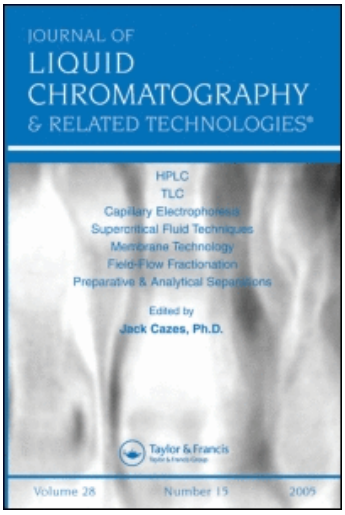

Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

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To cite this Article Thomas, B. R. and Ghodbane, S.(1993) 'Evaluation of a Mixed Micellar Electrokinetic Capillary Electrophoresis Method for Validated Pharmaceutical Quality Control', Journal of Liquid Chromatography & Related Technologies, 16: 9, 1983 — 2006
To link to this Article: DOI: 10.1080/10826079308019909
URL: http://dx.doi.org/10.1080/10826079308019909

JOURNAL OF LIQUID CHROMATOGRAPHY, 16(9&10), 1983-2006 (1993)

EVALUATION OF A MIXED MICELLAR ELECTROKINETIC CAPILLARY ELECTROPHORESIS METHOD FOR VALIDATED PHARMACEUTICAL QUALITY CONTROL

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ABSTRACT

A stability indicating, quality control analysis method was developed for VASOTEC, an antihypertensive drug. Mixed micellar capillary electrokinetic electrophoresis employing the anionic detergent, sodium lauryl sulfate (SDS), and the nonionic detergent, Brij 35, was used to separate enalapril maleate (EM), the active component of VASOTEC, and its degradates, the enalapril diketopiperazine (DKP) and enalaprilat (DA). The formation of easily separable rotary isomers by EM required that the capillary temperature be maintained at 50°C except in the presence of Brij 35. Adjusting the Brij 35 concentration allowed manipulation of selectivity for the neutral DKP degradate with respect to EM and DA. Detection sensitivity was enhanced ten-fold to 0.1 ug/mg by utilizing a 100 micron I.D. rather than a 75 micron I.D. capillary. Theoretical plates were determined to be 71,000 plates/m for the active component or 70 times that for the HPLC method. Critical factors in method stability were pH, temperature and applied voltage. Concentrations of SDS, Brij 35 and sodium borate could be varied ±5%

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without significant effects on the separation. Capillary electrophoresis was determined to be potentially useful for drug quality control analysis because of its resolution, speed, capillary longevity and versatility, low solvent consumption, long term expense, and simplicity of training and operation.

INTRODUCTION

Capillary electrophoresis (CE) is a newly introduced analytical technique which has promise for pharmaceutical analysis because of its inherently high resolution (1). Modification of this technique to include detergents in the capillary buffer which allowed separation of neutral, as well as charged molecules, was introduced by Terabe (2) and termed micellar electrokinetic chromatography (MEKC). The utility of MEKC for pharmaceutical analysis has been examined by Nishi and Terabe (3) with the conclusion that the technique had value for assay of active ingredients. The theory and practical applications of MEKC have recently been reviewed (4). Combinations of anionic and nonionic detergents, such as sodium lauryl sulfate (SDS) and Brij 35, have also been used to provide added selectivity in difficult separations. This technique was called mixed MEKC (5). A comprehensive review of CE advances, which included pharmaceutical applications, has recently been published (6).

VASOTEC contains an angiotensin-converting enzyme (ACE) inhibitor that acts by suppression of the renin-angiotensin-aldosterone system (7). Its primary effect is lowering blood pressure. The active component is enalapril maleate (EM) which forms chromatographically separable rotary isomers (8). Enalapril maleate is an ethyl ester prodrug that is hydrolyzed to a diacid form which is the ACE inhibitor *in vivo* (7). The diacid form, enalaprilat (DA), may occur as a degradate *in vitro* and must be assayed to ensure potency. Enalapril diketopiperazine

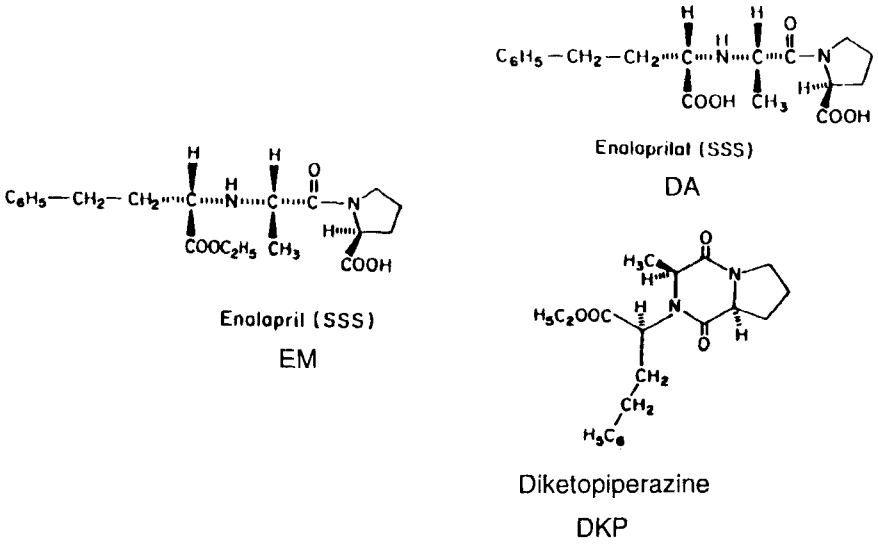

Figure 1. Structures of VASOTEC component molecules.

(DKP) is a cyclic degradate which is assayed to ensure purity with stability indicating methods. Figure 1 shows the chemical structures of these compounds. Note that the EM contains one carboxylic acid group and the DA contains two, while the DKP lacks a strongly charged functional group and is relatively hydrophobic.

The purpose of this study was to evaluate CE as an analytical tool for Quality Control (QC)use. QC methods must be precise, accurate and rugged. Federal regulation and good manufacturing practice require reliable testing to assure that demonstrably pure drugs are delivered at the proper strength. Economics dictate that accurate drug formulation will not waste expensive active ingredients. Methods to assay purity and potency must be rugged for use by operators in different laboratories, and often, different countries. Nielson and Rickard (9) have

discussed the limiting factors in quality control for their validation of a capillary zone electrophoresis method for human growth hormone. These factors included linearity, precision and sensitivity. Methodology and manipulable parameters in capillary electrophoresis useful for pharmaceutical analysis have been presented by McLaughlin et al. (10). These authors give practical guidelines for methods development in capillary electrophoresis.

Most capillary electrophoresis separation methods are developed to achieve maximum resolution or sensitivity, but for drug quality control applications, precision, accuracy and ruggedness are also very important. Partial validation of a capillary zone electrophoresis analysis method for VASOTEC components, enalapril maleate and enalaprilat, has been accomplished, but without the ability to quantitate the DKP degradate (11). We present an accurate, rapid and rugged pharmaceutical quality control method for all three VASOTEC components.

MATERIALS AND METHODS

Capillary electrophoresis was performed on a Spectra Physics (San Jose, CA) SpectraPhoresis 1000 system using version 1.04 software. A manually threaded 100 micron I.D. x 300 micron O.D. x 70 cm L x 63 cm detection length Chrompack (Raritan, NJ) capillary was used in a Spectra Physics cassette. In some development assays, a Spectra Physics 75 micron I.D. x 70 cm L capillary was utilized. The anode or capillary buffer consisted of 20 mM sodium tetraborate pH 8.5 containing 2% Brij 35 (Technicon, Tarrytown, NY) and 20 mM SDS (99%,Sigma Chemical Co., St. Louis, MO) in validation assays. Standards were dissolved in and samples extracted in 50% methanol (HPLC grade), 10 mM sodium borate pH 8.5. Spiked placebos were

sonicated and shaken for 30 minutes each in volumetric flasks containing one-half the volume extraction buffer, then brought to volume before injection. Separating conditions for validation were 15 kV applied voltage at 50°C. Injections were hydrodynamic for 2.5 s after a 2 min. capillary buffer fill time. Detection wavelength was 200 nm. A 10 min. buffer wash with capillary buffer at 60°C was used to equilibrate the system before starting a sequence of assays. Ten or less assays were performed from one 1.8 mL anode buffer vial to prevent buffer depletion. The cathode reservoir was flushed at the end of each sequence or after ten assays. At the start and end of each day, a five minute wash with 0.1 N sodium hydroxide and water prepared the capillary for use or storage.

The validation procedures employed were those used to validate HPLC methods for QC use at Merck & Co.. Precision, accuracy, limits of detection and quantitation, range, linearity and method stability were determined. Precision, range, accuracy and linearity were determined by spiking placebo samples with the active ingredient, EM, and the degradates, DA and DKP. The placebo consisted of the inactive ingredients or excipients found in VASOTEC tablets which were weighed and mixed in the same quantities as in 20 mg tablets. Spiked placebo assays were conducted at 50%, 75%, 100%, 125% and 150% of a 0.2 mg/mL concentration of EM in the presence of 0.01 mg/mL (5% of active) for each degradate, DA and DKP. On each of three days, triplicate samples were assayed at each of the five levels, 50-150%. Degradates were assayed similarly at 20%, 50%, 80%, 100% and 120% of 0.01 mg/mL in the presence of 0.2 mg/mL EM and 0.01 mg/mL of the other degradate. Recoveries were determined by comparing peak areas of standard solutions to extracted samples of spiked placebos.

Limit of detection was determined by finding the concentration of each degradate at which a peak three times the area of the ambient noise occurred in otherwise standard conditions. Limit of quantitation was the minimum concentration for each degradate that could be reproducibly and accurately integrated. Instrument precision was determined with ten injections of the same standard on the same day at identical injection times. Factors which could affect the assay were varied ±5% to establish the stability of the method.

RESULTS

Methods Development

Capillary Buffer Screening

A screening of separation conditions was performed in 10 mM sodium borate pH 9.5 at 50°C at 200 nm and an applied voltage of 20 KV (Figure 2.A). The capillary was 75 microns I.D., 375 microns O.D., 75 cm in length and 63 cm detection length. In the absence of detergents the standards separated well and stable rotamer formation by the EM was inhibited by the high temperature. The DKP, as a neutral uncharged molecule, migrated with the electroosmotic flow (EOF) which prevented accurate integration. The detergents, sodium lauryl sulfate (SDS), SDS with 2% Brij 35, cholic acid (CA) and deoxycholic acid (DOC) were screened to determine their effect on the separation. SDS at 20 mM yielded a good separation of the three components and DKP was separated from the EOF marker (Figure 2.B). The neutral DKP partitioned into the SDS micelle to a greater extent than the EM and DA. Figure 2.B also shows that the rotamer separation is inhibited at 50°C. When the temperature was lowered to 25°C, EM

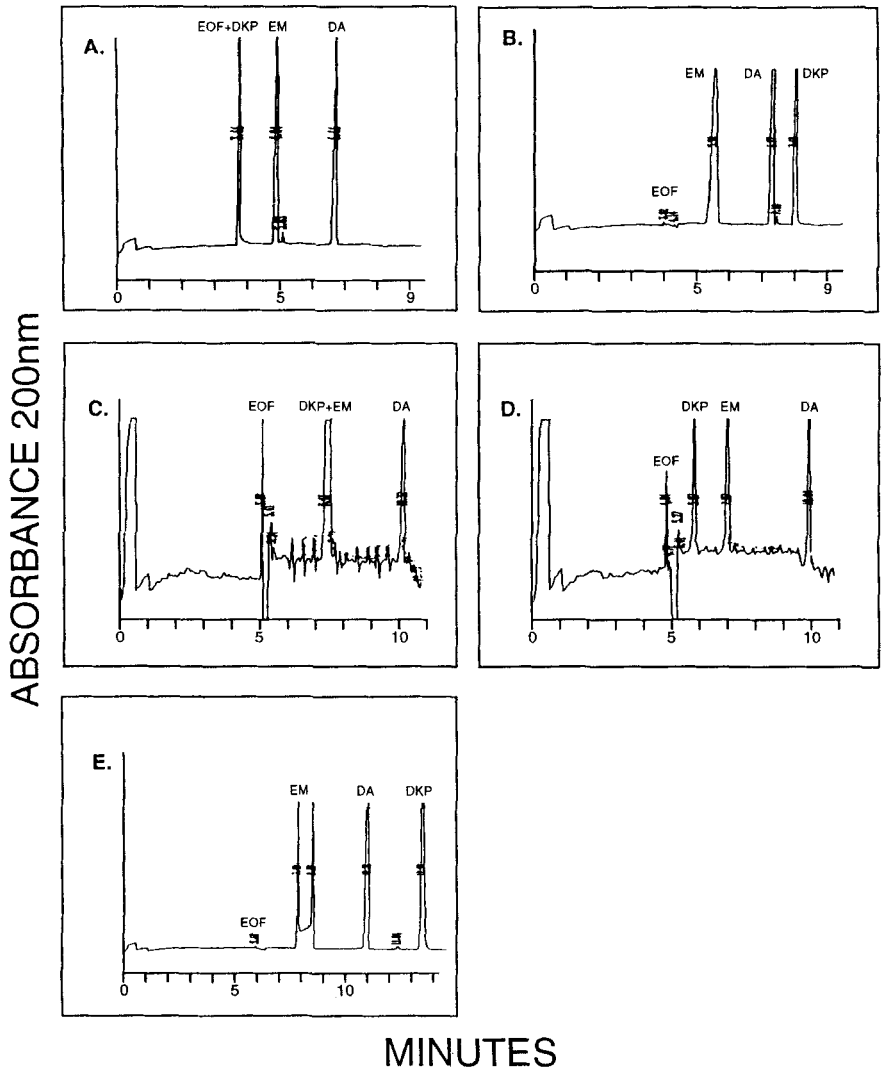

Figure 2. Detergent screening for VASOTEC assay. (A) 10 mM sodium borate pH 9.5 at 50°C and 20 kV, (B) same as A + 20 mM SDS, (C) same as A + 10 mM deoxycholic acid, (D) same as A + 10 mM cholic acid, (E) same as A at 25°C.

rotamers separated (Figure 2.E). This temperature effect on EM rotamers separation in CE was first observed by Qin et al.(11). Addition of 2% Brij 35, a nonionic detergent, to SDS affected the selectivity of the separation as shown in Figure 3. The DKP which migrated last with SDS alone, migrated first with the addition of 2% Brij 35. The separation resembled that found with cholic acid (Figure 2.D) except that the DKP is better separated from the EOF marker and the total assay time is reduced by 50%.

Deoxycholate in Figure 2.C separated all components well from the EOF marker, but DKP and EM comigrated. Cholic acid in Figure 2.D separated all components, but the DKP peak was not well separated from the EOF marker. The electroosmotic flow was differentially affected by detergents at 10 mM concentration and the bile acids increased EOF marker migration time by about 25%. The SDS micellar buffer did not affect the EOF marker migration rate, but the addition of Brij 35 decreased the migration time by almost 40% without degrading resolution.

Brij 35

The effects of Brij 35 addition were investigated by varying the Brij 35 concentration in 10 mM borate pH 9.5 with 20 mM SDS. Figures 2.B and 3.A illustrate the effect of increasing Brij 35 concentrations from 0 to 2%. The selectivity of the separation for DKP, a neutral molecule, was changed relative to the charged molecules, EM and DA. The EOF marker migration time and the total separation time was reduced to 50% of that with SDS alone.

Figure 4 details the relationship of a nonmicellar capacity factor (t_m-t_o/t_o) for all three components (tm

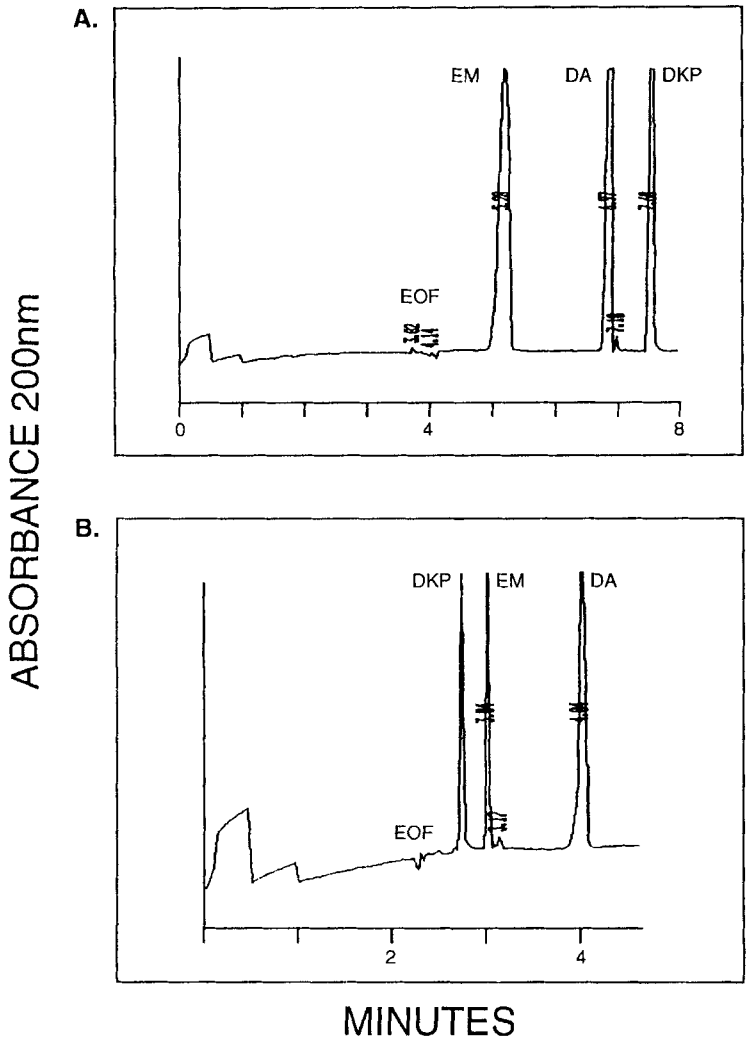

Figure 3. Electropherogram of a standard mixture of EM, DA and DKP under the final experimental conditions.

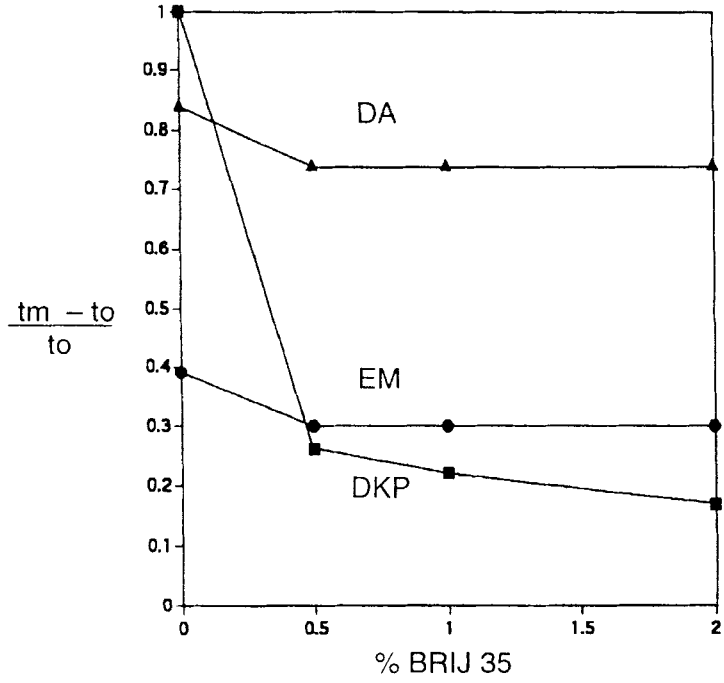

Figure 4. Selectivity effects of Brij 35 on migration times.

and to are the migration times of the solute of interest and the EOF marker). The ratio of the migration times of DA and EM (DA/EM) was constant with or without Brij 35 which clearly demonstrated that the nonionic detergent had no effect on the selectivity of the separation for these two acidic components. Effects of Brij 35 on the neutral molecule, DKP, were large. Migration time decreased three-fold and the separation of DKP from EM and DA varied with Brij 35 concentration. With higher Brij 35 concentration, there was more rapid DKP migration relative to the charged EM and DA. Brij 35 reduced partitioning of DKP into the SDS micelle in a concentra-

tion dependent fashion. This indicated that the Brij 35 was acting as a pseudomobile phase with the SDS micelle acting as a pseudostationary phase. The migration time of the DKP peak was adjusted relative to the EM peak by varying Brij 35 concentration.

The EM and DA acids were repelled by the strongly acidic surface of the SDS micelle and did not partition into it. The addition of Brij 35 also did not affect the two acidic components. The DKP was relatively hydrophobic and partitioned into the SDS micelle (Figure 2.B). Brij 35 is a polyoxyethylene detergent with a long polyether segment ($C_{12}E_{23}$). In the mixed micelles formed from SDS and Brij 35 the polyether groups of the Brij 35 decreased the hydrophobicity of the micelle and thereby the hydrophobic partitioning of the DKP into the micelle. As the Brij 35 concentration was increased the hydrophobicity of the micelle decreased which progressively decreased DKP hydrophobic interaction with the micelle until at 2% Brij 35 in 20 mM SDS the micelle did not become more hydrophilic with increasing Brij 35 concentrations. With 2% Brij 35 the DKP partitioning into the mixed micelle was minimal, but was enough to separate the DKP from the EOF marker peak and allow integration.

Internal Standard

The utility of an internal standard for quantitation was investigated using simvastatin as the internal standard. Simvastatin is the active ingredient of ZOCOR, a Merck & Co., Inc. drug. The internal standard migrated more slowly than EM, but faster than DA and was well separated, but in comparison to external standard quantitation, the internal standard quantitation was less reproducible. This may be a result of the greater hydrophobicity of the simvastatin compared to EM.

Validation assays were performed using an external standard method because integration was more accurate and sample preparation was facilitated.

Capillary Diameter

The 75 micron I.D. capillary originally used for the method development allowed only a 1 ug/ml minimum detection limit and 10 ug/ml of the degradates could be reliably integrated. Sensitivity was increased by increasing capillary internal diameter. A 100 micron I.D. capillary yielded an order of magnitude increase in sensitivity allowing reproducible integration at 1 ug/ml and minimum detection limit of 0.1 ug/ml for the degradates. The increased column diameter allowed a greater injection volume for the same injection time and a longer light path for the detector. Comparison of figures 3 and 5.A demonstrated the difference in the electropherograms for 75 micron and 100 micron I.D. capillaries, respectively. The peaks were broader with the 100 micron I.D. capillary, but good resolution was maintained. Maximum resolution was traded for sensitivity by use of the larger bore capillary. When a 250 micron I.D. (44 cm L) capillary was tested the resulting peaks were excessively broad and sensitivity was not increased over that obtained with the 100 micron I.D. capillary. Increasing capillary diameter can increase sensitivity in some cases, but the effect is limited by peak broadening.

Final Conditions

The strategy pursued in developing this method is evident in the final conditions used for the drug assays. The high resolution obtained with CE was partially sacrificed in order to obtain a fast, sensitive and selective assay. Figure 5.A demonstrates the spiked

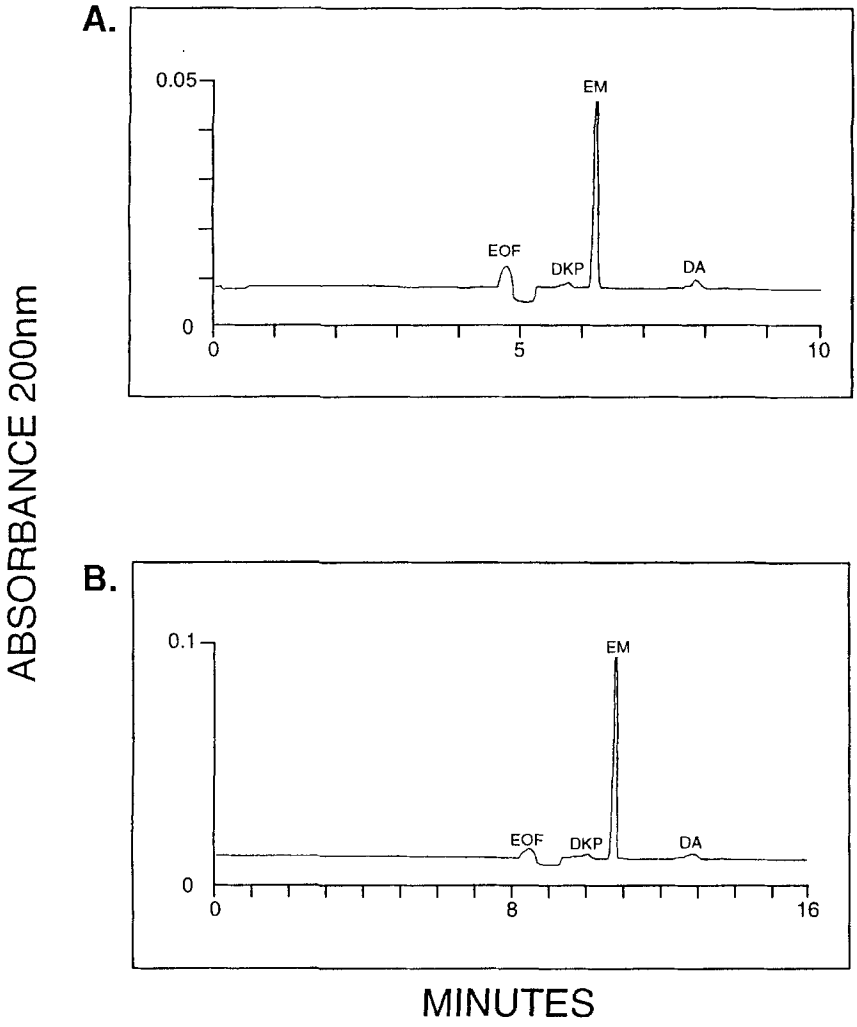

Figure 5. Electropherogram of a placebo spiked with EM, DA and DKP under final experimental conditions (A) and at a temperature of 25°C (B).

placebo electropherogram obtained under the final assay conditions of 20 mM sodium borate pH 8.5, 20 mM SDS and 2% Brij 35 at a temperature of 50°C and an applied voltage of 15 kV (approximately 45 uA current and 0.24 gigaOhms resistance). A 2.5 injection time and two min. capillary buffer washes between assays were used. Each buffer vial was calibrated with three injections of standards for external standard quantitation and six or less sample assays were run from each buffer vial after calibration. A 100 micron I.D. capillary was employed to increase detection sensitivity for the degradates. The ratio of 20 mM SDS to 2% Brij 35 was chosen to force the DKP to migrate faster and yet resolve it from the EM peak and the EOF marker peak. The Brij 35 allowed higher applied voltages to be used. In the absence of Brij 35, the assay run time was longer, the currents were higher and higher buffer concentrations (>20 mM) could not be used. The addition of Brij 35 allowed adjustment of the DKP peak to any position relative to the other components. The VASOTEC component molecules could be separated without Brij 35, but its presence added versatility to the assay.

Under identical conditions, except temperature, it was observed that in the presence of Brij 35 at 25°C, the separation of the rotary isomers of EM did not occur as had been observed in SDS alone at 25°C (Figures 5.B and 2.E). The Brij 35 decreased the hydrophobic interaction with the SDS micelles necessary to separate the rotary isomers. Despite the ability to eliminate the stable rotamer formation at 25° C with Brij 35, the final assay was performed at 50° C because of improved peak shapes and shorter assay run time.

Under the final conditions, the micellar migration marker, Sudan III, migrated at 6.5 min. with the EOF marker, methanol, at 4.8 min. The DKP was within this

relatively narrow micellar window. The micellar k' for DKP, (Tm-To)/[To(1-Tm/T micelle)], was 2.1 which is near the optimum for an analyte in a micellar system.

EVALUATION OF METHOD VALIDATION

Spiked Placebos

Table 1 shows the results for placebos spiked with the active component EM at five levels ranging from 50 to 150 % of label claim (c = 0.2 mg/mL). EM was assayed in the presence of 5% of each degradate, DKP and DA. Assays were conducted with fresh buffer and sample preparation each day for three days. The linearity results for EM (slope and correlation coefficient) conformed to the internal validation acceptance criteria for active ingredients over the range tested. When pooled over all days and all levels, the data exhibited an acceptable (2 % or less) bias (assayed mean less the actual quantity of active or X-theory) of 1.6 %. On a given day or over three days at a given level, the bias sometimes exceeded the limit of 2%. The 95% confidence interval, defined as twice the standard deviation, s (sigma), was 6.2 % and also exceeded the 2% limit.

Assays for the degradates, DKP and DA, as listed in Tables 2 and 3 were, as expected, not as precise or reproducible as for the EM which was assayed at a higher concentration. Data for the DA degradate were acceptable from the standpoint of accuracy (bias of 0.5%) and precision. They were more precise and reproducible than those for DKP. Linearity for the DA degradate was good, but inadequate for the DKP degradate. The minimum amount of the degradates that could be reliably integrated was 1 ug/mL. The minimum amount detectable was approximately 0.1 ug/mL. These minimums were possible only with the

TABLE 1
PLACEBO ASSAYS: ENALAPRIL RECOVERIES

Accuracy and Precision

Level	No. of Samples	Average Assay	Bias (X-Theory)	Standard Deviation	Precision (±2s)
50%	9	52.9%	2.9%	2.5%	± 5.0%
75%	9	78.0%	3.0%	2.0%	± 4.0%
100%	9	103.1%	3.1%	1.5%	± 3.0%
125%	9	124.1%	-0.9%	3.3%	± 6.6%
150%	9	149.7%	-0.3%	4.9%	± 9.8%
Pooled Overall					
100%	45	101.6%	1.6%	3.1%	± 6.2%

Day	n	Theoretical Average Assay	Average Assay	Bias (X-Theory)	Average Standard Deviation	Precision (± 2s)
1	15	100.0%	101.8%	1.8%	1.1%	± 2.2%
2	15	100.0%	100.3%	0.3%	2.6%	± 5.2%
3	15	100.0%	102.6%	2.6%	2.5%	± 5.0%

Linearity

Slope = 0.96%/%
R^2 = 99.17%

TABLE 2
PLACEBO ASSAYS: DKP RECOVERIES

Accuracy and Precision

Level	No. of Samples	Average Assay	Bias (X-Theory)	Standard Deviation	Precision (±2s)
20%	9	25.0%	5.0%	5.9%	±11.8%
50%	9	52.0%	2.0%	7.4%	±14.8%
80%	9	76.3%	-3.7%	10.6%	±21.2%
100%	9	109.1%	9.1%	6.5%	±13.0%
120%	9	118.8%	-1.2%	11.1%	±22.
		Pooled Overall			
74%	45	76.2%	2.2%	8.6%	±17.2%

Day	n	Theoretical Average Assay	Average Assay	Bias (X-Theory)	Average Standard Deviation	Precision (±2s)
1	15	74.0%	75.6%	1.6%	3.8%	± 7.6%
2	15	74.0%	77.8%	3.8%	7.6%	± 15.2
3	15	74.0%	75.3%	1.3%	5.6%	± 11.2

Linearity

Slope = 0.97%/%
R^2 = 93.39%

TABLE 3
PLACEBO ASSAYS: ENALAPRILAT RECOVERIES

Accuracy and Precision

Level	No. of Samples	Average Assay	Bias (X-Theory)	Standard Deviation	Precision (±2s)
20%	9	19.8%	-0.2%	2.7%	± 5.4%
50%	9	54.2%	4.2%	2.6%	± 5.2%
80%	9	76.9%	-3.1%	8.7%	±17.4%
100%	9	101.2%	1.2%	3.7%	± 7.4%
120%	9	120.6%	0.6%	4.4%	± 8.8%
		Pooled Overall			
74%	45	74.5%	0.5%	5.0%	±10.0%

Day	n	Theoretical Average Assay	Average Assay	Bias (X-Theory)	Average Standard Deviation	Precision (±2s)
1	15	74.0%	72.0%	-2.0%	1.9%	± 3.8%
2	15	74.0%	75.8%	1.8%	2.3%	± 4.6
3	15	74.0%	75.8%	1.8%	3.2%	± 6.4

Linearity

Slope = 0.99%/%
R^2 = 97.84%

100 micron I.D. capillary. In preliminary assays of other drugs, the minimum detectable amounts were approximately an order of magnitude better than enalapril maleate, because of their greater UV absorbance.

Although variability caused reproducibility over three days to exceed Merck's internal specifications, the results of this study were encouraging. More developmental work is underway in our laboratory to gain the performance increase necessary to meet the internal specifications.

Methods Stability

Stability of the method was determined by making small changes in the factors which have an effect on the assay. The parameters varied were SDS, Brij 35, and sodium borate concentration, pH, applied voltage and temperature (Table 4). The temperature, applied voltage and pH caused the largest changes in migration time for EM, DKP, and DA. Temperature and applied voltage are precisely controlled by the CE instrument; therefore, only the pH may vary in normal assays. The large changes in migration time as a result of small pH changes demanded that capillary buffer pH be maintained as precisely as possible. Capillary buffer was prepared daily and its pH was adjusted to within 0.05 pH units of pH 8.5.

DISCUSSION

Capillary electrophoresis is uniquely suited to quality control drug analysis. CE has often been and will continue to be directly compared to HPLC despite basic differences (13). The current situation between CE and HPLC is similar to that which occurred 10-15 years

TABLE 4

Method Stability

PARAMETER VARIED		MIGRATION TIME(min) Control*	+5%	-5%
SDS Concentration	DKP	5.2	5.3	5.2
	EM	5.6	5.6	5.7
	DA	6.9	7.0	6.9
Brij 35 Concentration	DKP	5.2	5.2	5.3
	EM	5.6	5.5	5.6
	DA	6.9	6.9	7.1
Sodium Borate Concentration	DKP	5.2	5.1	5.1
	EM	5.5	5.5	5.4
	DA	7.0	6.8	6.8
pH	DKP	5.2	4.9	9.2
	EM	5.6	5.2	>10
	DA	6.9	6.1	>10
Applied Voltage (kV)	DKP	5.1	4.8	5.5
	EM	5.5	5.0	5.9
	DA	6.8	6.2	7.2
Temperature	DKP	5.1	4.7	5.2
	EM	5.5	5.0	5.6
	DA	6.8	6.3	6.9

*Standard conditions used for controls were 20 mM Borate pH 8.5 with 20 mM SDS and 2% Brij at 15 KV applied voltage and 50°C. Parameters were varied ±5% with all other conditions held constant.

ago between HPLC and gas chromatography (GC). Just as GC is still in common use and complements HPLC, HPLC will continue to be used for many analyses.

The utility of CE for drug analysis is the result of a number of advantages of CE over HPLC some of which are listed in Table 5. Resolution is a primary advantage. CE is an inherently high resolution technique which commonly yields theoretical plate counts 10 to 1000-fold

TABLE 5

Comparison of VASOTEC HPLC and CE Analysis Methods

Factor*	Component	HPLC	CE
Capacity K' = Tm-To/To	DKP	>4	>0.1
	EM	7<K'<20	>0.2
	DA	>1.3	>0.5
Theoretical Plates $N=16\ (Tm)^2/W$	DKP	>1500	19,000
	EM	> 900	71,000
	DA	>1000	34,000
Peak Tailing T = (a + b)/2a	EM	<2.5	1.0
Resolution R=2(Tm1-Tm2)/(W1+W2)		(EM/Maleate) >5	(EM/EOF) >3
%RSD (10 Replications)	EM	≤2.0%	≤2.0%
Run Time		20 Min.	10 Min.

* Tm = Migration time of a peak, To = Migration time of the EOF marker peak, W = Width of peak at baseline, a + b = Width of peak on each side of a perpendicular through the peak center at the baseline.

higher than HPLC. In practical terms, this makes methods development easy. If there is even a small migration behavior difference between two molecules, they will separate as a result of the large number of theoretical plates. Table 5 indicates that for the VASOTEC separation, the theoretical plate count (N) is at least 10-fold higher for all three components.

Speed is another advantage of CE. The run times of CE separations are usually faster than that of HPLC separations. The VASOTEC CE separation is twice as fast as an HPLC separation (Table 5). Solvent volume and composition favor CE assays. A CE system used continuously will use one or two liters of anode and cathode buffer each week while HPLC can consume that volume in less than a day. HPLC produces large amounts of organic solvent waste which can be a difficult and expensive disposal problem, while CE produces little or no organic solvent waste.

Theoretically, a CE capillary can be used for a long time and can be used for many drug analysis methods. The capillary is an empty silica tube filled with frequently changed buffer. There is no packing material to degrade as in HPLC columns. The silica capillary can be cleaned and conditioned easily and often under stringent conditions using strong acids, bases and organic solvents. A fused silica capillary may last for a long time and the capillary threaded for the VASOTEC assay cost less than $15 to replace. Expensive and short-lived HPLC columns can be eliminated.

Simplicity is an often overlooked advantage of CE (14). Commercial CE instruments are generally totally integrated units controlled by computer which result in a simplified user interface. Experience in this laboratory has indicated a shorter training period for inexperienced CE operators that contrasts with longer HPLC training. The well known disadvantage of CE compared to HPLC is sensitivity leading to detection limits that are not useful for trace analysis. In quality control drug analysis, this limitation does not apply. Drug concentrations may be adjusted to the levels necessary for adequate sensitivity and reproducible peak integration.

Further development of the MEKC method for VASOTEC is planned. Specifically, a ten-fold increase in concentration of the active in the extraction buffer may provide greater sensitivity necessary for better degradate detection and less quantitation error. A change in the capillary buffer pH, detergent and buffer concentration may provide better peak shape and more consistent integration of peak areas.

This method for VASOTEC has allowed evaluation of CE for quality control applications. More work is needed however to generate improvements in reproducibility and accuracy which are necessary to meet internal specifications. Experience gained in this project suggests that CE is potentially useful for quality control analysis of pharmaceutical compounds. The second generation of CE instruments should exhibit better injection and detection performance. This would result in the improvement of accuracy, precision and reproducibility for CE methods.

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