Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Chromatography A, 995 (2003) 1–10
JOURNAL OF CHROMATOGRAPHY A
www.elsevier.com/locate/chroma

Systematic error in automated in-tube solid-phase microextraction

Anil R. Raghani*, Kristin N. Schultz
Pharmacia Corporation, Global Chemical Process R&D, 4901 Searle Parkway, Skokie, IL 60077, USA

Received 18 November 2002; received in revised form 7 March 2003; accepted 14 March 2003

Abstract

The widely employed configuration for automated in-tube solid-phase microextraction (SPME) involves modification of a commercial liquid chromatographic autosampler into an automated extraction device. This popular configuration is demonstrated to result in an inherent systematic error in the quantitation of analyte in a given matrix. The source of error is traced to the accumulation of analyte in the extraction and the pre-extraction segment (i.e., sample loop, metering valve and tubing prior to the metering valve) of the autosampler where the analyte comes in contact with the residual mobile phase. This results in cross-contamination due to sample/mobile phase mixing. The quantity of analyte accumulated in these segments is shown to consistently increase with the increasing number of draw/eject cycles. As a result of the accumulation, the amount of analyte recorded leads to inaccurate quantitative information, leading to overestimation of the limit of detection and limit of quantitation, when automated in-tube SPME is employed as an approach for sample enrichment. Insertion of a 100-μl air plug prior to extraction step was able to significantly minimize sample/mobile phase mixing of analyte with the residual mobile phase in the pre-extraction and extraction step, thus minimizing the systematic error.

Keywords: Solid-phase microextraction; In-tube solid-phase microextraction; Automation; Systematic errors; Instrumentation; Ranitidine

1. Introduction

Accurate analysis of components present in difficult matrices such as plasma, serum, whole blood, tissue homogenates, saliva or urine requires sample pretreatment procedures to allow clean up of these matrices prior to the quantitation of components of interest. On the other hand, determination of analytes in dilute samples such as environmental samples often needs a sample enrichment step where analytes of interest are concentrated before they are analyzed. Various sample pretreatment methods are utilized for sample clean-up or enrichment. These include, but are not limited to protein precipitation, liquid–liquid extraction (LLE), solid-phase extraction (SPE), solid-phase microextraction (SPME), liquid-phase microextraction (LPME) and membrane methods, e.g., dialysis, and ultracentrifugation, etc. An extensive review on the subject of sample preparation is discussed in Ref. [1]. Of these techniques, SPME has gained the widest acceptance due to better sensitivity compared to direct injection, high analyte recovery, extraction reproducibility, and commercial availability of SPME devices for gas chromatographic analysis. For liquid chromatographic analysis, SPME and most recently automated in-tube SPME have gained

*Corresponding author. Tel.: +1-847-982-7081; fax: +1-847-982-4657.
E-mail address: anil.r.raghani@pharmacia.com (A.R. Raghani).

0021-9673/03/$ – see front matter
doi:10.1016/S0021-9673(03)00499-0

wide acceptance as a method of choice for the sample clean up and enrichment for the analysis of non-volatile analytes. Automated in-tube SPME is successfully applied for the analysis of environmental [2,3], biological [4,5], food [6], forensic [7], and drug samples [8,9]. A review on the applications automated sample preparation using in-tube SPME has recently been published [10].

Most of the published work on automated in-tube SPME has predominantly employed one commercial high-performance liquid chromatography (HPLC) system (Model HP1100, Agilent Technologies, USA) where the autosampler of this system is configured for the purpose of automated extraction. This commercial equipment was thoroughly compared with other available system for the application of in-tube SPME [11]. Because of the ease of automation with the autosampler of the HP1100 for in-tube extraction, this commercial equipment is employed for almost every recently published work.

In this paper, we report a systematic error in the quantitation of the extracted analyte when the commonly employed configuration is used for the automated in-tube SPME using the HP1100 system. It is important to emphasize that the reported systematic error is inherent exclusively to this configuration and procedure widely employed for automated in-tube SPME analysis. To alleviate the systematic error involved with the commonly used configuration, we suggest a few precautionary steps that must be taken when using an autosampler modified for the application of automated in-tube SPME.

2. Experimental

2.1. Sample preparation

A stock solution of ranitidine was prepared by dissolving a Zantac 75 tablet (approximately 75 mg/tablet ranitidine, Warner-Lambert, USA) in 100 ml Milli-Q water by sonication. This stock solution was further diluted with 25 m*M* Tris–HCl (pH 8.5) to obtain desired concentration of ranitidine in this buffer solution.

2.2. Chromatographic conditions

A HP1100 (Agilent Technologies) liquid chromatograph equipped with Model a G1313A autosampler and an atmospheric pressure electrospray ionization mass spectrometric detector was used for analysis of ranitidine. A Zorbax SB-CN column (150 mm×0.46 mm I.D., 5.0 μm particle size) was used. A mobile phase containing methanol–2-propanol–5 m*M* ammonium acetate (50:49:1)–water (62:38, v/v) was used for isocratic elution. A flow-rate of 0.5 ml/min with a run time of 15.0 min was employed. Electrospray ionization mass spectrometric conditions employed for the detection of ranitidine were as follows: ionization mode: positive; mass scan range: 100 to 400 u; selected ion monitoring (SIM), *m*/*z* 270, 315 and 337, dwell times of ions in SIM was 132 ms; fragmentor voltage, 70 V; capillary voltage, 3500 V; nebulizer and drying gas used was nitrogen; nebulizer pressure at 40 p.s.i. (1 p.s.i.=6894.76 Pa) drying gas flow-rate of 10 l/min at 350 °C . Consistent with the study reported by Kataoka et al. all three ions (*m*/*z* 270, 315 and 337) were extracted for quantification of ranitidine [12].

2.3. Automated in-tube SPME

The autosampler of the HP1100 LC system was modified for automated in-tube SPME. The system diagram for in-tube SPME is illustrated in Fig. 1A–C. Between the autosampler needle assembly and the 100 μl stainless steel sample loop, an extraction capillary (Omegawax-250, 60 cm×0.25 mm I.D., 0.25 μm film thickness, purchased from Supelco, USA or stainless steel capillary, 60 cm×0.25 mm I.D.) coupled with a 90 cm×0.25 mm I.D. uncoated fused-silica capillary was inserted. Only for studies involving quantitation of the amount of analyte in the extraction (i.e., extraction capillary and the needle assembly) and pre-extraction segment of the system (i.e., sample loop, metering pump and uncoated fused-silica capillary, hatched lines in Fig. 1), a two-position switching valve (valve 1) was incorporated between these segments of the in-tube SPME configuration. The details on the procedure for the determination of analyte in these segments will be discussed in the following section. An additional two-position switching valve (valve 2) was incorporated between the analytical column, injection valve (valve 3) and valve 1. Valve 2 was synchronized with valves 1 and 3 such that these valves together would

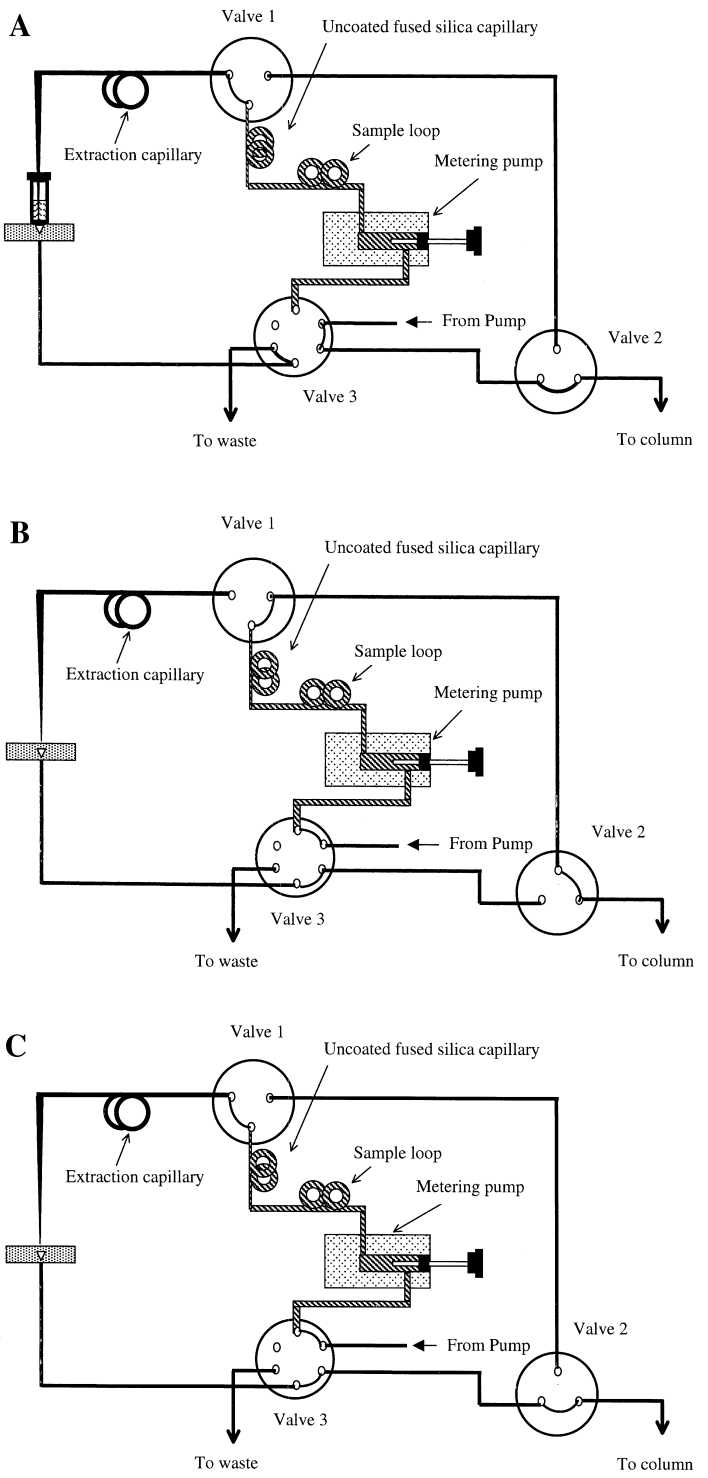

Fig. 1. Configuration of automated in-tube SPME using an HP1100 autosampler (A) during extraction; and (B) after extraction: desorption from pre-extraction segment after completion of extraction step (C) after extraction: desorption from extraction segment after completion of extraction step.

serve to divert the mobile phase flow between the pre-extraction and extraction segments (see Fig. 1B and C). For other part of the work, however, the commonly adopted configuration (i.e., without incorporating two-position switching valve) as reported in the literature, was used.

Extraction parameters such as number of draw/eject cycles and data acquisition were controlled by Chemstation software (Agilent Technologies). Prior to extraction, two aspirate/dispense cycles of 40 μl methanol and mobile phase were performed to condition the extraction medium. Extractions of ranitidine were conducted from 1 ml aliquots of 50 ng/ml ranitidine solutions taken in 2 ml autosampler vials. The total internal volume of a 60 cm×0.25 mm I.D. capillary is 29.4 μl and the volume of the needle assembly is approximately 10 μl. Thus, the total volume of the extraction capillary and the needle assembly is approximately 40 μl. Therefore, for the first step of extraction, 40 μl of the sample was drawn from a sample vial containing ranitidine solution. This volume is sufficient to allow the sample to be in maximum contact with the coated stationary phase of the capillary. In the next step, 30 μl was dispensed back to the same vial from which the sample was drawn. Subsequently, 30 μl of the sample was drawn and ejected back to the vial numerous times. In the final step, 40 μl of the sample was ejected back to the vial. In this way, the sample is completely removed from the capillary and the needle assembly. The extraction was conducted at a flow-rate of 100 μl/min when the injection valve is in the LOAD position (see Fig. 1A). After washing the injection needle with 10 μl of methanol, analyte was desorbed from the extraction capillary by diverting the mobile phase flow to the column. Each determination was conducted in triplicate and the average of the peak area of ranitidine was reported.

3. Results and discussion

To demonstrate the systematic error involved with the widely used approach for the automated in-tube extraction, work similar to that reported by Kataoka et al. was reproduced as a representative example to the current practice employed for automated in-tube SPME [12]. The choice of the representative example was based on the simplicity of the analytical procedure. Briefly, the published approach involved extraction of ranitidine in 25 m*M* Tris–HCl (pH 8.5) using a 60 cm×0.25 mm I.D., 0.25 μm film thickness Omegawax-250 capillary using the HP1100 LC system. Extracted ranitidine was eluted using a mobile phase containing methanol–2-propanol–5 m*M* ammonium acetate (50:50:1) at a flow-rate of 0.5 ml/min on a Supelcosil LC-CN column (3.3 cm×4.6 mm I.D., 3 μm particle size). Ranitidine was detected using electrospray ionization (ESI) MS.

While most extraction conditions such as pH and concentration of the buffer solution and the extraction capillary (Omegawax-250) used in this study were similar to that reported in the literature, only chromatographic conditions were different in this study. Obviously, use of different chromatographic conditions should not have any influence on the extraction efficiency of ranitidine.

In the preliminary experiments, extraction of 50 ng/ml ranitidine in 25 m*M* Tris–HCl (pH 8.5) was carried out on a 60 cm×0.25 mm I.D., 0.25 μm film thickness Omegawax-250 column by using multiple draw/eject cycles of 30 μl solution at the aspiration/dispense speed of 100 μl/min. Fig. 2A shows the extraction profile of ranitidine on the Omegawax-250 capillary. An increase in the peak area of ranitidine with increasing number of draw/eject cycles was observed. This observation is consistent with that reported by Katoaka et al. [12] leading to the conclusion that it was possible to reproduce the extraction profile of ranitidine even if the different chromatographic conditions were employed in this study. Surprisingly, a similar extraction profile was observed for ranitidine when a 60 cm×0.25 mm I.D. stainless steel capillary was used as an extraction medium (see Fig. 2B). Because stainless steel is a rather inert material, it should not be able extract ranitidine similar to that extracted by Omegawax-250. Ironically, from the extraction profile observed with stainless steel capillary, it appears that stainless steel follows a partitioning mechanism similar to a coated capillary. There has been a report that stainless steel does adsorb analyte to a certain extent during extraction by automated in-tube SPME when similar instrumentation is employed [11]. To eliminate adsorption of the analyte on the stainless

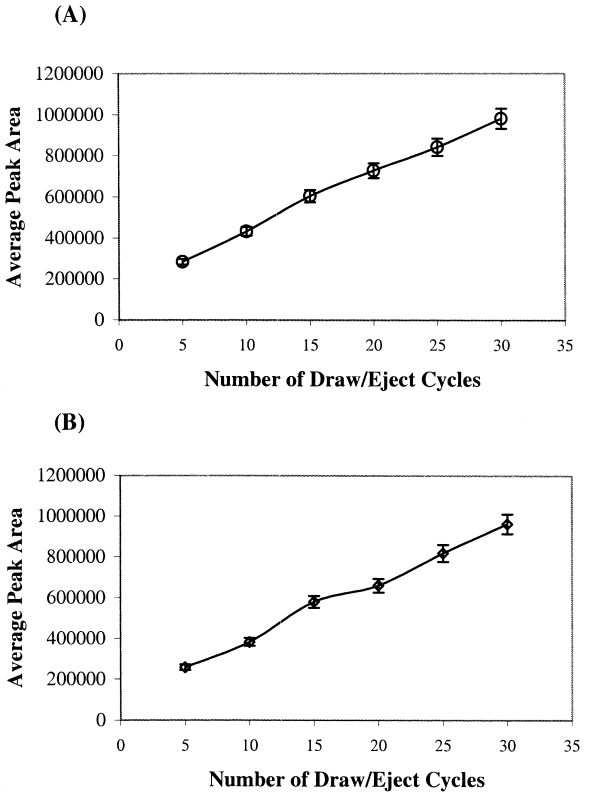

Fig. 2. Comparison of extraction profiles on (A) Omegawax-250 and (B) stainless steel capillaries. Concentration of ranitidine=50 ng/ml.

steel surface, it was recommended to connect a 90 cm polar deactivated uncoated fused-silica capillary between the extraction capillary and the stainless steel injection loop to prevent the sample loop from becoming contaminated with the analyte during extraction. Accordingly, in this study, a piece of fused-silica capillary was incorporated between the stainless steel injection loop and the extraction capillary (either Omegawax-250 or stainless steel capillary). In any case, because stainless steel capillary lacks a coated stationary phase, it should not exhibit a partitioning mechanism similar to the one demonstrated by Omegawax-250. When the concentration of ranitidine in 25 m*M* Tris–HCl was varied in the range of 20 to 200 ng/ml and extracted on the stainless steel capillary using 10 draw/eject cycles at the draw/eject speed of 100 μl/min, it showed a linear increase in peak area (R^2=9963). Once again, being an inert material, the stainless steel capillary should not be able to retain an increasing amount of ranitidine since it does not possess a stationary phase coating to allow retention of increasing amounts of analyte. The linear increase in the amount of ranitidine on the stainless steel surface could not be attributed to adsorption since stainless steel may allow adsorption only to a minimal degree. There has also been a report of sample/mobile phase mixing with the mobile phase when the mobile phase contained in autosampler tubing prior to the extraction capillary comes in contact with the analyte during extraction, when a similar configuration of automated in-tube SPME is employed [13]. However, no extensive study of the influence of sample/mobile phase mixing on the quantitation has been documented in the literature. Accordingly, therefore, a linear increase in the peak area of ranitidine on stainless steel capillary could possibly be due to an increase in the concentration of analyte as a result of sample/mobile phase mixing with the residual mobile phase in the pre-extraction and/or in the extraction segment. In this work, the unusual extraction profile demonstrated by stainless steel is attributed to the sample/mobile phase mixing, and we present a set of data leading to this conclusion.

During extraction, the six-port injection valve is in the LOAD position. However, there is residual mobile phase contained in both the pre-extraction and the extraction segment. When the sample is drawn, traces of analyte migrate to these segments of the autosampler causing contamination of the mobile phase residing inside the segments. In the subsequent steps, the concentration of analyte cumulatively increases as a result of sample/mobile phase mixing as the number of draw/eject steps is increased. To eliminate sample/mobile phase mixing, Mullett et al. incorporated a short rapid wash step immediately after extraction step by pumping a solvent of lower strength through the extraction segment using a second switching valve [13]. However, this approach needed optimization of solvent strength such that the loss of analyte absorbed by the extraction capillary could be minimized while effectively removing any contaminant resulting from sample/mobile phase mixing. While it was demonstrated in the literature that a wash step eliminates contamination due to

sample/mobile phase mixing, the exact point of contamination was not located. In this study, we investigate the source of error when the autosampler is converted to an automated in-tube SPME system, and show how this error could affect the quantitative results.

To identify the location of potential sample/mobile phase mixing, we incorporated two-position switching valves between the stainless steel sample injection loop connected to a piece of 90 cm uncoated fused-silica capillary and the Omegawax-250 extraction capillary, as shown in Fig. 1. A 50 ng/ml ranitidine solution in 25 m*M* Tris–HCl was extracted in triplicate from each fresh aliquot of ranitidine solution on this capillary with 20 draw/eject cycles, and it produced an average peak area of 727 970. Extraction was repeated with fresh aliquots of 1 ml ranitidine solution at 50 ng/ml. Immediately after the completion of the extraction step, the two-position switching valves 1 and 2 were directed to the pre-extraction segment where any analyte contained in this segment is carried to the analytical column (see Fig. 1B). Any analyte detected from the pre-extraction segment corresponds to an extraneous amount of analyte as a result of systematic error. This experiment was repeated three times and the average of peak area was taken into account. The average peak area of ranitidine in the pre-extraction segment was 472 993, which corresponds to approximately 65% (i.e., 100·472 993/727 970) relative to the average total peak area when both the segments are combined.

After desorption of analytes from the pre-extraction segment, the two-position switching valves were changed to their original positions to allow mobile phase to carry any analyte present in the extraction segment to the analytical column (see Fig. 1C). The average peak area produced in this segment was 173 967, which corresponds to approximately 24% (i.e., 100·173 967/727 970) relative to the total average peak area when both segments are included, as discussed above. When compared, these total peak areas were in close agreement (89% recovery) with the average peak areas of rantidine when the extraction was conducted (i.e., the normal mode of extraction without the use of two-position switching valves 1 and 2). No carry over was detected in any of these segments once the mobile phase was pumped through these segments. Fig. 3 shows chromatograms of ranitidine observed in pre-extraction and extraction segments when extracted using the Omegawax-250 capillary. For the purpose of comparison, a chromatogram of ranitidine extracted with Omegawax-250 without bifurcation of the mobile phase flow into these segments is also shown in this figure. When the number of draw/eject cycles was varied, the average peak area of ranitidine in the pre-extraction segment increased with the number of cycles (see Fig. 4A). For all draw/eject cycles, more than 85% of the analyte was accounted for, as determined by comparing the average peak area obtained when both the segments are combined. This indicates that there was less than 15% loss of analyte somewhere other than pre-extraction and extraction segment. One possible location where the loss of analyte could occur is the channels of switching valve 3, where mobile phase may not be able to reach to carry the trapped analyte to the analytical column. No easy practical way to prove this loss of analyte was found.

The extraction was repeated with the stainless steel capillary. A profile similar to that shown by the Omegawax-250 capillary was observed with stainless steel capillary when ranitidine was analyzed in both pre-extraction and extraction segments. In addition, an increase in average peak area of ranitidine in the extraction segment of the stainless steel capillary was noticed with increasing number of draw/eject cycles. Before the extraction process begins, there is still residual mobile phase contained in the extraction segment. This residual mobile phase is sufficient to allow accumulation of analyte when the number of draw/eject cycle increases, thus causing the observed increase in the amount of ranitidine in the extraction segment of the stainless steel capillary. It can be concluded from these observations that the analyte keeps accumulating proportionately in these segments with the increasing number of draw/eject cycles. When the extraction capillary has selectivity towards the analyte, the coated stationary phase allows partitioning between the stationary phase and the sample solution. Any increasing amount of analyte beyond a minimal amount observed in the extraction segment with increasing number of draw/eject cycles on the surface of an inert material such as a stainless steel capillary could be attributed to the

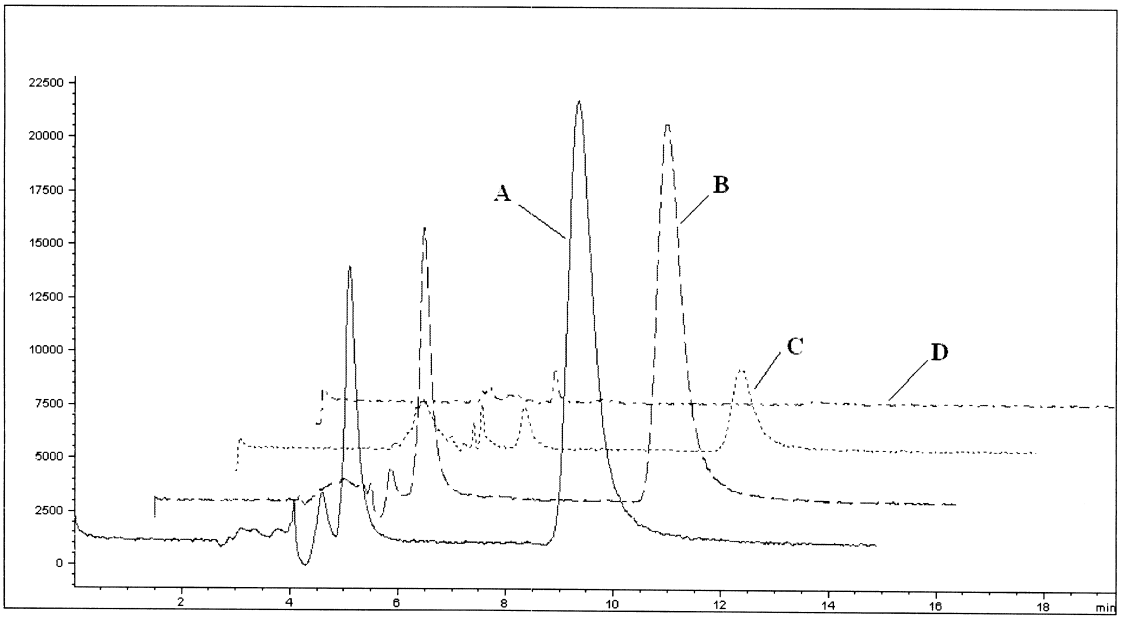

Fig. 3. Ranitidine observed (A) in the pre-extraction segment and extraction segments combined together, (B) in the pre-extraction segment; (C) in the extraction segment, and (D) Tris–HCl blank using the Omegawax-250 capillary. Concentration of ranitidine=50 ng/ml.

accumulation of analyte allowed by the residual mobile phase in the extraction segment.

The accumulated analyte in these segments is not carried to the column until the injection valve switches to the INJECT position, and thus any analytical signal generated is due to a combination of analyte extracted by the extraction capillary due to the partitioning mechanism, and the analyte accumulated in the extraction and pre-extraction segments due to sample/mobile phase mixing. This conclusion is consistent with the observed extraction profile shown by the stainless steel capillary, indicating that this chemically inert capillary does not demonstrate partitioning. In other words, the extraction profile observed with the stainless steel capillary is attributed to the increase in concentration of analyte due to its accumulation in the extraction and pre-extraction segments when the number of draw/eject cycles is increased.

In order to facilitate modification of an existing HP1100 autosampler into an automated in-tube SPME without a need of additional hardware, introduction of an air plug drawn from the headspace of the sample vial prior to the beginning of draw/eject cycles was attempted to circumvent sample/mobile phase mixing. An air plug between any residual mobile phase in the pre-extraction segment and the extraction segment would prevent the sample from coming in contact with the mobile phase where sample/mobile phase mixing results. Because the injection loop volume is 100 μl, an air plug of this volume may be sufficient to effectively minimize sample/mobile phase mixing. To demonstrate this approach, after conditioning the extraction capillary with methanol and mobile phase, 100 μl of air from the headspace of the sample vial is drawn followed by multiple draw/eject cycles of a 50 ng/ml ranitidine solution on the stainless steel capillary. At the end of the extraction process, the two-way positioning valves were switched such as to determine the amount of analyte in the pre-extraction segment. As noted in Fig. 5 (chromatogram C), no ranitidine was detected in the pre-extraction segment under these conditions, indicating that sample/mobile phase mixing could be substantially minimized when an air plug was inserted before beginning an extraction. On the other hand, a very minimal amount of ranitidine was detected in the extraction segment when an air plug was inserted. The average peak area of ranitidine in the pre-extraction segment

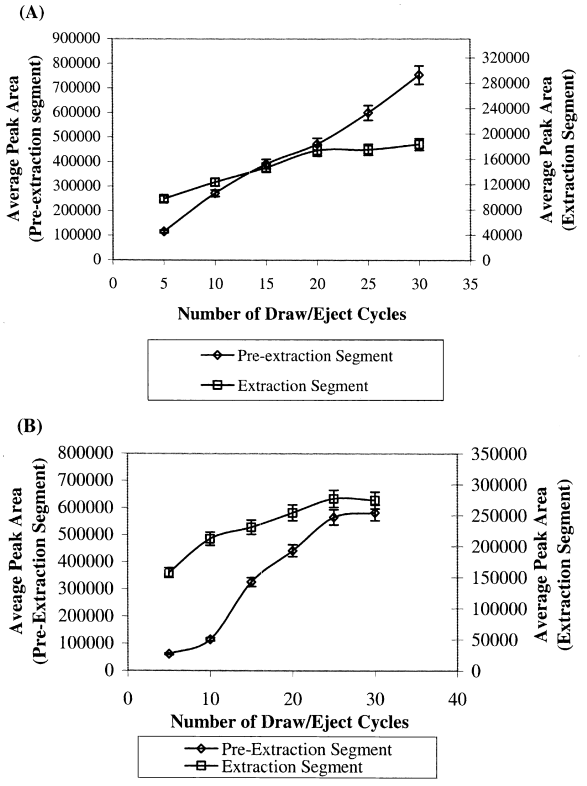

Fig. 4. Profiles of ranitidine found in pre-extraction and extraction segments when extracted with (A) Omegawax-250, and (B) stainless steel capillaries. Concentration of ranitidine=50 ng/ml.

at 20 draw/eject cycles was only 29 483 counts in the presence of the air plug, as opposed to its average peak area 288 523 in this segment when no air plug was inserted.

With the incorporation of an air plug prior to the extraction step, an extraction profile of ranitidine on the stainless steel capillary was constructed (see Fig. 6) using 50 ng/ml ranitidine solutions. On comparison of this profile with the one shown in Fig. 2B, it becomes evident that there is no increase in the amount of ranitidine extracted on the stainless steel capillary when the air plug is inserted prior to the beginning of extraction, hence the stainless steel capillary does not demonstrate a partitioning mechanism, as would be expected. When this extraction profile was constructed for Omegawax-250 with the insertion of an air plug, an increase in the extracted amount of ranitidine was observed with the increase in the number of draw/eject cycles, as seen in Fig. 6. Note that the peak areas of extracted ranitidine in the extraction segment of Omegawax-250 capillary when an air plug is inserted are 3–4-fold lower than those produced in the extraction segment without the insertion of an air plug. In addition, these peak areas are about 9–20-times lower than the average peak area produced when the two segments are combined together and no air plug is inserted, as is normally done in in-tube SPME using the commonly adopted system configuration. For example, the average peak area produced at 20 draw/eject cycles of 50 ng/ml ranitidine solution on Omegawax-250 capillary was 727 970 when the extraction is conducted following the widely adopted configuration for in-tube SPME. With the insertion of an air plug, an average peak area of only 46 497 at 20 draw/eject cycles was observed. It can be concluded from these data that incorporation of an air plug prior to the extraction step greatly minimizes systematic error resulting from sample/mobile phase mixing in the extraction and pre-extraction segments of the autosampler. Furthermore, the systematic error resulting from the sample/mobile phase mixing in the two segments increases proportionately with higher number of draw/eject cycles. For example, a ninefold increase in the average peak area due to system error at five extraction cycles was found whereas a 20-fold increase in the average peak area at 30 draw/eject cycles was observed. Because the accumulation of analyte in these segments contributes to the total amount of analyte detected, it would lead to significant overestimation in quantitation if this error is not taken into account.

4. Conclusion

Under the widely employed configuration for automated in-tube SPME, the amount of analyte observed is a combination of the amount of analyte that originates from three sources: (i) analyte accumulated in the the pre-extraction due to sample/mobile phase mixing with the residual mobile phase in this segment, (ii) analyte accumulated in the the extraction segment due to sample/mobile phase mixing with the residual mobile phase in this segment, and (iii) analyte extracted by the coated capillary via a partition mechanism. In this study, the

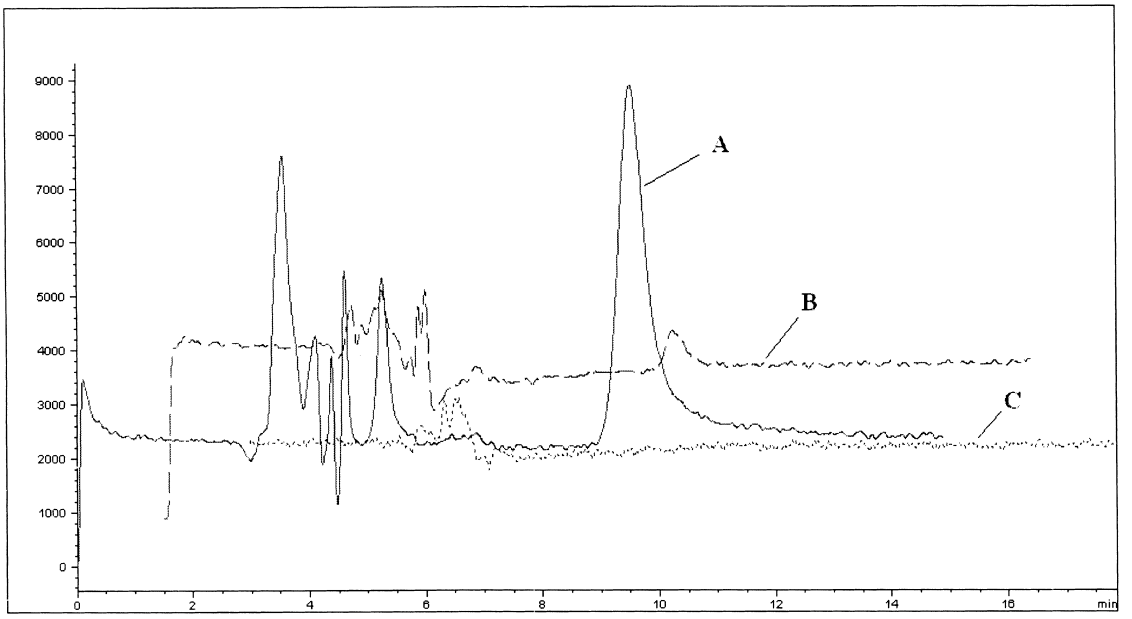

Fig. 5. Comparison of chromatograms of ranitidine in the extraction segment of stainless steel capillary (A) without air plug insertion; (B) with air plug insertion, and (C) in the pre-extraction segment with air plug. Concentration of ranitidine=50 ng/ml.

systematic error in automated in-tube SPME is shown to be generated from the extraction and the pre-extraction segment of the autosampler where analyte is concentrated as a result of cross-contamination with the residual mobile phase. Consequently, an overestimation as much as 20-fold in the quantitative results could be encountered. In addition, this error would lead to inaccurate information on the limit of detection and limit of quantitation, when automated in-tube SPME is employed as an approach for sample enrichment. If the extraction phase has a significantly higher affinity towards the analytes, then the systematic error would be expected to be much lower in magnitude because lesser amount of analyte would be migrating to the residual mobile phase in the pre-extraction and extraction segments. However, it should be proven in such cases that there is no accumulation of analytes in these segments due to sample/mobile phase mixing.

Finally, insertion of an air plug dramatically reduces the amount of analyte in both the pre-extraction and extraction segments of the autosampler by preventing the sample from coming in contact with the mobile phase.

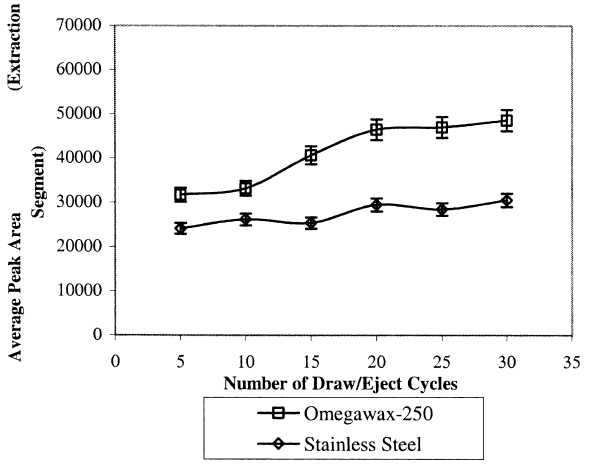

Fig. 6. Extraction profile of 50 ng/ml ranitidine extracted in the presence of air plug with Omegawax-250 and stainless steel capillaries.

References

[1] M. Gilar, E.S.P. Bouvier, B.J. Compton, J. Chromatogr. A 909 (2001) 111.
[2] J. Wu, J. Pawliszyn, Anal. Chem. 73 (2001) 55.

[3] J. Wu, Z. Mester, J. Pawliszyn, J. Anal. At. Spectrom. 16 (2001) 159.
[4] W.M. Mullett, P. Martin, J. Pawliszyn, Anal. Chem. 73 (2001) 2383.
[5] J. Wu, H.L. Lord, J. Pawliszyn, H. Kataoka, J. Microcol. Sep. 12 (2000) 255.
[6] H. Katoaka, M. Ise, S. Narimastu, J. Sep. Sci. 25 (2002) 77.
[7] J. Wu, H. Lord, J. Pawliszyn, Talanta 54 (2001) 655.
[8] H. Kataoka, H.L. Lord, J. Pawliszyn, Chromatography 20 (1999) 142.
[9] M. Walles, W. Mullett, K. Levsen, J. Borlak, J. Pawliszyn, J. Pharm. Biomed. Anal. 30 (2002) 307.
[10] H. Katakoa, Anal. Bioanal. Chem. 373 (2002) 31.
[11] Y. Guo, C. Tragas, H.L. Lord, J. Pawliszyn, J. Microcol. Sep. 12 (2000) 125.
[12] H. Kataoka, H.L. Lord, J. Pawliszyn, J. Chromatogr. B 731 (1999) 353.
[13] W.M. Mullett, K. Levsen, D. Lubda, J. Pawliszyn, J. Chromatogr. A 963 (2002) 325.