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Analytical characteristics of sample evaporation with the micro-Kuderna–Danish concentrator

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Abstract

The analytical behaviour of dichloromethane, pentane, and hexane solutions was studied through their evaporation in a micro-Kuderna–Danish concentrator. Initial sample concentration, final sample volume, matrix composition, system geometry and temperature gradients influence the process greatly. In general, solute loss increases exponentially when the final volume decreases. During the concentration process with very dilute solutions, the loss of solutes can be higher than 50% if the final volume reached is lower than 200 μl. This means a decrease in the quantification of the process, even though it is seemingly kept under control (low R.S.D.). The solute losses are not just due to the decrease in the number of plates during the reflux disappearance, but also to the probability that droplets of the evaporating sample are projected against clean and dry walls of low retention capacity. This hypothesis may explain the observed effects when concentrating more concentrated samples, or samples with a high content of some solutes. A more advantageous design for the micro-Kuderna–Danish receptor flask is also proposed. The replacement of the cylindrical flask by a flask with a conical bottom permits a decrease in the volume of losses and an improvement in the quantification of the process.

Introduction

When analysing volatile compounds at trace concentrations, it is common practice to transfer them from the starting matrix to an organic solvent, regardless of the sample characteristics and almost independently of the isolation method applied, with the exception of compounds isolated by automatic purge and trap or by automatic supercritical fluid extraction [1–4]. This organic phase must often be concentrated in order to achieve the required concentration for analysis. This is usually done by solvent evaporation within a suitable system and on very limited occasions is it possible or convenient to carry it out inside the chromatographic system [5,6]. During the development of gas chromatography, numerous concentration methods have been proposed, varying in purpose, applicability and ease of use. Classical methods when working with food matrices were proposed by Muller et al. [7], Ahrenst-Larsen and Hansen [8], Weurman [9], Blakesley and Loots [10], Guichard [11] and more recently by Boison and Tomlinson [12]. In environmental sample analysis and microanalysis, the work by Dünges [13], Grob et al. [14] and Grob and Muller [5] is particularly outstanding. Almost all the methods developed have more than satisfactory analytical characteristics. However, they must be performed in several

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steps with not readily available materials, and generally require experience and close attention to the process. In this context, the more widely used methods are concentration under a stream of nitrogen, with a Rotavapor and with a micro-Kuderna–Danish concentrator. The last approach is particularly attractive owing to its capability to concentrate relatively large volumes of sample in one step, requiring minimum attention. Among the most widely used procedures it is the only one that involves a fractional distillation. It is also the method proposed in the EPA 608 and 605 methods to determine priority pollutants, and it is used in virtually all kinds of analyses related to volatile compounds.

However, even though these methods are widely used, and are a necessary step in many analyses for trace compounds by gas chromatography, there have been hardly any studies evaluating the impact of this step of the analysis on the analytical quality of the final results. It is believed that each system must be separately studied as its behaviour cannot be extrapolated or related to any other. This situation makes it difficult to find general working rules, and to predict *a priori* the analytical behaviour of the processes. The results offered by some researchers on the recovery of solutes during the concentration steps are sometimes contradictory [5,12,15] and no conclusions except that concentration to dryness is problematic and may lead to important solute losses can be made. It is generally accepted that if the process is stopped before reaching dryness, the solute losses are reproducible and independent of the chemical nature of the solutes; therefore, they do not affect the reproducibility or accuracy of the process if the quantification is done by means of an adequate internal standard [14]. A closer look at the results reported in the literature will show that few researchers have taken precautions in checking if their sample concentration process is the most suitable or if it is dependent on the matrix composition, or to verify which percentage of the analytical variance is due to this aspect.

We are aware that each system has a particular behaviour as a consequence of its physico-chemical properties (Vapour pressure, solubility, azeotrope formation, deviation from ideal behaviour, etc.), and therefore it is difficult to develop a method that will suit all situations. Without pretending to give a global answer, this work is an attempt to understand the phenomena that take place during sample evaporation and to derive conclusions about the optimum evaporation conditions, with special emphasis on the micro-Kuderna–Danish concentrator.

2. Experimental

The standards were of analytical-reagent grade from Chemservice (West Chester, PA, USA). Standard solutions containing 200, 100, 40, 20, 10, 5, 2, 1, 0.8 and 0.08 mg l^{-1} of the following selected volatile compounds were prepared in dichloromethane or hexane: ethyl butyrate, 2-hexanone, ethyl benzene, *m*-xylene, 3-hexanol, ethyl caproate, 1-hexanol, 2-nonanone, ethyl caprylate, 1-heptanol, 1-octanol, ethyl decanoate, nerol, phenylethyl acetate and ethyl laurate.

Internal standard solutions of 2-ethylhexanol in ethanol (1 and 10 g l^{-1}) were prepared.

Dichloromethane, pentane, hexane, ethanol and 2-propanol were of analytical-reagent grade from Merck (Darmstadt, Germany). The first three were redistilled before use.

2.1. Concentration systems

A Vigreaux column (25 cm × 1.3 cm I.D.) was obtained from Afora (Barcelona, Spain). Other equipment consisted of a micro-Snyder column 158 mm long with three balls, a micro-condenser 112 mm long a 2-ml receiving vessel and a 40-ml flask from Supelco (Bellefonte, PA, USA). All joints were grease-free.

2.2. Chromatography

An HP 5890 Series II gas chromatograph (Hewlett-Packard) fitted with a Model 7673

automatic injector was used, with a split–splitless injection system and flame ionization detection. The signal was received and processed in a NEC computer equipped with the Maxima–Waters 820 program, version 3.0 (Waters Software). A Supelcowax 10 column (60 m × 0.32 mm I.D., film thickness 0.5 μm) was used.

2.3. Chromatographic conditions

With split injections, the conditions were as follows: carrier gas, hydrogen; head pressure, 50 kPa (1.5 ml min^{-1}); splitting ratio, 1 : 40; purge flow, 1.5 ml min^{-1}; injection temperature, 220°C; detector temperature, 220°C; initial column temperature, 50°C, held for 5 min and then raised at 3°C min^{-1} to 200°C. With splitless injection, the conditions were as follows: carrier gas, hydrogen; head pressure, 120 kPa (3.5 ml min^{-1}); split flow-rate 27 ml min^{-1}, splitless time, 3 min; purge flow, 1.5 ml min^{-1}; column temperature, 40°C, held for 5 min and then raised at 3°C min^{-1} to 200°C; make-up gas, nitrogen at 30 ml min^{-1}. The injection volume was 1 μl in both instances; a 5-μl Hamilton syringe was used.

Quantification with an internal standard (50 and 5 mg l^{-1} 2-ethylhexanol for split and splitless injection, respectively) was based on peak areas. The peak areas obtained from each analysis were interpolated on calibration graphs constructed with 20, 50, 100 and 200 mg l^{-1} standard solutions with split injection and 1, 2, 5 and 10 mg l^{-1} with splitless injection.

2.4. General procedure for concentration with the micro-Kuderna–Danish concentrator

The micro-Kuderna–Danish concentrator was filled, in all instances, with 30 ml of the solution to be concentrated. Two or three boiling chips no larger than 2 mm in diameter were added. The rectification column was conditioned with 2 ml of additional solvent before starting the concentration process. The concentrator was then immersed in a water-bath to the widest part of the flask body, then the evaporation was begun. When it was completed the apparatus was removed from the water-bath and immediately introduced into a water–ice bath where it was allowed to settle for 5 min. The liquid was recovered with a syringe and introduced into a 2-ml volumetric flask. The bottom of the micro-Kuderna–Danish concentrator was washed without agitation with three fractions of a volume similar to the recovered volume, and the washings were added to the volumetric flask. The internal standard was then added (10 μl) and the mixture was diluted to volume with solvent. The sample obtained was analysed by GC as indicated previously.

Study of the influence of the final volume reached and the initial sample concentration

Three sets (4, 0.8 and 0.08 mg l^{-1}) of 30 solutions in dichloromethane were prepared. Each solution was concentrated using a micro-Kuderna–Danish concentrator equipped with a Snyder column with three balls in a thermostated bath at 47°C. Six solutions from each set were concentrated to a volume of ca. 1 ml, another group of six to 0.5 ml (just before the reflux began to diminish), another group to 0.4 ml (reflux disappearance), another group to 0.2 ml (just before boiling stopped) and a last group to the point where the liquid phase disappeared at the bottom of the flask (final volume about 0.1 ml). The concentrated solutions were analysed as described previously.

Influence of the presence of third compounds

Fifteen sets of three solutions in dichloromethane containing 0.08 mg l^{-1} of the volatiles were prepared. The following compounds were added to the solutions of each set: set 1, 1 mg of ethanol; set 2, 2 mg of ethanol; set 3, 4 mg of ethanol; set 4, 6 mg of ethanol; set 5, 12 mg of ethanol; set 6, 25 mg of ethanol; set 7, 2 mg of 2-propanol; set 8, 4 mg of 2-propanol; set 9, 6 mg of 2-propanol; set 10, 12 mg of 2-propanol; set 11, 1 mg of isoamyl alcohol; set 12, 2 mg of isoamyl alcohol; set 13, 6 mg of isoamyl alcohol; set 14, 0.012 mg of heptanol and set 15, 0.12 mg of heptanol. The three solutions of each set were

concentrated to 0.1 ml and analysed as described previously.

In addition to sets 5, 10, 14 and 15, three more groups of solutions were prepared and concentrated to final volumes of 1, 0.4, and 0.2 ml.

Influence of the rectifying capacity of the column

Two other columns were used besides the Snyder three-ball column: a Vigreaux column and the standard micro-Kuderna–Danish column (microconcentrator). Sets of six concentrated and diluted solutions were prepared and concentrated in both systems as done previously, to a final volume of about 0.1 ml.

Influence of micro-Kuderna–Danish receiving flask geometry

A 2-ml flask with a conical bottom and of 1.5 cm I.D. was used to replace the cylindrical standard micro-Kuderna–Danish flask. Twelve solutions in dichloromethane containing 0.08 mg l^{-1} volatiles were concentrated in the system equipped with the new flask to final volumes of 0.2 and 0.1 ml. Also, 1-ml volume samples were directly concentrated to 0.1 ml in the new flask without fitting the rest of the microconcentrator.

Influence of the room temperature and the type of solvent containing the sample

Solutions in hexane and pentane with concentrations of volatiles of 4 and 0.08 mg l^{-1} were prepared: two sets, each of a different concentration, of six solutions in pentane were concentrated to ca. 0.1 ml in the standard system. The bath temperature was 41°C and room temperature was 24°C. Two other sets of solutions with the same characteristics were concentrated in a cold room at 15°C. In a similar manner, two sets of solutions in hexane were concentrated keeping the bath temperature at 87°C (below this temperature the reflux was not satisfactory). Two other sets of solutions in hexane were concentrated in a chamber at 45°C. On this occasion the bath temperature was kept at 76°C.

3. Results

3.1. Study of the influence of the final volume reached and the initial sample concentration (Figs. 1–4)

Losses increase exponentially with respect to the final volume, and the curve shape is a function of the starting concentration (Fig. 4). If the final volume is higher than 400 μl (concentration factor around 80), losses are kept at acceptable levels and are lower than those obtained with other systems which reach a similar concentration factor [5,13]. The results were not different on changing the initial sample concentration, as shown in Fig. 1a and b and in the first part of Fig. 4.

Below 400 μl the evaporation process depends to a great extent on the solute concentration of the media, as shown in Fig. 1a and b and 4. If the solution is somehow concentrated, solute losses are not very important until the volume approaches 100 μl (only data relating to 4.0 mg l^{-1} solutions are shown). If the solution is very dilute, the losses are substantial below 400 μl. Losses in this and for these solutions are much larger than those reported in the literature for other concentration systems. The absolute amounts of lost solute are hardly reproducible. The greater the losses, the more difficult is reproducibility. The corresponding data are shown in Fig. 2a and b. During the concentration of dilute solutions, the amount of lost matter is random when the concentration reaches final volumes lower than 0.4 ml (R.S.D. > 10% when the final volume is 0.4 ml and >20% when the final volume is 0.1 ml). On the other hand, the concentration of concentrated solutions was a much more repetitive process, the average R.S.D. being ca. 10% when the evaporation was stronger. However, the relative amounts of lost solutes are much more reproducible (see Fig. 3). It is important to point out that the low recoveries of solutes do not mean an important decrease in precision if the quantification is done with an internal standard which is also being concentrated.

The losses depend slightly on the boiling point

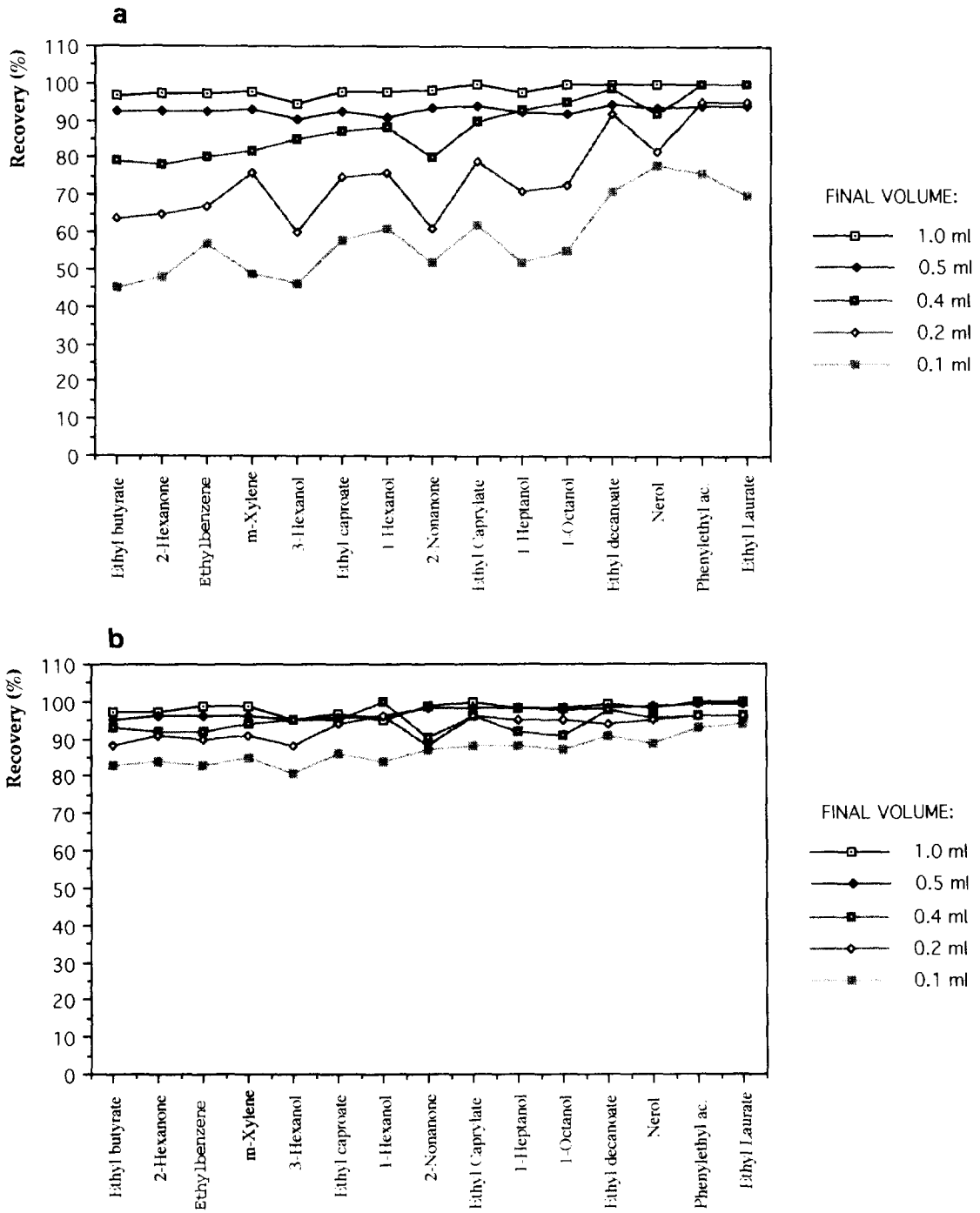

Fig. 1. Solute recovery throughout the sample concentration process: influence of final volume, (a) dilute solutions, 0.08 mg l^{-1}; (b) concentrated solutions, 4.0 mg l^{-1}.

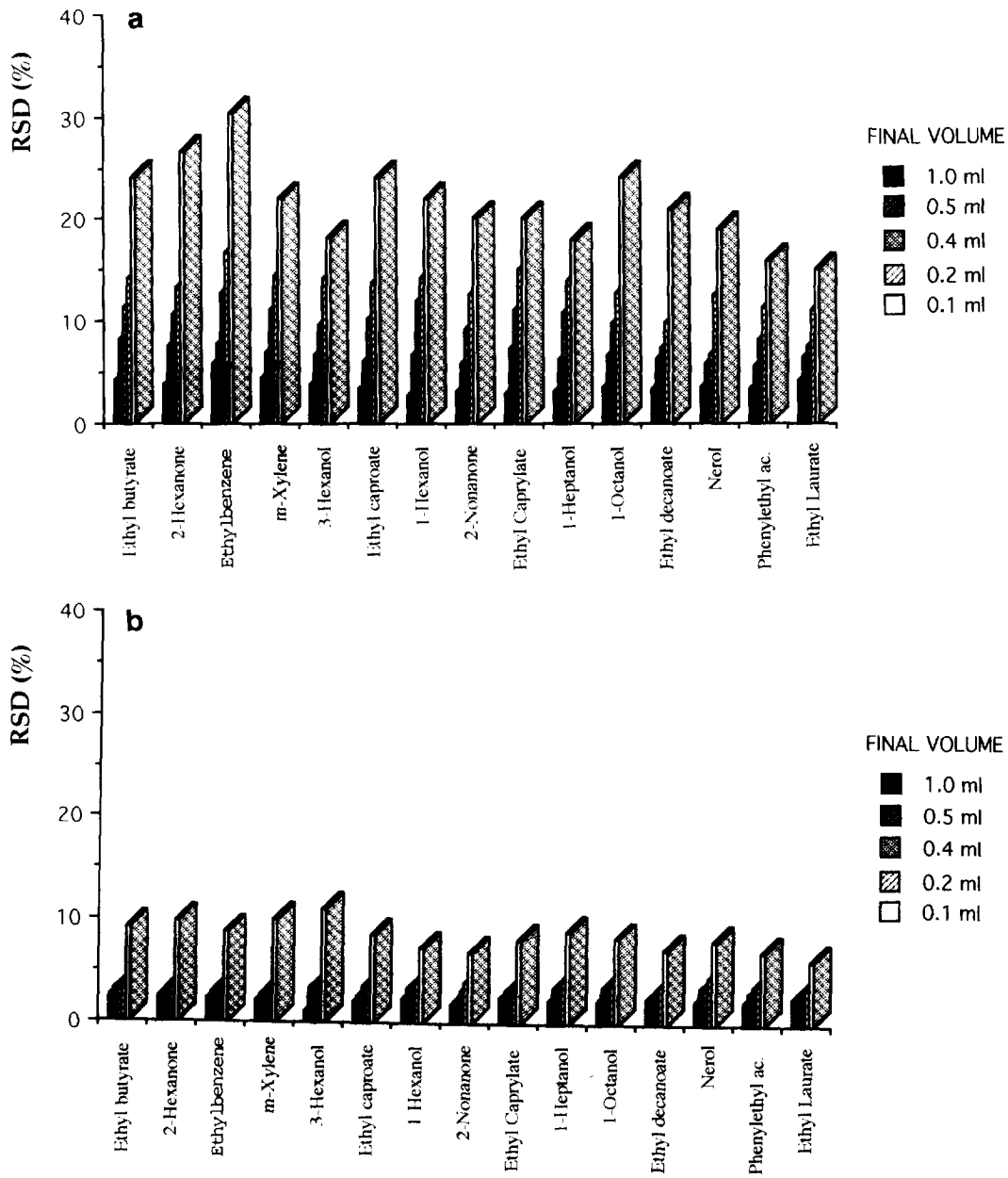

Fig. 2. Reproducibility of solute recovery throughout the sample concentration process as a function of final volume reached. (a) Concentration of 0.08 mg l^{-1}; (b) concentration 4.0 mg l^{-1}.

of the solutes: the higher the volatility, the higher the losses; however, relatively constant levels are maintained. Only when concentrating dilute solutions to 0.1 ml were the losses of the more volatile compounds almost double those observed for the less volatile compounds (see Fig. 1a). However, the differences were lower in the evaporation of the more concentrated solu-

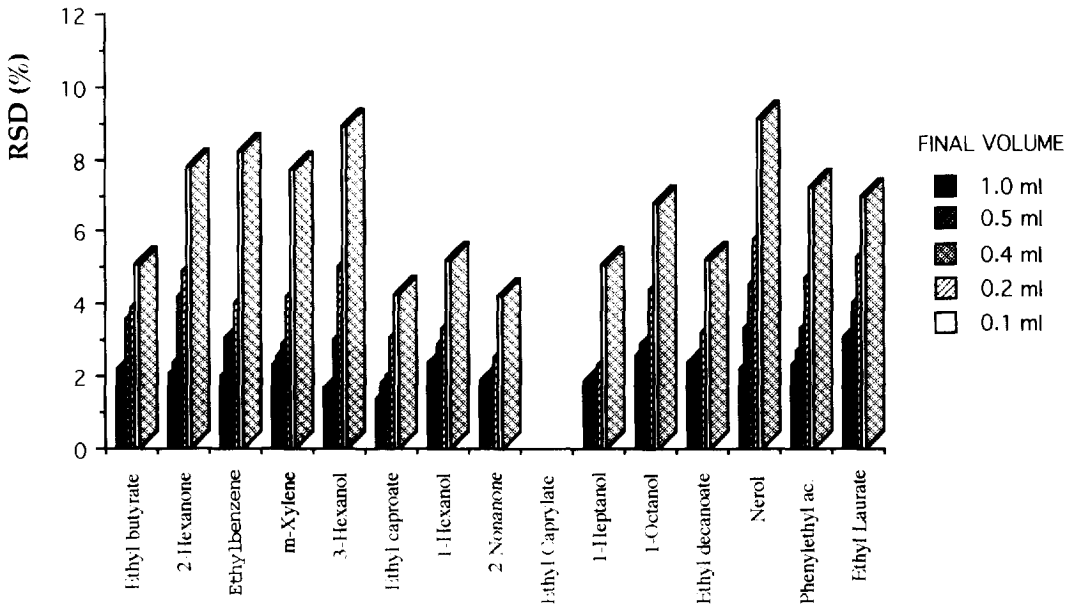

Fig. 3. Reproducibility of solute recovery relative to that of ethyl caprylate as a function of final volume reached. Concentration 0.08 mg l^{-1}.

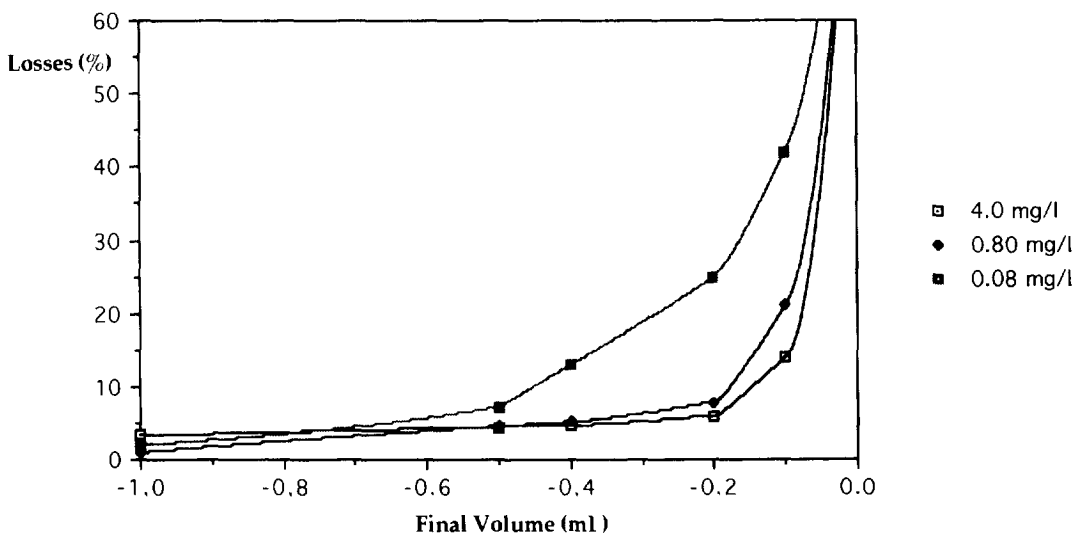

Fig. 4. Amount of ethyl caproate lost through the sample evaporation process as a function of both final volume reached and initial sample concentration.

tions (see Fig. 1b). The nature of the solute does not seem to affect the amount lost significantly.

3.2. Study of the effect of adding third compounds (Fig. 5a and b; Table 1)

After a certain critical amount, the addition of a third compound causes a severe decrease in the amount lost and a substantial improvement in the precision of the process. The critical amount of the compound to be added depends on its boiling point. The addition of four compounds was studied: ethanol, 2-propanol, isoamyl alcohol and heptanol. The minimum amounts needed to observe a significant improvement of the evaporation process were ca. 6 mg of ethanol, 4 mg of 2-propanol, 1 mg of isoamyl alcohol and 0.1 mg of heptanol. A dilute solution containing a sufficient amount of a third compound tends to behave like a concentrated solution.

3.3. Study of different rectification systems (Fig. 6)

The results obtained are diverse and are a function of the number of plates of the systems (Fig. 6). In all instances evaporation was forced almost to complete evaporation of the liquid phase, reaching a final volume of ca. 0.1 ml after its partial recondensation. In accordance with previous observations, the behaviour is strongly dependent on the initial concentration of the solution. In all instances the worst results were obtained with the microconcentrator, which is the system with the worse rectification capacity. During the concentration of more dilute solutions, some of the most volatile solutes were almost completely lost. The estimated losses of less volatile solutes were not very different from those observed with other systems. The results were slightly better for the Vigreaux column, but the losses were higher than those with the standard system.

3.4. Change in the geometry of the receptor system (Table 2)

The replacement of the cylindrical flask of the micro-Kuderna–Danish concentrator by a conical-bottomed flask lead to a great reduction in the losses during the concentration of the most diluted solutions. The results shown in Table 2 refer to strong concentration, almost to disappearance of the liquid phase. The precision of the process was improved, the average R.S.D. being ca. 7%. The behaviour of the system with the new flask is similar to that observed in the concentration of the more concentrated solutions.

3.5. Use of other solvents: pentane and hexane (Fig. 7a and b)

Fig. 7a and b show the results obtained in the concentration of solutions in pentane and hexane. The losses in both instances are higher than those when using dichloromethane as solvent. The differences found in the evaporation of the most concentrated solutions (Fig. 7a) are particularly important. The observed losses in the hexane solutions are the highest. When concentrating more dilute solutions, hexane solutions behave similarly to dichloromethane solutions, whereas the pentane solutions behave differently. The concentration of hexane solutions does not follow the same behaviour as observed so far. The losses hardly depend on the initial sample concentration under these conditions.

However, when the concentration system is kept in a chamber at 15°C for pentane or at 45°C for hexane, the results are similar to those obtained when concentrating dichloromethane solutions (data not shown). For both concentrated and dilute solutions, the best recoveries were obtained when working with pentane.

4. Discussion

Based on the results, we should consider two well differentiated steps. During the first step, an equilibrium is established between the evaporating solvent mass and the recondensing solvent mass, even though there is a net flux of solvent lost from the upper part of the column. The equipment works like a distillation system with several theoretical plates depending on the system geometry and the operating conditions. The

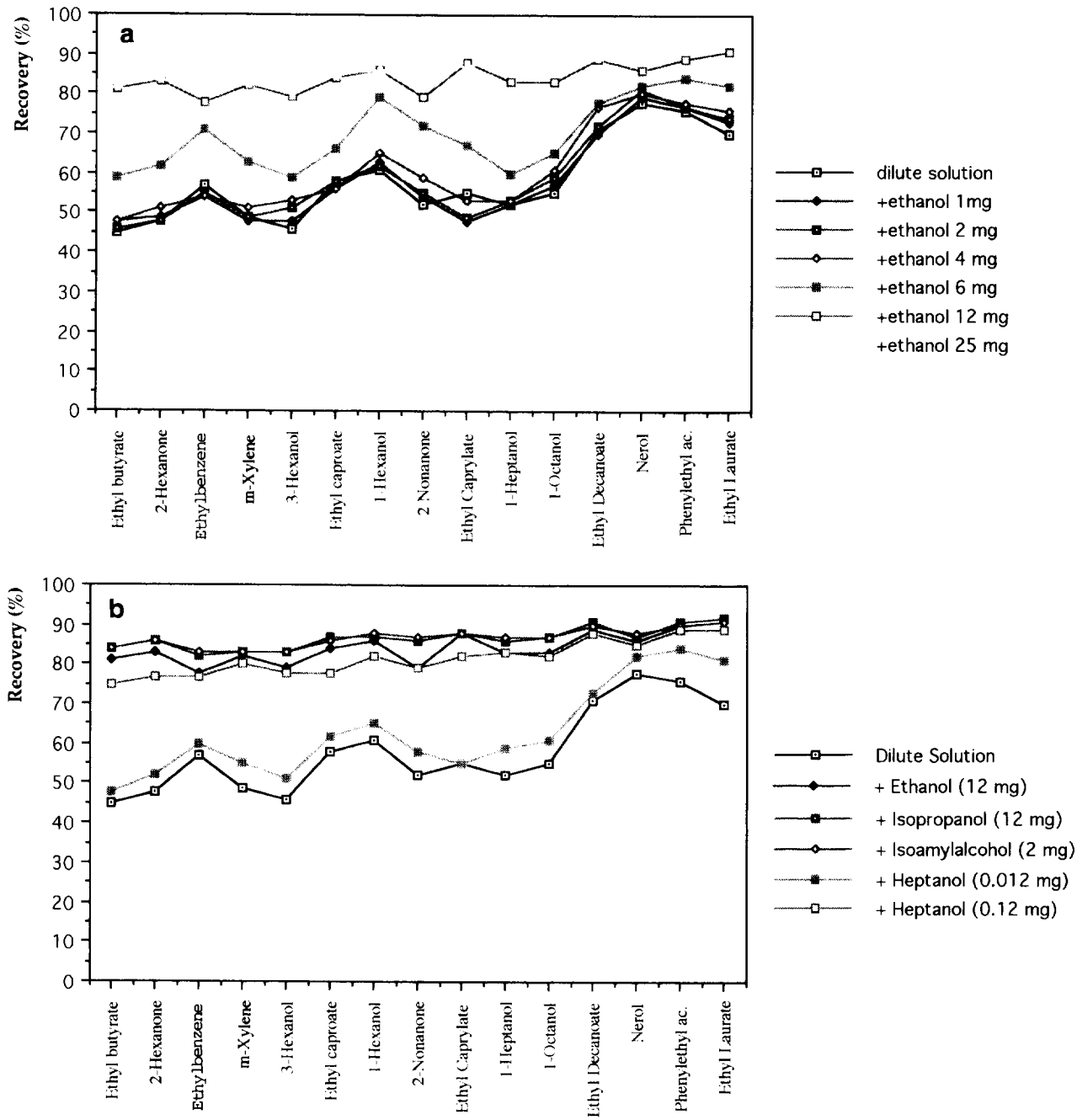

Fig. 5. Influence of the presence of a third party compound on solute recovery. (a) Study of amount of ethanol needed to achieve a significant decrease in solute losses. (b) Stabilization of matrix effect. The final point observed is independent of the added solute and tends to be similar to the evaporation process of a concentrated solution.

Table 1
Effect of addition of a third compound addition on the analytical behaviour of the evaporation process

Compound	R.S.D. (%)[a]				
	+ Ethanol (12 mg)	+ 2-Propanol (12 mg)	+ Isoamyl alcohol (6 mg)	+ Heptanol (0.012 mg)	+ Heptanol (0.12 mg)
Ethyl butyrate	14	3.1	3.4	18	11
2-Hexanone	13	3.0	3.1	14	10
Ethylbenzene	17	3.2	3.0	19	12
m-Xylene	14	2.8	3.5	21	9.1
3-Hexanol	14	3.8	3.7	18	8.5
Ethyl caproate	14	3.3	3.2	18	7.8
1-Hexanol	12	3.4	3.6	21	7.5
2-Nonanone	15	3.1	2.9	22	8.2
Ethyl caprylate	14	3.0	3.4	13	9.3
1-Heptanol	9	2.8	3.4	15	9.4
1-Octanol	11	2.9	3.0	16	8.6
Ethyl decanoate	8	4.4	4.2	11	7.2
Nerol	12	3.3	3.9	13	6.8
Phenylethyl acetate	11	3.5	3.8	14	8.0
Ethyl laurate	11	4.5	4.7	15	9.3

[a] $n = 3$.

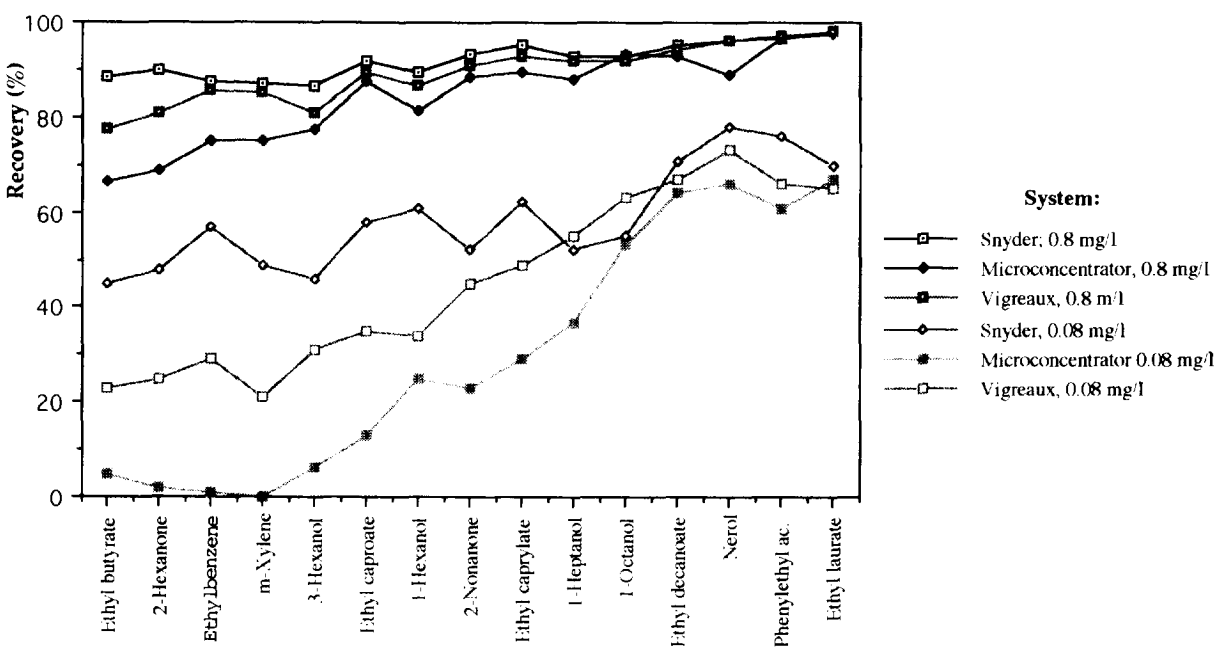

Fig. 6. Study of the influence of the rectifying system coupled to a micro-Kuderna–Danish concentrator for two different concentrations.

Table 2
Analytical results from concentration of dichloromethane solutions using a modified micro-Kuderna–Danish flask.

Compound	Recovery (%)			
	Cylindrical flask	Conical Flask	Absolute R.S.D. (%)[a]	Relative R.S.D. (%)[a]
Ethyl butyrate	45	72	9.8	5.7
2-Hexanone	48	78	8.3	7.2
Ethylbenzene	57	81	7.2	4.9
m-Xylene	49	83	8.7	5.2
3-Hexanol	46	85	7.9	5.0
Ethyl caproate	58	87	5.6	3.4
1-Hexanol	61	90	6.0	4.5
2-Nonanone	52	78	10.1	6.4
Ethyl caprylate	62	88	5.7	0.0
1-Heptanol	52	94	6.5	4.1
1-Octanol	55	91	5.6	3.9
Ethyl decanoate	71	96	4.8	2.0
Nerol	78	89	7.7	4.4
Phenylethyl acetate	76	96	5.1	3.5
Ethyl Laurate	70	97	4.3	2.7

Concentration until almost dryness. Initial concentration, 0.08 mg l^{-1}. Final volume, 100 μl.
[a] $n = 6$.

rectification capacity of the system is sufficient and the solute losses are almost non-existent if its boiling point is above 100°C. The behaviour of the micro-Kuderna–Danish concentrator from an analytical point of view is more than satisfactory during this step. The lower limit of this step is within the range 0.4–0.5 ml, which means a concentration factor of between 60 and 80. The lack of precision introduced in the analytical result by this step is also acceptable, being in the worse scenario ca. 7% if the quantification is done without adding an internal standard before concentration. However, if an internal standard is added to the solution before the evaporation process begins, then the lack of precision introduced by the process is close to 3%, which is negligible for most trace analyses.

The second step begins when the amount of evaporated solvent is not sufficient to maintain the reflux. The differentiating characteristics of this step are the strong dependence on the sample concentration, the important solute volume losses during the concentration of diluted solutions, the way in which these losses increase when the final volume reached is decreased and the strong lack of precision associated with high losses. From an analytical point of view, the control over the process is drastically lowered and, what is worse, this fact can even go unnoticed. Examination of the data in Fig. 3 may lead one to think that the lack of precision associated with the process of evaporation is not relevant and can be accepted, given the important improvement in sensitivity. However, a closer look at Fig. 1a and 2a shows that at least three internal standards should be used to avoid these effects, and even then the process would be subject to matrix effects.

These effects are not a direct consequence of a decrease in the efficiency of the system, but to the fact that the lack of solvent makes most of the area of the glass walls dry. A drop, e.g., from a splash, that is placed on a hot and dry surface will evaporate slowly (owing to low thermal transfer from the glass walls and to the high calorific capacity of the solvent [16,17]). First the solvent will be lost, while the solutes concentrate in a progressive way and will not be

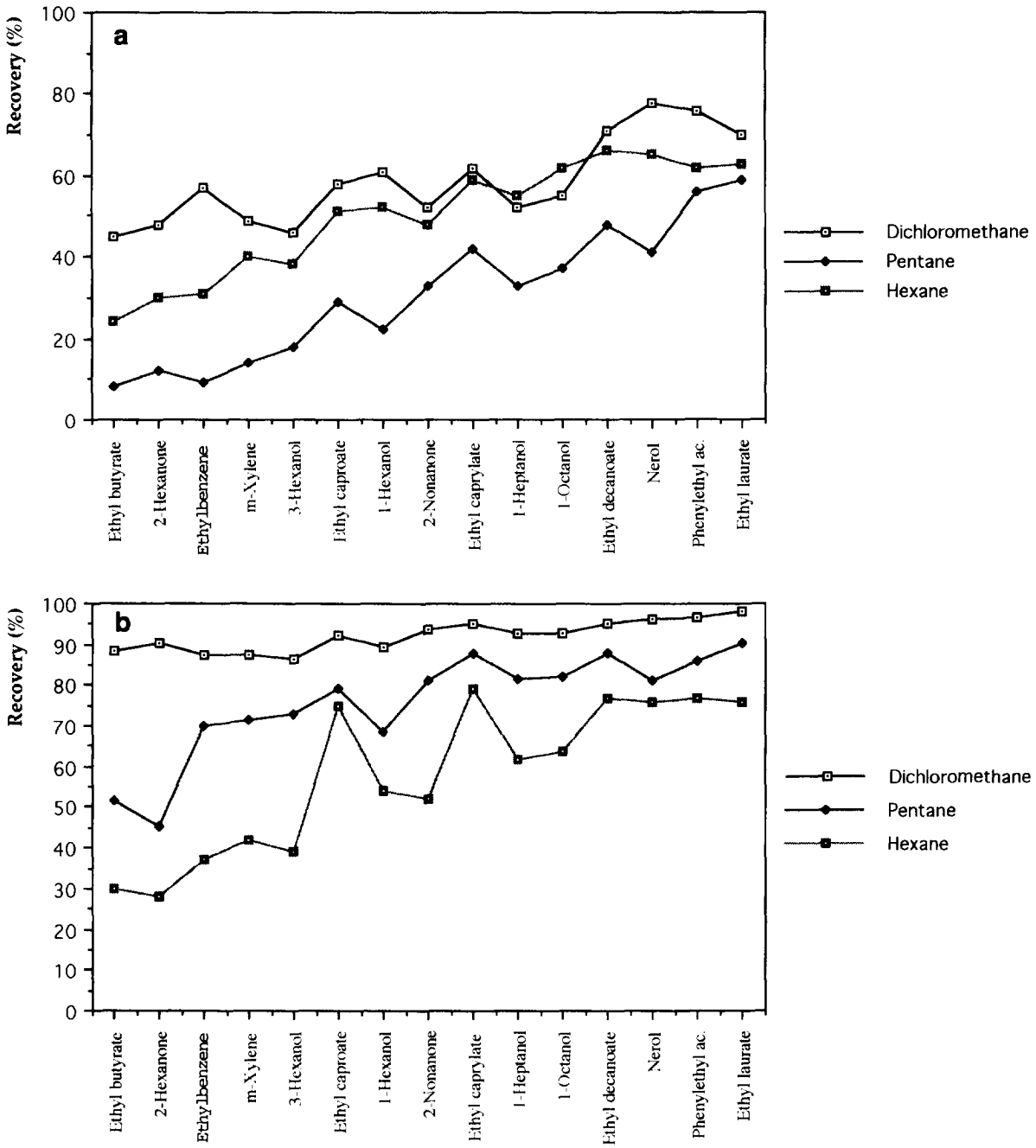

Fig. 7. Solute recovery as a function of type of solvent contained in the sample (a) Concentration 0.08 mg l^{-1}; (b) concentration 4.0 mg l^{-1}.

released until the drop of solvent that contained them has disappeared. Once it has disappeared, solutes are flushed by the solvent gas in a similar way to the evaporation of solutes in a gap of low retention surface, as has been thoroughly studied by Grob, Jr. [18, 19] and Grob [6,20].

If this previous statement is true, then it follows that the amount of lost solute depend on (1) the number of sample drop projections on dry walls and (2) the lifetime of those drops. Therefore,, all processes that contribute to increasing the number of splashes will contribute to increasing the amount of solute losses, and the shorter the life of the drop placed on the glass wall the greater is the probability of losing volatile matter. The validity of this hypothesis is discussed next based on the experimental results obtained.

4.1. Final volume dependence

Fig. 8 show two diagrams which represent the sample evaporation in the final steps of evaporation in the micro-Kuderna–Danish concentrator. They show how the violent sample boiling in the final steps of this process makes the surface constantly wet even when the volume of remaining liquid decreases drastically. According to our model, losses should be proportional to the wet surface area/remaining liquid volume ratio. In fact, this is the behaviour represented in Fig. 4. The existing parallelisms between evaporation in the cylindrical-bottomed flask and evaporation in the conical-bottomed flask are shown in Fig. 9. It can be seen that in this case the wet surface area/remaining liquid volume ratio is much lower, and this would explain the results with evaporation in this type of system which are shown in Table 2.

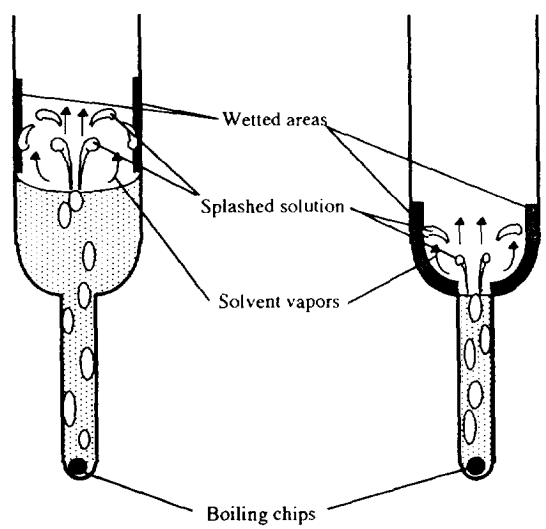

Fig. 8. Diagram of sample evaporation process in the final concentration step showing how the wet surface area/liquid volume ratio increases exponentially.

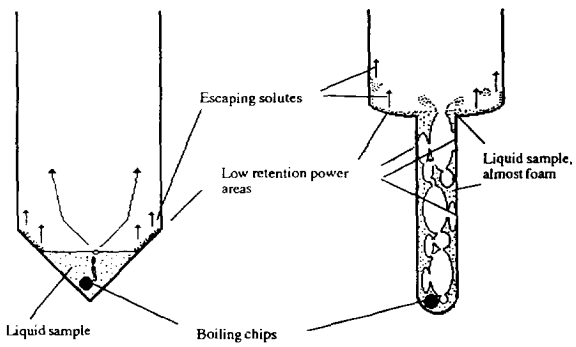

Fig. 9. Diagram of sample evaporation process in the final concentration step in two different systems. With a conical-bottomed flash the wet surface area/liquid volume ratio remains constant, whereas with a cylindrical-bottomed flash it increases exponentially. Consequently, the probability of a solute finding a low retention power area is much higher.

4.2. Effect of initial sample concentration

Studies on sample vaporization in gas chromatography with classical injectors have found that the presence of less volatile material can modulate the sample evaporation behaviour, prolonging the lifetime of solvent droplets that evaporate around a nucleus made of less volatile material [21–23]. Some of these solutes may play a similar role to non-volatile material in chromatographic injectors, adhering solvent droplets coming from splashes or recondensation. In fact, a simple observation of the behaviour of the evaporating solution shows that the size of droplets generated from solutions containing a larger amount of solutes is larger than the size of droplets generated during evaporation of dilute solutions, in spite of having a greater capacity to adhere to the glass wall and a lower mobility on it. As a result, the droplets take longer to evaporate, delaying the release of solutes contained in them, and diminishing the amount of

solute lost during this step. Once the majority of the solvent that forms the drop has evaporated, we have to consider that the thickness of the solute coating covering the glass wall makes the retention capacity of this type of stationary phase temporarily larger while avoiding solute loss, or at least delaying its release.

The addition of third components showed how important retention capacity of the pseudo-chromatographic system is. The boiling point of the compound added plays a key role in the intensity of the phenomena, as has already been mentioned. The concentration of the third component added needs to be progressively smaller as its volatility decreases in order to increase its action on the evaporation, which must be attributed to its higher capacity to form a temporary chromatographic coating. The practical importance of this observation is great, as it makes it possible to decrease the observed matrix effect and to cancel the error in quantification, and also to decrease the solute losses to acceptable levels for the concentration factors referred to here ($F_C > 400$). However, we must take into consideration that these "boiling regulator" compounds could form azeotropes with the solvents, which could lead to different results from those presented here, and should be considered with caution in each instance.

4.3. *Irreproducibility of the process*

This observation fits the proposed model. In fact, the number of projections is highly unpredictable and imprecise because it depends on uncontrolled factors such as the glass surface behaviour or the boiling of chips. Since solute losses are a direct consequence of these projections, the absolute solute losses are highly irreproducible. On the other hand, and according to the simplified model, the solutes contained in a droplet from a projection will be completely evaporated almost independently of their boiling point. This implies that losses will be constant from a relative point of view, and therefore reproducible.

4.4. *Concentration of solutions contained in other solvents*

The results with other solvents (see Fig. 7) are worse than those obtained with dichloromethane. With pentane under conditions used, almost no recondensation of pentane took place in the rectifying column, and evaporation occurred at high speed. In contrast, with hexane, solvent rectification at room temperature was so intense that an increase in the temperature of the thermostated bath to well above the solvent boiling point was needed. The higher losses in the case of poor refluxing are easily explained and must be attributed to a low process efficiency. Losses when the reflux is excessive are due to the large amount of energy needed to achieve a constant release of solvent at a reasonable speed from the rectifying system. This aggravates the coevaporation problems, as droplet lifetime decreases while their number increases as boiling becomes more violent. Experiments with both solvents were repeated at different external temperatures (15°C with pentane and 45°C with hexane). The behaviour of these systems was then very similar to that observed previously, supporting the hypothesis and demonstrating that there is an optimum concentration point for each rectification system depending on the boiling point of the solvent and its calorific capacity.

5. Conclusions

Micro-Kuderna–Danish concentration is a very suitable approach for the concentration of average sample volumes (10–40 ml) by solvent evaporation, providing the process with very good analytical characteristics of precision and accuracy.

Solute losses in systems that do not form azeotropes are mainly due to flushing of solute layers from dry surfaces (coevaporation) during the final sample evaporation step (below 0.5 ml). Therefore, special precautions are needed at this stage. Very dilute solutions should not be concen-

trated below that volume, but concentrated solutions or solutions containing a large amount of a third, miscible component can be concentrated below that limit. If the cyclindrical collector flask is replaced by a flask with a conical bottom, even dilute solutions can be concentrated below 100 μl without losing the analytical characteristics. The use of rectifying systems less efficient than the Snyder column leads to higher solute losses.

There are optimum operating conditions for each system. The temperature gradient between the reflux system and the ambient air around it should not be below 20°C (no reflux) or above 40°C (excessive risk of coevaporation).

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