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Optimisation, validation and application of a capillary electrophoresis method for the determination of ranitidine hydrochloride and related substances

M.A. Kelly[a], K.D. Altria[b], C. Grace[b], B.J. Clark[a,*]

[a]*Pharmaceutical Chemistry, School of Pharmacy, University of Bradford, Bradford, West Yorkshire BD7 1DP, UK*
[b]*GlaxoWellcome Research and Development, Ware, Herts. SG12 0DP, UK*

Abstract

Ranitidine hydrochloride is an H_2-antagonist which is widely prescribed for the treatment of peptic ulcers. The drug is marketed in a variety of dosage forms including tablets, syrups and injection solutions. A range of synthetic and degradative impurities of ranitidine are known and currently, these impurities are routinely determined using thin-layer chromatography (TLC). Alternatively a high-performance liquid chromatography (HPLC) method has also been employed in the assay of the pharmaceutical preparation. Unlike TLC, capillary electrophoresis (CE) offers the capability to quantify simultaneously both the active drug content and the levels of the related substances. The advantages of simplicity, selectivity, versatility and ease of use of CE offers a complementary separation technique to the established methods of HPLC and TLC in the determination of ranitidine and its related substances. This work represents a comprehensive evaluation of the performance of a developed CE method in the determination of drug-related impurities in both drug substance and various pharmaceutical formulations. The data obtained clearly shows that the performance of an optimised CE method can be equivalent in terms of sensitivity and precision to that of a HPLC method employed for a similar purpose and offers better selectivity against TLC and HPLC.

Keywords: Ranitidine hydrochloride; Validation; Pharmaceutical analysis

1. Introduction

Ranitidine hydrochloride is a H_2-antagonist which is widely prescribed for the treatment of peptic ulcers [1], reflux oesophagitis [2] and dyspepsia [3]. The drug is marketed in a range of dosage forms including tablets, syrups and injection solutions and a number of synthetic and degradative related substances of ranitidine are known [4]. The structures of ranitidine and seven of the major related substances are shown in Table 1. Currently, these substances are routinely determined using thin-layer chromatography (TLC) [5], achieving limits of detection (LODs) of 0.05% (a/a) (% area of the main peak), within an analysis time of approximately 30 min. But with the disadvantage that the TLC procedure is a two-stage analytical process. In addition HPLC has been used to separate ranitidine from its related substances [4].

Alternatively capillary electrophoresis (CE) offers a highly efficient separation technique which may be capable of simultaneously quantifying both the ranitidine and the levels of the related substances in bulk and pharmaceutical preparations. It is proposed

*Corresponding author.

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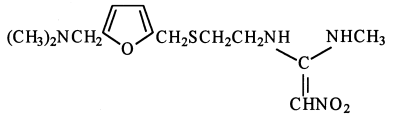
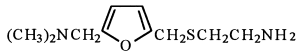
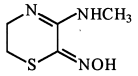
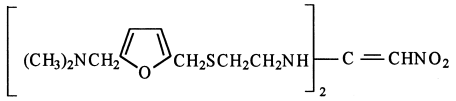
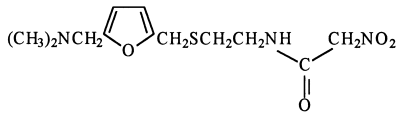
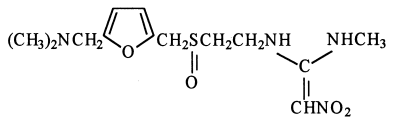
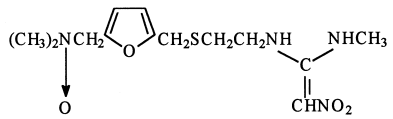
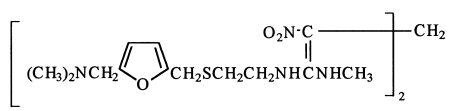
Table 1
Chemical structures of ranitidine and related substances

Compound number	Structure	Potential source (S-synthesis D-by degradation).
		-
	Ranitidine.	
1		S,D
	5-[[(2-aminoethyl)thio]methyl]-*N*,*N*-dimethyl-2-furanmethanamine.	
2		S,D
	5,6-dihydro-3-methylamino-2H-1,4-thiazin-2-one oxime	
3		S
	N,*N*'-bis[2-[[[5-[(dimethylamino)methyl]-2-furanyl]methyl]thio]ethyl]-2-nitro-1,1-ethenediamine.	
4		D
	N-[2-[[[5-[(dimethylamino)methyl]-2-furanyl]methyl]thio]ethyl]-2-nitroacetaminde.	
5		D
	N-[2-[[[5-[(dimethylamino)methyl]-2-furanyl]methyl]sulphinyl]ethyl]-*N*'-methyl-2-nitro-1,1-ethenediamne.	
6		D
	N-[2-[[[5-[(dimethylamino)methyl]-2-furanyl]thio]ethyl]-*N*'-methyl-2-nitro-2,2-ethenediamine N-oxide.	
7		D
	Formaldehyde adduct	

that the advantages of CE in simplicity, selectivity, versatility and ease of use should offer a complementary separation technique to the established TLC and HPLC procedures for this complex separation problem.

In examination of the safety and quality of a drug, changes in the profile of drug-related substances can occur in several ways, from the route of synthesis and the cleanup at different stages, to the quality of the reagents and the conditions used for the reactions. Modification may also occur during storage where the presence of light, moisture or oxygen can alter the identities and quantities of the related substance levels. To test these aspects and to ensure the safety of a drug, it is necessary to set up efficient methods and define certain specifications in the analytical procedure. In the pharmaceutical industry these controls are adhered to using a strict protocol of validation and are essential to satisfy the regulatory agencies.

When developing methods by CE for pharmaceutical products the method validation performance criteria has generally been taken as being similar to those applied to HPLC assays [6], however within certain limitations [7].

A number of reports have discussed aspects of validation for specific CE methods, such as in the determination of potassium counter-ion levels in pharmaceuticals by CE [8], where good agreement was achieved with an ion exchange chromatographic method. Also the determination of quinolone and its related impurities using a low pH buffer and ''pH mediated sample stacking'' for improved method sensitivity [9]. Other examples include statistical comparisons giving close agreement between HPLC and CE assays for cephradine [10], domperidone [11] and salbutamol and their related substances [12]. Quality control examples have also been reported where it has been possible to replace HPLC as the method of choice [13]. Procedures which include ion-pair reagents and cyclodextrin have been successfully validated for remoxipride and its related substances [14], where detection of all the required substances was at the 0.1% m/m level.

For validation experiments the extent of the tests will vary according to the nature of the application and the intended usage. A full method validation normally involves demonstrating acceptable selectivity and accuracy (through, linearity of response and determination of total error and by recovery experiments). In addition precision under conditions of repeatability is obtained from identical test material with the same conditions (of same sample solution, analyst, and apparatus in the same laboratory), over a relatively short time interval. In contrast, reproducibility involves assessment of the variability with the same method on identical test material but under different conditions (i.e. different sample solutions, analysts, apparatus and laboratory), over a relatively prolonged period of time.

In this work a comprehensive evaluation of the validation and performance of a developed CE method for the determination of the important drug, ranitidine and all its related substances is described, where the assay is applied to the drug substance and commercial pharmaceutical formulations.

2. Experimental

2.1. Instrumentation

A Beckman P/ACE 5100 or 2050 instrument (Fullerton, CA, USA) connected to a Hewlett-Packard (Bracknell, UK) HP1000 data system was used. Fused silica capillaries were obtained from Composite Metal Services (Hallow, UK) 27 cm (20 cm to window)×50 μm I.D.. Capillary detection windows were produced using an electrical filament device purchased from Electrokinetic Technologies (Capital HPLC, Edinburgh, UK). Inorganic reagents (trisodium citrate dihydrate and citric acid monohydrate) were obtained from Aldrich (Poole, UK). (+)-Chlorpheniramine (internal standard) was obtained from Sigma (Poole, UK).

2.2. Method conditions

The optimised method conditions are given in Table 2. As buffer pH was considered to be very important, an accurately weighed amount of trisodium citrate and citric acid (Table 2) was added to a 100 ml volumetric flask to obtain a final concentration equivalent to 190 m*M* trisodium citrate at a pH of 2.6. Under these operating conditions a current of 90 μA was typically generated. (A maxi-

Table 2
Details of the CE method parameters

Step	Beckman P/ACE 5100
Capillary dimensions	27 cm (20 cm to the detector)×50 μm
Detection aperture	800×100 μm
Rinse 1	1.0 min (0.1 *M* NaOH)
Rinse 2	1.0 min (buffer)
Injection	1 s
Buffer (mass of trisodium citrate)	5.52±0.06 g and
(mass of citric acid)	21.09±0.1 g/100 ml water
Voltage	+6 kV
Temperature	25°C
Detection wavelength	230 nm
Typical current	90 μA

mum of 100 μA is taken as the allowable limit both in our laboratories and in GlaxoWellcome. It is suggested as being good working practice to include an indication of the expected level of current in a method as a system suitability type check).

2.3. Sample preparation

The method development and optimisation were performed using an aqueous mixture of ranitidine at 10 mg/ml and the seven potential related substances at 0.01 mg/ml (i.e. 0.1%, m/m, of nominal ranitidine content). The related substances are either potential synthetic or degradative products of ranitidine. The chemical structures of these compounds are shown in Table 1. Ranitidine hydrochloride samples and related substances were obtained from within GlaxoWellcome Research and Development. The Zantac injection solutions (containing 10 or 25 mg/ml, with or without phenol) were diluted with internal standard solution [(+)-chlorpheniramine, 3 mg/ml, dissolved in water] to contain approximately 5 and 10 mg/ml ranitidine hydrochloride in water. The bulk drug substance was analysed by weighing 100 mg of ranitidine hydrochloride drug into 10 ml of internal standard solution.

3. Results and discussion

3.1. Method optimisation

The main prerequisite of the CE method for the determination of ranitidine related substances was that it should offer at least equivalent performance to that by established TLC and HPLC methods. These requirements were:

1. Separation of ranitidine and potential related substances of interest.
2. Detection limits of 0.1% (a/a) or less.
3. Total analysis time of 30 min or less.

A preliminary investigation for the determination of ranitidine suggested the use of a low pH borate electrolyte [15]. However, when challenged with a mixture of potential related substances, this method was found to be incapable of achieving the stipulated resolution and a further study was necessary to attain the requirements listed above.

3.1.1. Choice of electrolyte ionic strength and pH

From the chemical information it was anticipated that ranitidine and its related substances could be separated using low ionic-strength buffer system. Several buffer salts were investigated including sodium phosphate, sodium acetate, Tris and sodium citrate. As the UV-detection wavelength was to be 230 nm, low UV absorbing buffers with good buffering capacity were required and electrolyte concentrations between 20 and 200 m*M* were investigated. Using these conditions the resolution of the most critical peak pair, was that of the formaldehyde adduct (structure 7) from ranitidine and although, resolution of the seven related substances was obtained, the partial separation of this pair prevented accurate determination of the degradation product at trace levels.

With sodium citrate as electrolyte the first separation of the formaldehyde adduct from ranitidine

was provided, whilst still maintaining resolution of all the other substances. To further improve the resolution, ionic strength and pH of the electrolyte was further varied. From this it was found that an exact combination of ionic strength for both citric acid and citrate was essential to achieve the desired separation. As a result, the best conditions for the ranitidine sample spiked with the seven related substances (at 0.1%, a/a) was 190 m*M* trisodium citrate, adjusted to pH 2.6 with 2 *M* citric acid and run at an applied voltage of 6 kV. However to achieve the optimum separation of all components the pH was closely controlled by accurately weighing the buffer salt and the adjusting acid to obtain the final concentration as above.

Thus the accuracy of preparation of the electrolyte highlights the need on some occasions to be very specific in preparing and reporting details in CE, as slight inadvertent modifications in preparation can lead to nonrepeatable methods. Therefore it is considered essential to detail fully the exact form (i.e. salt type and level of hydration) and weight of electrolyte salt, the diluent and composition and the volume, with the exact concentration and nature of any solution used to adjust pH. In the preparation of buffers it is proposed that pH adjustment is likely to be the highest source of inaccuracy. One possible solution is to mix whenever possible weighed amounts of solutions/reagents to generate the desired buffer and pH.

3.1.2. Choice of capillary length, diameter and applied voltage

Uncoated fused silica capillaries were used during method development, of 27 cm (20 cm length to detector window)×50 μm I.D.. The particular capillary diameter and length was chosen in conjunction with the applied voltage and other operating parameters to maximise both method sensitivity and resolution, in tandem with achieving low currents and acceptable analysis times. As a result the separation in Fig. 1 was achieved with an applied voltage of +6 kV. With voltages lower than this, unacceptably long analysis times were found with no improvement in resolution. In contrast higher voltages decreased the analysis times at the expense of reduced resolution, which is accompanied by increased current and associated heating within the capillary.

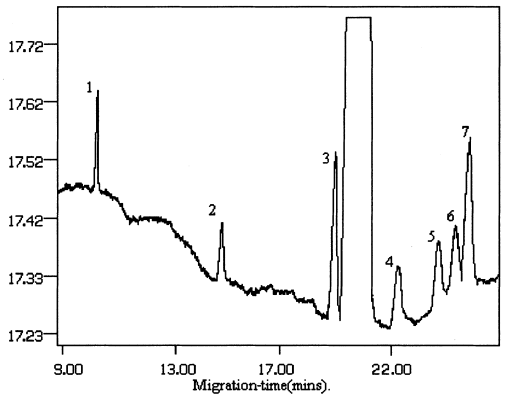


Fig. 1. Optimised separation of ranitidine and the seven related substances. Separation conditions: Fused silica capillary was 27 cm×50 μm I.D.; buffer was 190 m*M* trisodium citrate adjusted to pH 2.6 with citric acid (see Section 2). Voltage 6 kV; detection wavelength 230 nm; operating temperature 25°C.

3.1.3. Choice of detection wavelength

A number of commercial CE instruments have the facility of a UV diode array detector which can be used for spectral scanning of the main peak [16] or related substances, even at the 0.1% level [17]. This type of detector was used in this case for examination of the best mean detection wavelength for ranitidine and three of the seven related substances, which was at, or near, 230 nm, which is the same as that employed in the HPLC method [4].

3.2. Method validation performance

Thus the full parameters for the CE separation of ranitidine and related substances are as shown in Table 2. The next stage was to check these as optimised parameters through validation of the methods selectivity, reproducibility, precision, linearity of response and ruggedness and robustness, along with assessing the detection limits.

3.2.1. Selectivity

A solution of ranitidine (10 mg/ml) and related substances (0.01 mg/ml) in water were used to confirm migration times, by taking seven aliquots of the solution and spiking separately with each of the related substances and analysing with the CE method. The results of this clearly identified the migration

order and showed the selectivity and the migration time reproducibility.

3.2.2. Analyte response factors

Relative response factors were obtained for each related substances and the individual values ranged between 0.9 and 1.1 relative to ranitidine (Table 3). This was in agreement with the HPLC reported procedure [4], which employs a similar detection wavelength. Thus since all response factors are approximately unity, it was appropriate to calculate ranitidine related substances as % a/a of the ranitidine peak.

3.2.3. Precision

3.2.3.1. Migration time and peak area

Ten repeat injections of ranitidine and the seven related substances (0.1% m/m) were used to determine the precision of the relative migration times and peak area ratio (relative to ranitidine). The resulting data (Table 4) demonstrates acceptable R.S.D. values for both the parameters measured. In the assay the electroosmotic flow (EOF) will be considerably suppressed at the operating pH of 2.6 and therefore operational variation can be accentuated (e.g., capillary performance or fluctuation in injection volume). For this reason the internal standard was used for compensation.

3.2.3.2. Calibration and sample preparation

Repeatability in preparation of samples and standards was demonstrated using five individual standards and five samples of the drug substance. With each solution injected in duplicate, the calculated response factors for ranitidine produced precision values of R.S.D.=0.65% (n=10). In addition the levels of the ''Bis compound'' (Fig. 1, peak 3) were determined in each sample prepared and a R.S.D. of 0.89% (n=10) was obtained. It was also necessary to demonstrate acceptable precision using a second analyst preparing fresh solutions and using a different CE instrument. From this work an acceptable precision of R.S.D.=1.1% (n=10) was obtained for the peak area of the ''Bis compound'' using freshly prepared solutions by the second analyst, compared to R.S.D.=0.95% (n=10) for analyst 1.

Table 4
Precision data for the migration time and peak area (relative to ranitidine) for a sample of ranitidine and the seven related substances at the level of 0.1% (a/a)

Analyte	R.S.D. (%, n=10)	
	RMT	PAR
Diamine	0.01	0.07
Oxime	0.02	0.15
Bis	0.03	0.24
I.S.	–	–
Ranitidine	0.01	0.01
Acetamide	0.01	0.34
S-Oxide	0.01	0.17
N-Oxide	0.01	0.36
Adduct	0.01	0.33

RMT=Relative migration time and PAR=peak area ratio (for experimental conditions see Table 2).

3.2.4. Sensitivity

It was proposed earlier that a minimum working detection limit should be 0.1% m/m, and this was determined using a series of standards containing ranitidine spiked with trace levels of ''Bis com-

Table 3
Response factors for ranitidine and related substances determined by HPLC and CE (for experimental details see Table 2)

Name	Standard purity (%, a/a)	Salt-to-base correction factor	Response factor (compared to ranitidine)	
			HPLC	CE
Diamine	96.0	1.271	1.042	1.023
Oxime	98.5	1.229	1.015	1.012
Bis compound	98.0	1.000	0.995	0.982
Nitroacetamide	98.6	1.073	1.083	1.062
S-Oxide	99.2	1.000	1.017	1.022
N-Oxide	98.7	1.000	0.934	0.959
Formaldehyde adduct	98.4	1.000	0.984	0.975

pound''. As a result the absolute detection limit was found to be 0.05% m/m (based on a signal-to-noise ratio in excess of 3). Whereas the limit of quantitation or working limit of detection was 0.1% m/m (based on a signal-to-noise ratio in excess of 10). This solution (0.1% m/m) was then injected ten times and at this working limit a precision of 3.8% R.S.D. was obtained for peak area. This value is within the normal limit applied within GlaxoWellcome of <10%.

3.2.5. Linearity of detector response

The linearity of detector response was obtained for both ranitidine and the related substances. For ranitidine (concentration range 2 to 15 mg/ml) an acceptable correlation coefficient (R^2) of 0.998 was demonstrated ($y=0.7946x-0.7682$, $n=10$).

In a separate exercise a 10.0 mg/ml ranitidine solution was prepared and aliquots of this solution were spiked with levels of ''oxime'' (Fig. 1, peak 2) in the range 0.05 to 1% of the ranitidine concentration. An acceptable correlation coefficient (R^2) of 0.996 was obtained for levels of oxime, with the equation of the line being $y=0.0011x-0.0012$ ($n=10$).

3.2.6. Freedom from interference

To demonstrate freedom from interference a sample of the ranitidine bulk drug from a placebo tablet formulation was analysed. Under the separation conditions of low pH, ranitidine and related substances are positively charged and migrate before the EOF, whereas the tablet excipients in the formulations are generally neutral and migrate with the EOF.

In one particular test formulation, phenol is present as a preservative. When compared to the reference HPLC procedure [4] the peak for phenol is obtained in the middle of the chromatogram causing some interference. However, there is no corresponding interference with CE, as the neutral phenol at pH 2.5 migrates with the EOF, which is poor at this pH, and therefore the phenol is detected very late in the electropherogram.

3.2.7. Robustness

The method was then tested for robustness to deliberate changes in the operating parameters and in examination of worst case changes. In the industry it is normal to test a procedure for its stability, either side of the optimum operating conditions, against small errors in prepared concentrations or in analytical measurements. Moreover, if small changes in parameters result in a large change in response then the suitability of the method may be in question. This however has to be examined in the context of the method. Variability in the performance may be examined very closely in an academic environment but acceptance criteria may be widened in an industrial environment, where the final result and its relationship to the product has a different emphasis. From the method development work on ranitidine it was known that resolution of all the related substances was difficult. It was clear from Section 3.1 and the resultant electropherograms that many effects were contributing to the resolution of each peak pair. One of these effects was the buffer and the need for accurate preparation has been described. For other effects on robustness three further factors were examined in a programme of 17 experiments. To ease the tedium of this programme the structured simultaneous procedure of central composite design was set up, which speeded up the experimental process considerably. The factors considered and the ranges were:

Voltage (kV) 5.70–6.30.
Trisodium citrate (g) in 100 ml 5.46–5.58.
Citric acid (g) in 100 ml 20.97–21.21.

To pick up on any major fall off in performance, the resolution of the peak pairs of three of the related substances were measured, which included:

1. the bis compound and ranitidine (peak 3 and the main peak, Fig. 1),
2. complex nitroacetamide and S-oxide, (peaks 4 and 5, Fig. 1),
3. N-oxide and formaldehyde adduct (peaks 6 and 7, Fig. 1).

In addition the migration time of the formaldehyde adduct (peak 7, Fig. 1) was also used as a measure of the robustness of the method.

3.2.7.1. Resolution between peak 3 and ranitidine

For visualisation of the large data set from the central composite design, response surfaces were developed for the critical peak pairs. In the first case (peak 3 and ranitidine), small changes in citric acid concentration led to relatively large changes in

resolution. This was also the case with trisodium citrate, where initially in the concentration range (5.46–5.52 g/100 ml) relatively large changes in resolution were given but from 5.52–5.58 g/100 ml very little change was observed. Therefore the best resolution occurs at low citric acid and trisodium citrate concentrations and low citric and high trisodium citrate, within the working ranges.

3.2.7.2. Resolution between peak 4 and 5

In contrast for this peak pair, trisodium citrate and citric acid were best at high concentrations. However, at these concentrations the resolution for the other peaks deteriorates.

3.2.7.3. Resolution between peak 6 and 7

No statistically significant model was found for the response to changes in buffer concentrations. However, when resolution was observed outside the above concentration ranges the peaks tended towards comigration. At the opposite end although the resolution increased, this was offset by a loss of resolution between the bis compound and ranitidine, (peak 3 and the main peak, Fig. 1).

3.2.7.4. Migration time for peak 7

For this peak (formaldehyde adduct) changes in applied voltage gave small changes in migration time. Generally an increase in voltage resulted in a decrease in migration time but within the voltage range the migration difference was negligible. Outside the range, applied voltage changes had small to large effects on the resolution of all peak pairs which together with joule heating effects was associated with the magnitude of the voltage increase.

Thus the robustness results indicate that aspects such as the concentration of sodium citrate and citric acid have a considerable effect on individual peak pairs. But when the separation of all components is taken into account the chosen parameter combination appears to give good mean conditions for the separation of ranitidine from its related substances. In examination at the trace level it has to be taken into account that not all the related substances will be observed in a pharmaceutical preparation and that levels down to 0.1% (m/m) are in excess of the pharmacopoeial limits. Therefore the separation involves a difficult mixture of seven closely related substances and the results show that acceptable resolution can be obtained within the specified method limits.

3.3. Application of the method

To assess the CE method against an established GlaxoWellcome HPLC method [4], authentic bulk drug and pharmaceutical preparations of ranitidine were assayed. The standard TLC method was also included in the assay comparison. Statistical comparison of the results for CE and HPLC were carried out with an *F*-test where the HPLC results were the reference method.

3.3.1. Assay of the ranitidine bulk drug

The purity of the ranitidine drug was determined against the HPLC procedure [4] from a bulk supply. Different masses were assayed by the two methods and for CE the purity value was 99.8% (m/m) and likewise 99.8% (m/m) for the HPLC procedure ($n=4$) against aqueous standards at 10 mg/ml in water. A statistical comparison (*F*-test) was also carried out between the two techniques and no statistical difference between the methods was observed.

3.3.2. Assay of Zantac (10 and 25 mg ranitidine hydrochloride/ml) as 5 ml commercial injections

3.3.2.1. Determination of the parent compound in the Zantac (10 and 25 mg/ml) injection

The ranitidine hydrochloride in four Zantac 10 mg/ml injection solutions was determined using the developed CE method. From this a mean assay value of 10.1 mg/ml ($n=4$) was obtained. For the 25 mg/ml injections a mean value of 25.2 mg/ml ($n=4$) was given. These values compare favourably with the labelled strengths and are well within the allowable limits for the active substance.

3.3.2.2. Determination of the related substances

In newly manufactured batches, levels of related substances in the injections are very low and therefore a Zantac injection (10 mg/ml) which was at 38 months an end of life batch (5 ampoules), were assayed. The ampoules were pooled and the internal standard (3 mg/ml) was added as the diluent to give 5 mg/ml sample solutions. Two batches of injection

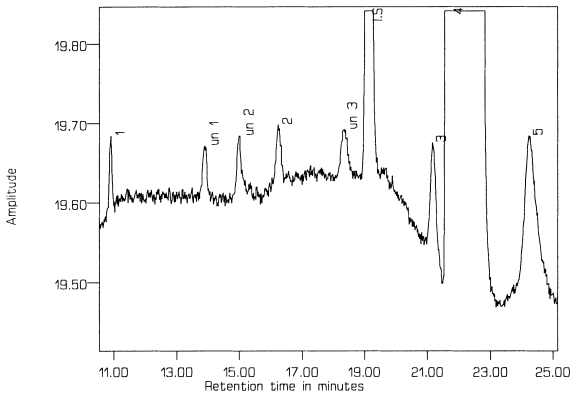


Fig. 2. Resolution of the components in a 10 mg/ml Zantac injection. Separation conditions: Fused silica capillary 27 cm×50 μm I.D.; buffer equivalent to 190 m*M* trisodium citrate adjusted to pH 2.6 with citric acid (see Section 2). Voltage 6 kV; detection wavelength 230 nm; operating temperature 25°C.

(B404 and B405) were assayed and the electropherogram in Fig. 2 was given for one of the samples. In the CE assay, the diamine (0.03%, a/a), oxime (0.04%, a/a), Bis (0.1%, a/a), nitroacetamide (0.24%, a/a) and a number of unknown peaks are seen in the CE assay (Table 5). The latter are not resolved in the other techniques. The nitroacetamide levels are higher in the CE method which gives an easily measured peak, than from HPLC and TLC, where there is poor peak shape and spot detection. The remaining related substances, S-oxide, N-oxide and the formaldehyde adduct, are not picked up in the CE and HPLC procedures and are not considered to be the main degradation products. However, small amounts were observed (0.05%, a/a) by TLC.

It is interesting to note the variation of ~1 min in the migration time in Fig. 2 compared to Fig. 1. This is likely to be due to the difference in the viscosity of the Zantac formulation against the aqueous standard. However, compensation for this variation in the CE assay is achieved by calculating relative migration times (relative to the main peak).

4. Conclusions

A CE method was developed and validated for the assay of ranitidine and potential related substances in bulk drug and pharmaceutical preparation. The ionic strength and pH of the electrolyte were shown to be the most critical parameters affecting selectivity. The capability of detecting the appropriate limits of the related substances at (0.05%, a/a, against ranitidine) has been demonstrated, which is in line with the TLC limits as proposed by the British Pharmacopoeia. Acceptable method performance has been demonstrated for precision and peak migration times and areas, and linearity of detection is very good. Robustness of the method with individual peak pairs is slightly variable, but taken over all the related substances the best conditions fulfil the acceptance criteria. The method was thus successful-

Table 5
Determination of ranitidine related substances in injections which were at 38 months an end of life batch (10 mg/ml in 5 ml ampoules) (for experimental conditions see Table 2)

Name (peak No.)	% (a/a) of ranitidine Batch					
	B404 (6 months)			B405 (6 months)		
	HPLC	TLC	CE	HPLC	TLC	CE
Diamine (1)	0	0	0.03	0	0	0.03
Oxime (2)	0.01	0	0.04	0.01	0	0.04
S-Oxide	0	0.05	0	0	0.05	0
N-Oxide	0	0	0	0	0	0
Adduct	0	0.05	0	0	0.05	0
Nitroacetamide (5)	0.03	0.05	0.24	0.03	0.05	0.26
Bis- (3)	0.06	0.05	0.1	0.06	0.2	0.12
Unknown 1 (un 1)	0	0	0.03	0	0	0.03
Unknown 2 (un 2)	0	0	0.03	0	0	0.03
Unknown 3 (un 3)	0	0	0.04	0	0	0.04

ly applied to examination of bulk drug and injections.

Overall the results support the view that CE is a useful alternative or complementary method to the established TLC and HPLC methods and can be used to generate quantitative related substance information on ranitidine hydrochloride-containing formulations.

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