci. 14 (2000) 25–32.
[30] J. Rose, Advanced Physico-Chemical Experiments, Pitman, London, 1964, p. 67.
[31] J.C. Miller, J.N. Miller, Statistics for Analytical Chemistry, Wiley, New York, 1984.
[32] Analysis certificate from Sedico Laboratories for solid preparations, personal communication.
[33] J.G. Hardman, A.G. Gilman, L.E. Limbird, in: L.S. Goodman, L.E. Limbird, P.B. Milinoff, R.W. Ruddan, A. Goodman Gilman, J.G. Hardman (Eds.), The Pharmacological Basis of Therapeutics, 9th ed., McGraw-Hill, New York, 1996, p. 745.