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Quantitative analysis of cefradine by liquid chromatography on poly(styrene-divinylbenzene)

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Abstract: A method is described for isocratic analysis of cefradine by liquid chromatography on a poly(styrene-divinylbenzene) column (PLRP-S, 250 × 4.6 mm i.d.) at 50°C. Cefradine is separated from its related substances using a mobile phase of acetonitrile–0.02 M sodium 1-octanesulphonate–0.2 M phosphoric acid–water (14.5:10:5:up to 100, v/v/v/v). The flow rate was 1.0 ml min^{-1} and UV-detection was performed at 254 nm. The method was employed for the quantitative analysis of reference substances, bulk samples and pharmaceutical dosage forms.

Keywords: *Cefradine; liquid chromatography; poly(styrene-divinylbenzene).*

Introduction

Cefradine is a semi-synthetic β-lactam antibiotic belonging to the group of the cephalosporins. The relative unstability of the molecule and its complex degradation pathways explain the need for a discriminating liquid chromatography (LC) method. The most important related substance, always present in cefradine, is the antibiotic cefalexin, which also gives origin to degradation products.

A number of papers on LC of cephalosporins describe the determination of cefradine in biological samples [1–5]. These methods highlight the separation of the antibiotic from the background of biological material. The determination of cefalexin in cefradine samples is also reported [1, 6, 7]. Other papers deal with the separation of cefradine from other cephalosporins [8–18]. Some papers discuss the determination of cefradine in pharmaceuticals [1, 7, 8, 17, 19].

None of these LC methods described in literature has been validated for the separation of cefradine from its related substances. Comparison of different reversed-phases demonstrated the poor reproducibility of the selectivity of C_8 and C_{18} stationary phases towards the cephalosporins [16].

In this paper, a LC method is described for both qualitative and quantitative analysis of cefradine, using poly(styrene-divinylbenzene) (PSDVB) as stationary phase. This method is able to separate cefradine from its related substances and gives reproducible results on different brands of PSDVB available on the market. Using this method, the United States Pharmacopeia Reference Standard, the British Pharmacopoeia Chemical Reference Substance and the European Pharmacopoeia Chemical Reference Substance were compared. Results for a number of bulk samples and pharmaceuticals are also reported.

Experimental

Reference substances and samples

The United States Pharmacopeia Reference Standard (USP-RS; Lot H; 906 μg mg^{-1}), the European Pharmacopoeia Chemical Reference Standard (Ph. Eur.-CRS; 93.5%) and the British Pharmacopoeia Chemical Reference Standard (BP-CRS; Batch No. 1385; 96.0%) of cefradine were available, as well as the Ph. Eur.-CRS of cefalexin (93.4%).

For a number of bulk samples the origin was unknown. Other bulk samples were kindly donated by Gema (Barcelona, Spain) and Gist-Brocades (Delft, The Netherlands). Specialities (Velosef®, Squibb) from the Belgium market were provided by the Ministry of Health.

Related substances

Cefradine is converted into cefalexin by influence of UV light. Cefalexin is the most

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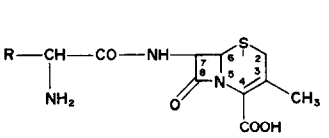
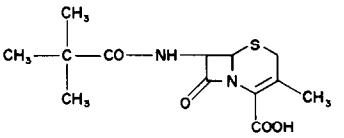
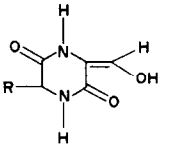
important related substance, always present in cefradine samples. Other impurities originate from the semi-synthetic preparation and from degradation. Figure 1 shows the structures of cefradine and its potential impurities. **VII** and **VIII**, which are the basic constituents of the cefradine molecule, are commercially available (**VII**: Gist-Brocades; **VIII**: Janssen Chimica, Beerse, Belgium). **II**, **VI** and **IX** can arise from the semi-synthetic preparation of cefradine. **VI** and **IX** were prepared in the laboratory, **II** was provided by a manufacturer. The other related substances are decomposition products formed in acidic (**III**, **IV**), neutral (**III**, **V**) and alkaline



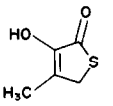
I : D-cefradine
a : D-cefalexin



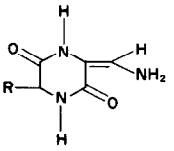
II (=b) : pivalamide of 7-ADCA



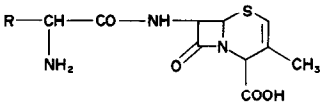
III : 3-hydroxymethylene-6-(cyclohexa-1,4-dienyl)-piperazine-2,5-dione
c : 3-hydroxymethylene-6-phenyl-piperazine-2,5-dione



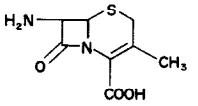
IV (=d) : 3-hydroxy-4-methyl-2(5H)-thiophenone



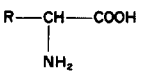
V : 3-aminomethylene-6-(cyclohexa-1,4-dienyl)-piperazine-2,5-dione
e : 3-aminomethylene-6-phenyl-piperazine-2,5-dione



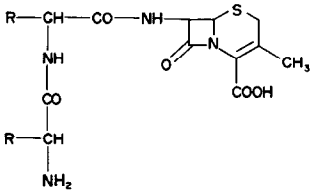
VI : Δ^2 - cefradine
f : Δ^2 - cefalexin



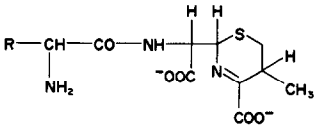
VII (=g) : 7-aminodesacetoxycephalosporanic acid (7-ADCA)



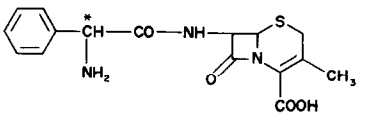
VIII : D-cyclohexa-1,4-dienylglycine
h : D-phenylglycine



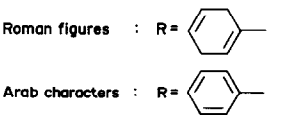
IX : cyclohexa-1,4-dienylglycylcefradine
i : phenylglycylcefalexin



X : cefradine Δ^4 - cephalosporates
j : cefalexin Δ^4 - cephalosporates



k : L-cefalexin



Roman figures : R =

Arab characters : R =

Figure 1
Structures of cefradine (I) and its related substances (Roman figures) and of cefalexin (a) and its related substances (Arab characters).

(**X**) media. **III**, **IV**, **V** and **X** were prepared in the laboratory. Since cefalexin is always present in cefradine, its corresponding related substances were also considered. Cefradine and cefalexin both have the D-configuration in the side chain. The L-configuration has no significant biological activity. Since L-phenylglycine is commercially available (Janssen Chimica), it was possible to prepare L-cefalexin (**k**) in the laboratory. Unfortunately, L- or DL-cyclohexa-1,4-dienylglycine were not available. The preparation of the related substances will be described elsewhere. **X** and **j** were never isolated but were prepared *in situ* by dissolving cefradine or cefalexin in 0.1 N NaOH (1 mg ml^{-1}) and storing the solution at room temperature for 10 min.

Chemicals and mobile phase

Acetonitrile 99% (Janssen Chimica) and methanol (Roland, Brussels, Belgium) were distilled before use. 2-Methyl-2-propanol 99.5% (Janssen Chimica) was used as such. Phosphoric acid 85% and potassium dihydrogen phosphate Zur Analyse were from Merck (Darmstadt, Germany). Sodium 1-octanesulphonate (NaOS) was from Janssen Chimica. Water was distilled twice.

The mobile phase finally used was prepared by mixing 500 ml of water, 145 ml of acetonitrile, 100 ml of 0.02 M sodium 1-octanesulphonate and 50 ml of 0.2 M phosphoric acid. The mixture was diluted to 1000 ml with water and degassed before use.

LC apparatus and operating conditions

Isocratic elution was used throughout the study. The equipment consisted of a L-6200 pump (Merck–Hitachi, Darmstadt, Germany), a 20 μl loop injector Model CV-6-UHPa-N60 (Valco, Houston, TX, USA), a 250 × 4.6 mm i.d. column, packed in the laboratory with PLRP-S 100 Å 8 μm (Polymer Laboratories, Church Stretton, Shropshire, UK), a 254 nm fixed wavelength UV monitor D (LDC/Milton Roy, Riviera Beach, FL, USA) and an integrator Model 3396 A (Hewlett–Packard, Avondale, PA, USA). The column temperature was maintained at 50°C by means of a water bath heated by a Julabo EM thermostat (Julabo, Seelbach, Germany). The selectivity of the method was also examined using other PLRP-S columns or other PSDVB stationary phases (PRP-1 10 μm and PRP-1 7–9 μm, Hamilton, Reno, NV, USA). For the examination of peak homogeneity the UV detector was replaced by a photodiode array detector Model 990 (Waters Assoc., Milford, MA, USA). A Marathon autosampler (Spark Holland, Emmen, The Netherlands) with sample-cooling (6°C) equipped with a fixed 20 μl loop and a cryomat Julabo C and F10 was used for the quantitative determinations.

Sample preparation

Samples for quantitative analysis were prepared by weighing 30 mg of cefradine into a 20 ml volumetric flask. Mobile phase containing 20% of the 0.02 M solution of sodium 1-octanesulphonate was used as solvent.

For the specialties an amount corresponding to 30 mg of cefradine was weighed into a 20 ml volumetric flask, 15 ml of solvent was added and the mixture was ultrasonicated at room temperature for 5 min. Solvent was then added up to volume.

The chemical reference substances of cefradine of different origin were dissolved the same way as the samples.

Results and Discussion

Development of the chromatographic method

Poly(styrene-divinylbenzene) (PSDVB) is a very stable material, even at extreme pH conditions (pH 1–13) and high temperature [20]. Recent work on LC of cefalexin proved that this stationary phase gave very satisfactory results for the separation of cefalexin from its related substances [21]. The mobile phase consisted of acetonitrile, 0.02 M sodium 1-octanesulphonate (NaOS), 0.2 M phosphoric acid and water. Using a very similar mobile phase, a mixture of cefradine and its related substances was analysed. The PSDVB stationary phase was heated at 60°C to enhance the mass-transfer and to reduce the backpressure. A chromatogram is shown in Fig. 2. Related substance **f**, which is Δ^2-cefalexin, was also present in the mixture, because Δ^2-cefradine was prepared starting from cefradine, which always contains a certain amount of cefalexin. Related substance **VIII**, which was eluted close to the dead volume, was not detected at 254 nm. The complex mixture of polar diastereoisomeric substances **X** and **j** was analysed separately. All these substances were eluted fast (capacity factors of 0–3) and were well separated from cefalexin and cefradine. Related substance **IX** was not added to the

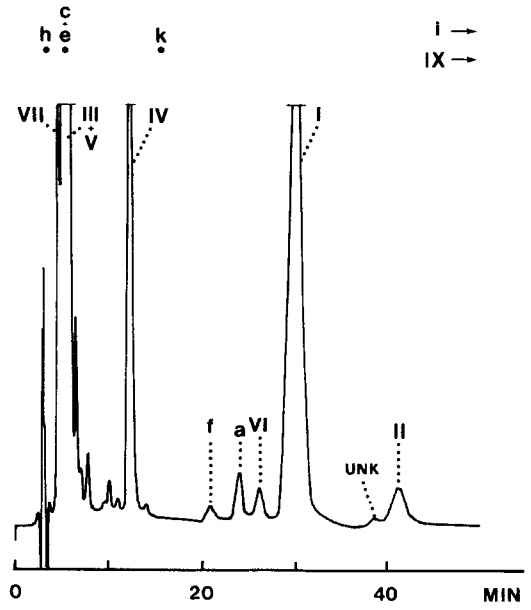


Figure 2
Chromatogram of cefradine spiked with its related substances (symbols: see Fig. 1). UNK = unknown. Stationary phase: PLRP-S 100 Å 8 μm. Mobile phase: CH_3CN–NaOS (0.02 M)–H_3PO_4 (0.2 M)–H_2O (14.5:10:5:up to 100, v/v/v/v). The position of the related substances of cefalexin, which were not added to the mixture, is indicated at the top of the figure.

mixture, because of its long retention time. Using a mobile phase containing 25% of acetonitrile instead of 14.5%, **IX** was eluted at 13.5 min, whereas cefradine was eluted at 5.6 min. Related substance **i** was strongly retained, like its analogue **IX**. The small peak (**UNK**) between cefradine and **II** was found to be present in all cefradine samples but did not correspond to any of the structures shown in Fig. 1. At the top of the figure the position of the related substances of cefalexin, which were not added to the mixture, is indicated. The main components **I** and **a**, which are both active antibiotics, were well separated from all the potential impurities.

The mobile phase was further evaluated by systematic examination of its components. Substances **IX**, **c**, **e**, **h**, **i** and **k** were no longer considered in these experiments because they were very well separated from the main components **I** and **a**. The 0.2 M phosphoric acid solution (pH 1.4) of the mobile phase was replaced by 0.2 M phosphate buffers pH 3.0 or 4.0, in order to examine the influence of the pH. For each pH, the amount of organic modifier was varied to optimize the separation (Fig. 3). The substances examined were cefradine and the related substances cefalexin, **II**, **VI** and **f**. This allowed the study of the most critical separation problems. At pH 1.4 a very good separation of cefradine from its related substances was achieved using 14–15% of acetonitrile. pH 3.0 and pH 4.0 were found to be less suitable because of the lower efficiency which was probably related to the poorer protonation of the molecules. In conclusion, phosphoric acid was chosen to regulate the pH of the mobile phase.

Other organic modifiers were also examined. 2-Methyl-2-propanol (Fig. 4) proved to be very suitable in LC of tetracyclines [22] and erythromycin [23] on PSDVB. The order of elution was changed in comparison with acetonitrile and good separation was not achieved. The Δ^2-isomers **VI** and **f** were now eluted on the tail of their respective Δ^3-isomers cefradine and cefalexin. Methanol gave very poor efficiency and separation. Acetonitrile was therefore preferred as organic modifier.

Figure 5 shows the influence of different concentrations of the ion-pairing reagent sodium 1-octanesulphonate (NaOS). Doubling the original concentration in the mobile phase to 20% of the 0.02 M solution caused an increase of the retention times, except for **II**, which was now eluted in the ascending part of the cefradine peak. Substance **II** had no positively charged amino group like the other related substances and consequently did not interact with the ion-pairing reagent. Lowering the amount of NaOS to half the original concentration decreased the separation of cefalexin and **VI**. The concentration of NaOS had also to be maintained at a certain level, to provide sufficient NaOS ions for ion pairing with cefradine and avoid formation of double peaks. In the final mobile phase, 10% of a 0.02 M NaOS solution was used. By reducing the amount of 0.2 M phosphoric acid from 5% to 2.5%, **VI** was eluted closer to cefalexin, resulting in a poor separation. At 7.5%, the retention times were nearly the same as for 5%. The initial concentration (5% of a 0.2 M solution) was therefore maintained. The mobile phase reported in Fig. 2 was therefore considered as suitable for further use.

The selectivity of different PSDVB-columns was also examined. Table 1 shows an overview of the columns used, with the respective concentrations of acetonitrile in the mobile phase, which was adapted for each column to

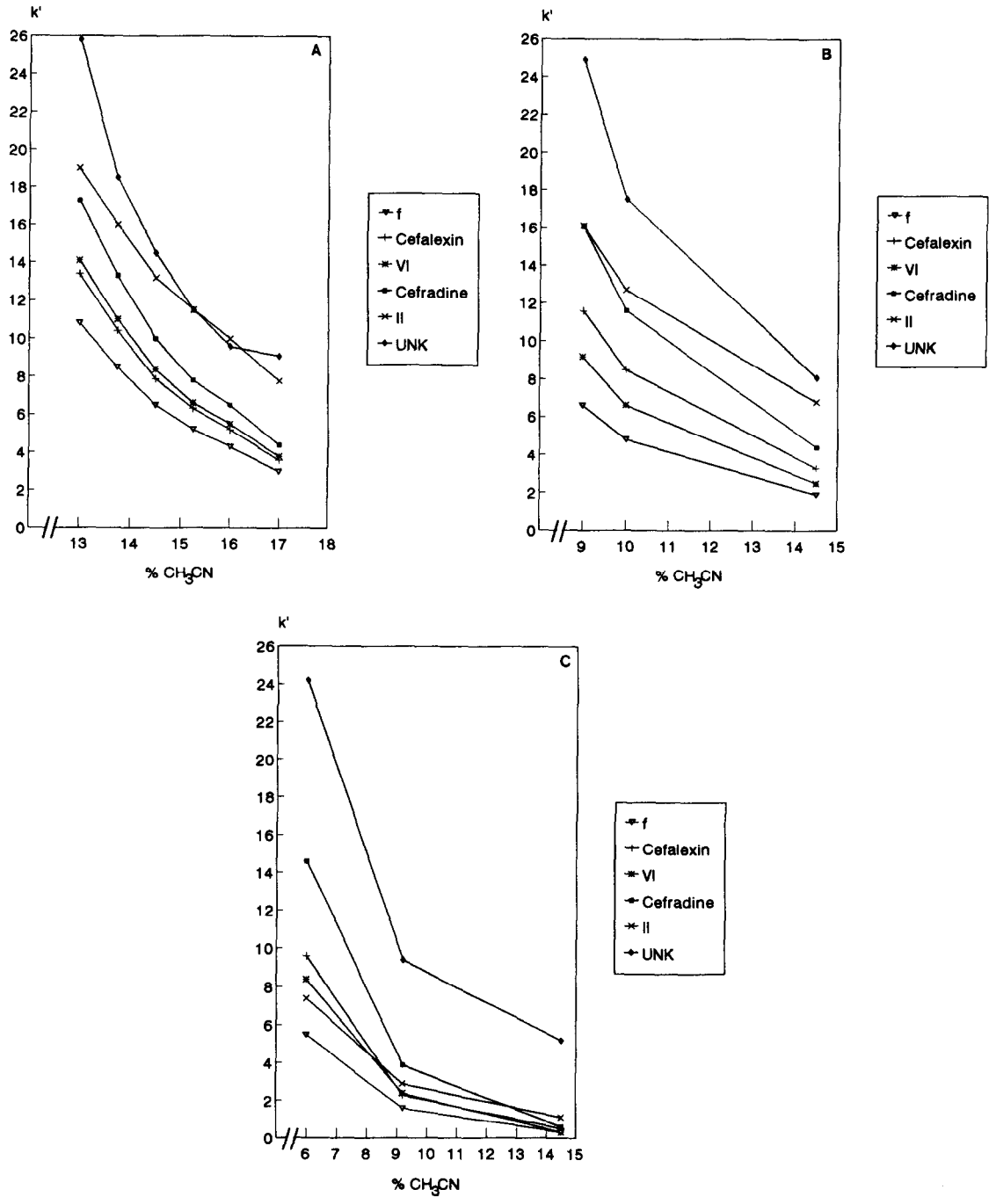


Figure 3
Influence of the concentration of acetonitrile on the separation of cefradine and related substances at (A) pH 1.4, (B) pH 3.0 and (C) pH 4.0 (symbols: see Fig. 1). Stationary phase: PLRP-S 100 Å 8 μm. Mobile phase: CH_3CN–NaOS (0.02 M)–H_3PO_4 (0.2 M) or phosphate buffer pH 3.0 or 4.0 (0.2 M)–H_2O (*x*:10:5:up to 100, v/v/v/v).

give a retention time of about 30 min for cefradine. As can be seen in Fig. 6, the elution order is the same for each column, except for **II** and **UNK**. Good reproducibility of the selectivity is a great advantage of PSDVB stationary phases.

The selective separation of cefradine from a sample stored in alkaline or acid medium was examined by means of photodiode array detection. A solution of cefradine in 0.1 N NaOH (1 mg ml^{-1}) was stored at room temperature for 10 min, neutralized and analysed. The

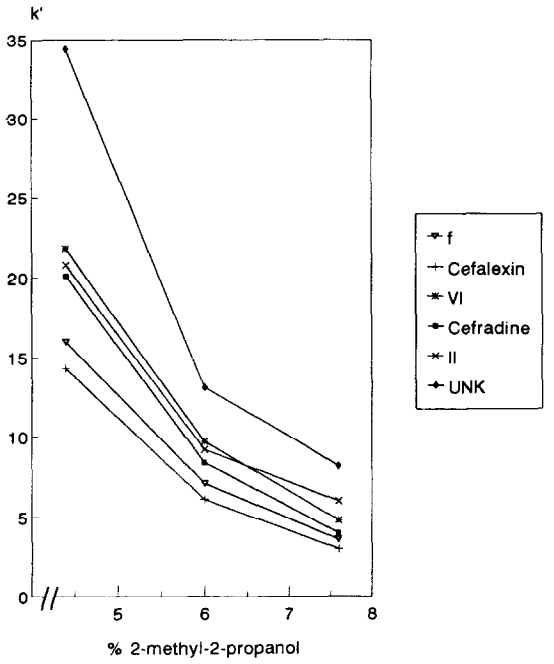


Figure 4
Influence of the concentration of 2-methyl-2-propanol as organic modifier on the separation of cefradine and related substances (symbols: see Fig. 1). Stationary phase: PLRP-S 100 Å 8 μm. Mobile phase: 2-methyl-2-propanol–NaOS (0.02 M)–H_3PO_4 (0.2 M)–H_2O (*x*:10:5:up to 100, m/v/v/v).

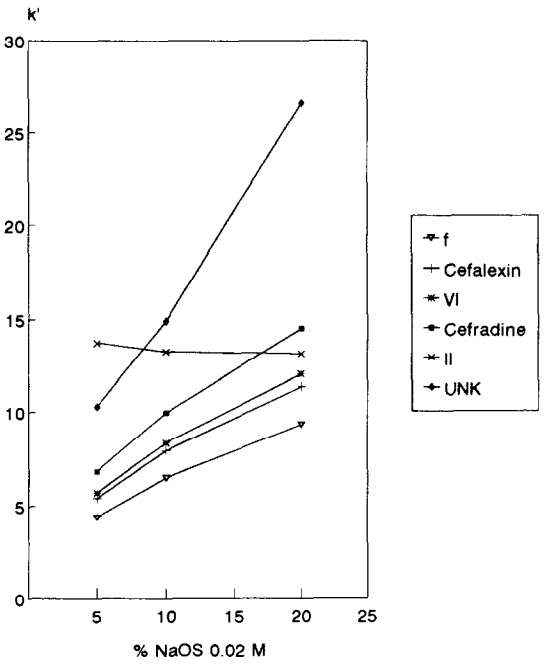


Figure 5
Influence of the concentration of sodium 1-octanesulphonate (NaOS) in the mobile phase on the separation of cefradine and related substances (symbols: see Fig. 1). Stationary phase: PLRP-S 100 Å 8 μm. Mobile phase: CH_3CN–NaOS (0.02 M)–H_3PO_4 (0.2 M)–H_2O (14.5:*x*:5:up to 100, v/v/v/v).

Table 1
Characteristics of the poly(styrene-divinylbenzene) columns used, with the respective concentrations of acetonitrile in the mobile phase

Column	Age (years)	Stationary phase (250 × 4.6 mm i.d.)	Batch	% CH_3CN
A	5	PLRP-S, 100 Å 8 μM	(10-12-85)B	15.25
B	5	PLRP-S, 100 Å 8 μM	(10-12-85)B	14.95
C	2	PLRP-S, 100 Å 8 μM	35	14.50
D	1	PLRP-S, 100 Å 8 μM	35	14.95
E	1	PLRP-S, 100 Å 8 μM	35	15.40
F	new	PLRP-S, 100 Å 8 μM	8M-RPS-1-64	15.25
G	6	PRP-1, 10 μm	79400	14.80
H	new	PRP-1, 7–9 μm	457	15.70

cefradine peak was found to be homogeneous, notwithstanding the high level of degradation. A solution of cefradine in 0.1 N HCl (1 mg ml^{-1}) stored at 60°C for 5 h was examined using the same method. The cefradine peak remained homogeneous.

The influence of the column temperature on the separation was investigated at 50, 60 and 70°C. The selectivity at the three temperatures was comparable. The stability of a solution of cefradine in mobile phase stored at these three temperatures was examined. After 45 min storage at 60 or 70°C a decrease of, respectively, 1.3 or 6.3% of cefradine was observed, while at 50°C no decomposition occurred. In order to guarantee stability of the samples during analysis, the column temperature was decreased to 50°C.

A solution of cefradine in water gave several disturbing system peaks. Using mobile phase as the solvent, some system peaks disappeared. After systematic variation of the composition of this solvent, mobile phase containing 20% instead of 10% 0.02 M NaOS was chosen as the solvent for the samples. Unfortunately, one small system peak was still present.

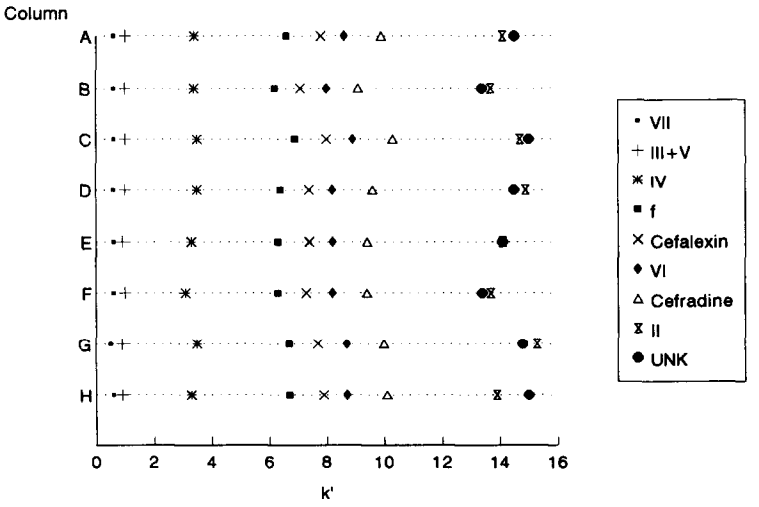


Figure 6
Elution order of cefradine and its related substances (symbols: see Fig. 1) on different PSDVB columns (see Table 1 for characteristics). Mobile phase: CH_3CN–NaOS (0.02 M)–H_3PO_4 (0.2 M)–H_2O (*x*:10:5:up to 100, v/v/v/v), *x*: see Table 1.

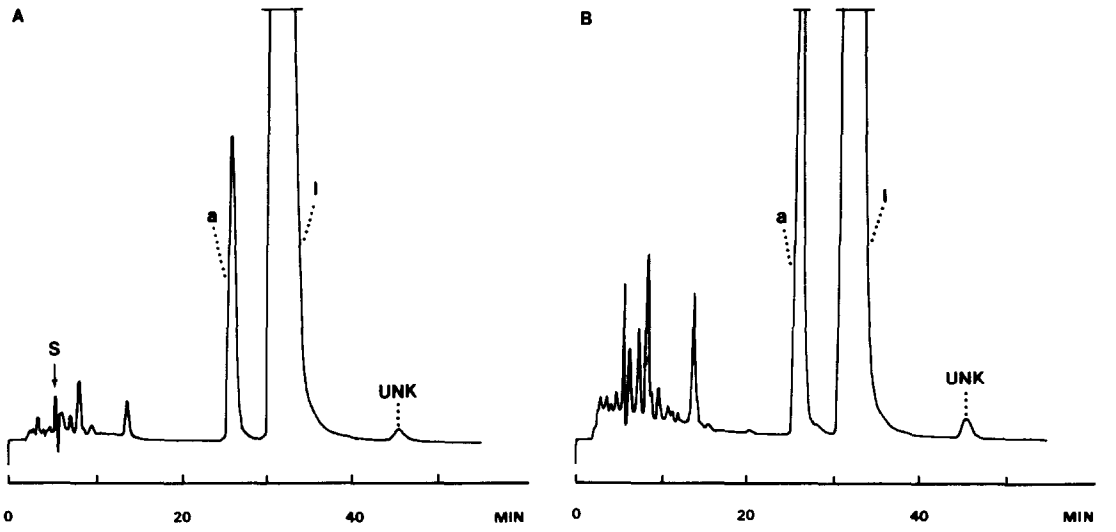


Figure 7
Typical chromatogram of (A) a recent and (B) an older sample of cefradine (30 μg). Stationary phase: PLRP-S 100 Å 8 μm. Mobile phase: CH_3CN–NaOS (0.02 M)–H_3PO_4 (0.2 M)–H_2O (14.5:10:5:up to 100, v/v/v/v). Column temperature: 50°C. S = system peak.

Figure 7 shows a typical chromatogram of a recent and an older cefradine sample obtained with the selected chromatographic conditions. The content of decomposition products of the older sample is obviously higher, as well as the cefalexin content.

At last, a resolution test was developed for the LC method. Using mobile phases containing an increasing amount of organic modifier, a mixture of cefradine, cefalexin and the closest eluting related substances **f**, **VI** and **II** was analysed for evaluation of the separation of the main components cefradine and cefalexin. In the same conditions, a mixture of equal amounts of cefalexin and cefradine was analysed for the calculation of the resolution. These analyses were performed on each of the columns listed in Table 1. It was observed that when a resolution of 4.0 was obtained between cefalexin and cefradine, sufficient separation was obtained also between cefalexin, cefradine and the most closely eluted related substances.

Quantitative aspects of the LC method

The loadability of the column was found to be 50 μg of cefradine. By injection of a higher amount, peak splitting of the small cefalexin peak occurred. It was decided to use a 30 μg amount for quantitative analysis of cefradine samples. For this quantity the limit of detection (LOD), expressed as cefradine, was 0.05%. The limit of quantitation (LOQ) was 0.1% [n = 7, relative standard deviation (RSD) = 6.5%]. The repeatability was checked by analysing the same solution of cefradine six times (RSD = 0.16%) and by analysing subsequently six freshly prepared solutions of cefradine (RSD = 0.35%). At about 6°C solutions of cefradine in the solvent remained stable for at least 24 h. Linearity tests were performed (y = peak area/1000, x = amount injected in μg): $y = 10609x - 1315$, $S_{y,x}$ = 409, r = 1.000 (n = 9), range of x covered in the experiments = 24–36 μg.

Comparison of cefradine standards

This LC method was used to compare the USP-RS, the BP-CRS and the Ph. Eur.-CRS. The cefradine content of the USP-CRS and the BP-CRS was calculated by comparison with the Ph. Eur.-CRS, which has an assigned cefradine content of 93.5% on 'as is' [24]. Samples (30 μg) of each standard were analysed. The cefalexin content of the USP-RS and the BP-CRS was obtained by comparison with 1.2 μg injections of the Ph. Eur.-CRS of cefalexin (93.4% cefalexin on 'as is' [25]), which was also used to calculate the content of **UNK** and other impurities (0.1 μg injections). Results obtained for the standards are shown in Table 2. The BP-CRS and the USP-RS contain comparable amounts of cefradine, the BP-CRS contains more cefalexin and 'other impurities'. Due to the limited amount available of these standards, the content of water and solvents was not determined.

The declared content of the BP-CRS seems to be overestimated. The BP-CRS, however, is applied in an iodimetric assay, which does not discriminate between the different cephalosporin structures containing an intact β-lactam ring. In this case, cefalexin reacts in the same way as cefradine and the content can only be 90.6% at most. The overestimation of the BP-CRS may be explained by the hygroscopic character of cefradine, which might have caused water uptake in the period between the official determination of the content and the final packing in airtight containers.

For the USP-RS, 99.4% of the mass can be explained by addition of the content of cefradine, cefalexin, **UNK**, other impurities and water. The cefradine content found is obviously lower than the declared content (906 μg mg^{-1}), which is closer to the sum of cefradine and cefalexin (89.2%). The USP-RS is indeed applied in a microbiological assay, which cannot distinguish between cefalexin and cefradine. It needs to be emphasized that the content expressed as μg mg^{-1} should be interpreted as micrograms of activity compared

Table 2
Composition of cefradine standards

	Ph. Eur.-CRS 93.5%	BP-CRS 96.0%	USP-RS 906 μg mg^{-1}
Number of analyses	26	30	30
Number of solutions analysed	9	6	6
Number of days	3	3	3
Cefradine	93.5*	87.23	87.12
	(0.7)	(1.2)	(0.7)
Cefalexin	2.50*	3.38	2.13
		(11)	(2)
Unknown (**UNK**)		0.30	0.30
	0.28*	(6)	(5)
Other impurities		1.97	0.15
		(20)	(34)
Solvents	0.1*	ND	ND
Water determined	3.39	ND	ND
Water declared	3.66*	NA	9.67†
Total	100.0	—	99.4

Values in per cent (m/m), RSD is given in parentheses, ND = not determined due to limited amount of sample, NA = not available.

* Assigned content following ref. 24.

† W.W. Wright, United States Pharmacopeia, Rockville, USA, personal communication.

Table 3
Composition of bulk samples of cefradine

O	*N*	LC cefradine ($n = 8$) *A*	LC cefalexin ($n = 8$) *B*	LC **UNK** ($n = 4$) *C*	LC other impurities ($n = 4$) *D*	Water ($n = 3$) *E*	Total $A + B + C + D + E$	Non aqueous titration ($n = 3$) *F*	Total $E + F$
a	1	89.24 (0.6)	3.06 (1.5)	0.31 (1.5)	0.65 (6.8)	4.40 (3.1)	97.66		
a	2	89.44 (0.9)	2.77 (1.8)	0.33 (3.1)	0.40 (7.1)	5.22 (5.5)	98.16		
a	3	87.80 (0.7)	2.82 (1.2)	0.32 (0.9)	1.16 (1.6)	4.09 (4.7)	96.19		
a	4	91.26 (0.6)	3.11 (2.9)	0.28 (2.3)	0.36 (18)	3.53 (1.4)	98.54		
b	5	92.79 (0.6)	2.80 (1.3)	0.24 (4.8)	0.17 (7.4)	3.54 (2.0)	99.54	93.58 (0.3)	97.12
c	6	89.12 (0.8)	3.03 (1.5)	0.37 (1.7)	1.36 (3.8)	4.79 (2.1)	98.67	94.38 (0.2)	99.17
c	7	74.24 (1.1)	7.32 (5.3)	0.45 (2.3)	5.67 (3.6)	5.39 (5.1)	93.07	91.54 (0.1)	96.93
c	8	49.95 (0.8)	12.36 (1.7)	0.32 (3.1)	15.12 (6.0)	5.92 (0.7)	83.67	86.53 (0.2)	89.59

Values in per cent (m/m); RSD (%) are given in parentheses. Water was determined by Karl Fischer titration.
O = sample origin, *N* = sample number, **UNK** = unknown.

to a primary standard and not as micrograms of mass.

Analysis of commercial samples

Table 3 shows the results obtained for a number of bulk samples. The water content of the samples was determined by Karl Fischer titration. The RSD on the cefradine content mostly did not exceed 1.0%. All the samples except 7 and 8 complied with the Ph. Eur. requirements for content, which are 90.0% for cefradine and 95.0–102.0% for the sum of cefalexin and cefradine, both figures calculated on anhydrous. Samples 7 and 8 also did not comply with the limit for the cefalexin content, which is not more than 5.0% on anhydrous. All the samples complied with the requirement for water (not more than 6.0%). For the less pure samples, the total mass could not be explained entirely by addition of the content of cefradine, cefalexin, **UNK**, other impurities and water. This may be due to the fact that some of the degradation products have a lower specific absorbance at 254 nm than cefalexin. The base content of a few samples, of which enough was available, was determined by non aqueous titration to check the correspondence with LC figures. Cefradine (200 mg) was dissolved in 40 ml of acetic acid and titrated with $HClO_4$ 0.1 N using potentiometric endpoint detection. Each sample was titrated three times. These results are also shown in Table 3. However, the total of the non aqueous titration and water (E and F) was higher than the sum of the LC determinations and water (A, B, C, D and E), except for the purer sample 5. When impurities, carrying no basic function, are present, titration figures do not allow a correct estimation of the total non volatile mass.

Table 4 shows the results obtained for three commercial samples: capsules; a powder for suspension; and a powder for injection, all from the same manufacturer. The content of cefradine was expressed as a percentage (m/m) of the label claim. The content of the sample for injection was very high, which is probably due to overdosage, since the homogeneity of the cefradine peak was demonstrated by photodiode array detection.

Table 4
Cefradine content of specialties as a percentage (m/m) of label claim

Form	Mean content (n = 4)	RSD (%)
Capsules	98.5	1.2
Suspension	97.1	0.2
Powder for injection	131.5	0.9

It can be concluded that the method described is suitable for purity control and assay of bulk samples and different kinds of pharmaceutical preparations containing cefradine. Major advantages of the method are its selectivity and its applicability on PSDVB stationary phases from different manufacturers and of different age.

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