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Influence of the capillary dimensions on the performance of the preconcentration technique based on parallel current chromatography

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Abstract

The influence of the inner capillary diameter, length of the capillary, linear velocity of the mobile phase and volume of the retentive liquid segment on the dynamic range of the gradient and the efficiency of enrichment of the new preconcentration technique was studied. Phenol, 2,4-dinitroaniline, phenanthrene and fluoranthene in water solution were used as model analytes. Capillaries with inner diameters of 0.095 mm and 0.2 mm and lengths ranging from 3.5 to 15 m were examined. The segment of the retentive liquid in capillary was generated by the injection of suitable volume of cyclohexanol. It was found that the enrichment increases with decrease in the inner capillary diameter and with increase in its length. The working range of this technique was found. With analytes in sub-ppb concentrations in the injected sample, an enrichment factor up to 280 was achieved. The application possibilities of proposed procedure was demonstrated by off-line capillary GC analysis of collected enriched solution of model mixtures of alkyl phthalates and polyaromates. © 1997 Elsevier Science B.V.

Keywords: Capillary dimensions; Parallel current chromatography; Preconcentration; Phenol; 2,4-Dinitroaniline; Phenanthrene; Fluoranthene

1. Introduction

Recently, the preconcentration technique based on the focusing of the analytes in the tail of the retentive layer moving in open capillary was introduced [1]. The film of the injected retentive liquid dissolves continuously in the stream of the mobile phase. In the region of the tail of the zone of retentive liquid, the conditions of gradient elution are generated. The process can be repeated many times in a single capillary without contamination which can be seen as an advantage in comparison with enrichment on the conventional solid-phase extraction (SPE) cartridges or SPE disks [2–5].

In parallel current open-tubular chromatography, (PC-OTLC) [6], the flow friction force keeps the thickness of the retentive film constant round the internal circumference of the capillary. The thickness of retentive liquid layer is dependent on the interfacial force between the retentive and mobile phase, viscosities of both phases and quality of the suitably modified inner surface of the capillary [7]. In the preconcentration procedure based on PC-OTLC, the thickness and homogeneity of the layer of the retentive pulse, the shape and steepness of the gradient elution in the region of the tail of the zone

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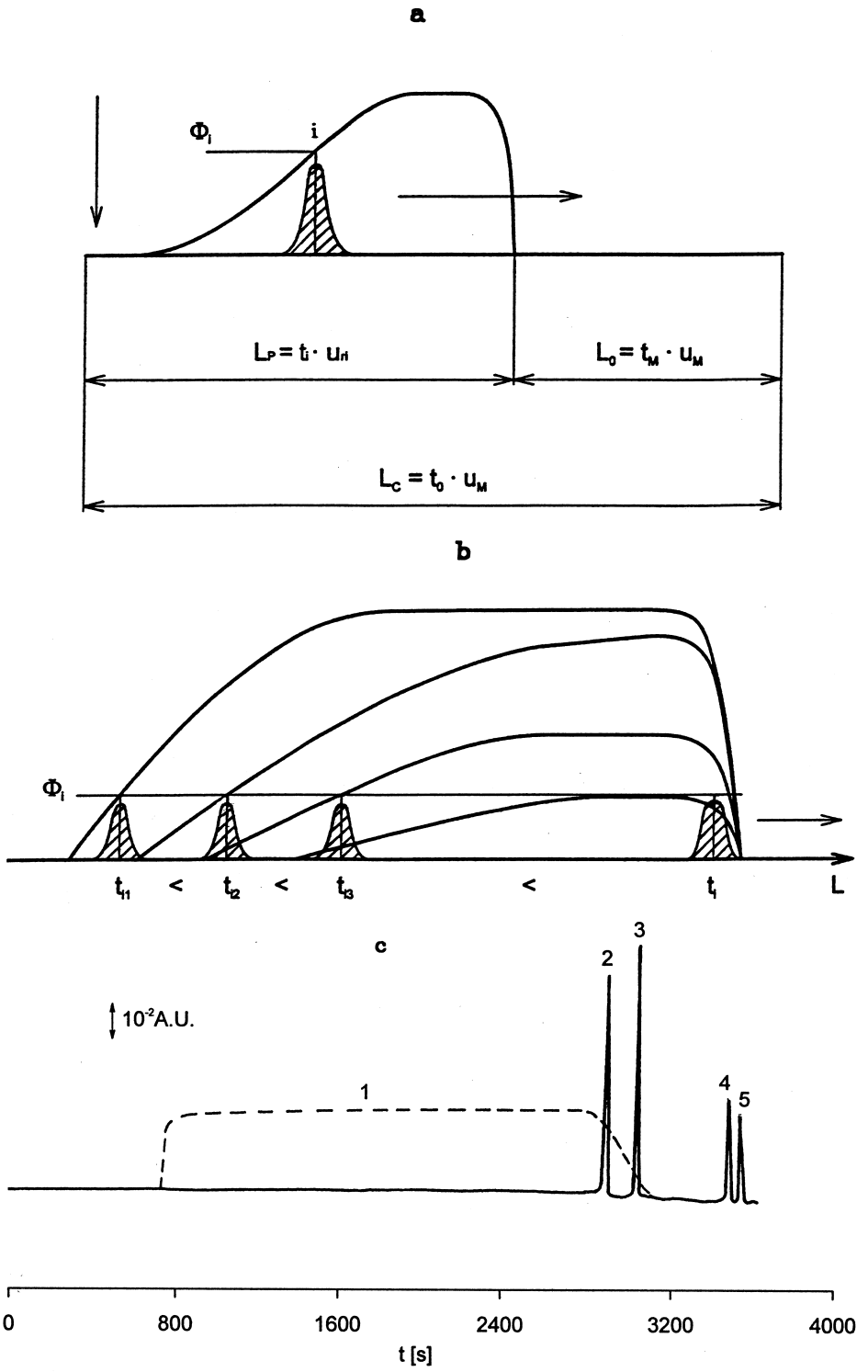
a
Φi
i
LP = ti · uп
L0 = tM · uM
LC = t0 · uM
b
Φi
t11 < t12 < t13 < t1 L
c
10-2A.U.
1
2
3
4
5
0
800
1600
2400
3200
4000
t [s]

of retentive layer [1] is also influenced by the dissolution of the retentive layer which is limited by the saturation of the mobile phase at the column temperature. That determines also the maximum injected pulse volume which can be dissolved without entering the undissolved retentive layer into the detector. Generally the shape of the gradient and the steepness of the gradient influence the retention in the gradient elution [1,8]. Thus, the change in the thickness of retentive liquid film leads to the change in the analyte retention and the peak dispersion [6,7,9–12]. However, one of the most commonly encountered complications affecting the homogeneity of capillary flow is that arising from variations in the solid–liquid interfacial tension and surface tension gradients [13–15]. In spite of the complexity of the phenomena involved, the enrichment and pre-separation by the proposed method is achieved simply by the injection of suitable volume of the retentive liquid followed by the injection of the sample solution on the suitably modified capillary. The feasibility of the procedure was shown in the previous paper [1] where the enriched concentration at the capillary outlet was achieved by more than one-order of magnitude higher than the concentration in the injected sample.

In this paper we study the influence of the internal diameter and the length of the capillary, the volume of the injected pulse of retentive liquid on the efficiency of the enrichment.

1.1. Model description

Under steady state conditions in PC-OTLC, the flow friction force keeps the thickness of the retentive film constant round the internal circumference of the capillary. Under transient conditions, when the pulse of retentive liquid is injected into the capillary, it is subjected to the retarding action of the walls; the initially flat velocity profile gradually approaches a Poiseuille profile [17] in a very short distance from the capillary inlet. When all liquid is transformed into the layer, the meniscus which coats the capillary wall by the retentive liquid disappears at a certain distance from the capillary inlet [11]. The diffusion coefficient in a liquid is small compared to its kinematic viscosity, the region over which the distribution of diffusing liquid becomes established should be longer than the region required for establishing the Poiseuille velocity profile [17]. The value of the diffusional flux of the retentive phase from the capillary wall into the flowing liquid depends on the inner diameter of the capillary, the solubility of retentive liquid in the mobile phase, on the diffusion coefficient of this liquid, velocity of the mobile phase and on the distance from the capillary inlet [17]. We supposed that for a capillary of a few tenth mm diameter and physical properties of the liquid used the length of the decreasing thickness of the retentive layer appears to be up to several tens of mm. The above estimation is based on the calculation with zero bulk concentration [17]. However, for the non-zero concentration of the retentive liquid in the bulk mobile phase, the length of the gradient should be even longer.

Again, due to the friction forces, the layer moves in the direction of the mobile phase, however, with substantially lower velocity [6]. In our water–cyclohexanol system, the average film velocity under steady state conditions is about 3% of the mobile phase velocity [16]. According to the model supposed (Fig. 1a,b), the analytes move on gradient of

Fig. 1. Model of the focusing on the pulse of retentive liquid. (a) Basic characteristics of the pulse of the retentive liquid. L_C: length of the capillary (mm); L_0: the length of the capillary from the place of the dissolution of the retentive pulse to the detection cell (mm); L_P: the length of the capillary on which the pulse of the retentive liquid dissolves during the preconcentration procedure. t_s: time of the dissolution of the segment of the retentive liquid; t_M: the time of the moving of the solute after dissolution of the retentive layer; t_i: the retention time of the solute i, on the capillary length L_P; t_0: the dead time; u_M: the linear velocity of the mobile phase (mm s^{-1}); u_{ri}: the average velocity of the solutes i on the part of the capillary, L_P, (mm s^{-1}); Φ_i: the phase ratio at position of the solute i. (b) Shapes of the pulses of retentive liquid before their dissolution in time t_i: t_{i3}; t_{i1}, t_{i2} and t_{i3} are different times after the pulse injection (s); designations as in (a); L: the length coordinate of the capillary. (c) Focusing of model analytes: curve 1 = sodium chromate (10^{-4} mol l^{-1}, 236 nm), marker of the dead time, t_0, peaks 2 = phenol (10^{-5} mol l^{-1}, 283 nm), 3 = 2,4-dinitroaniline (10^{-5} mol l^{-1}, 336 nm), 4 = phenanthrene (10^{-7} mol l^{-1}, 235 nm), 5 = fluoranthene (10^{-7} mol l^{-1}, 235 nm). Conditions: 15 000 × 0.095 mm I.D. fused-silica capillary modified by D_4 reagent; column temperature: 50°C; pumped liquid: 10 mmol l^{-1} CH_3COONa in water; linear mobile phase velocity, u_M: 18 mm s^{-1}; sample volume, V_S: 250 μl; volume of injected cyclohexanol, V_{CHOH}: 13 μl.

the decreasing thickness of the retentive layer. It follows from the general principle of the gradient elution at the constant retention strength of the mobile phase that the analytes with high retention are focused at the gradient positions with a characteristic phase ratio (Fig. 1b). Thus, the focused analytes migrate together with the layer thickness which is substantially smaller than the steady state one. When the thickness of the retentive layer or the phase ratio, respectively, becomes smaller than the characteristic one, the analyte leaves the retentive layer.

Since the average layer velocity is proportional to the square root of its thickness [6], it is reasonable to assume for the first approximation, that movement of the film is negligible compared to that of the mobile phase [16] and that the tail of its decreasing thickness moves along the capillary only due to its dissolution in the mobile phase. It allows us to assume that, at the particular focusing run, all focused analytes leave the two-phase region at the same distance from the capillary inlet, L_P. The distance from the place of leaving the retentive pulse to the detection cell is L_0. The whole length of the capillary, L_C, is then

$$L_C = L_P + L_0 \tag{1}$$

As long as the retentive layer can be regarded as negligibly thin in comparison with the capillary diameter, the length of the capillary can be expressed as

$$L_C = u_M \cdot t_0 \tag{2}$$

where u_M is the linear velocity of the mobile phase and t_0 is the elution time of the unretained analyte.

When the thickness of the front of the retentive pulse becomes so thin that it equals that in the gradient position of the solute *i* (Fig. 1b), the focused peak of solute *i* leaves the front of the pulse of the retentive liquid, which occurs at time t_i. The elution time from the moment of leaving the retentive layer to the position of the detection cell is t_M. The total retention time from injection to the detection place, t_{ri}, is

$$t_{ri} = t_i + t_M \tag{3}$$

where t_{ri} is the whole retention time of the solute on the capillary length L_C. Since the position where all analytes leave the retentive layer is considered the same, their migration time in part of the capillary with uncovered surface, L_0, can be calculated as

$$t_M = L_0 / u_M \tag{4}$$

The average velocity of the solute *i* in the part of capillary with the tail of retentive liquid, u_{ri}, can be expressed with help of L_P and t_i as

$$u_{ri} = L_P / t_i \tag{5}$$

By insertion of Eqs. (2)–(5) into Eq. (1) we obtain

$$u_M \cdot t_0 = u_{ri} \cdot (t_{ri} - t_M) + u_M \cdot t_M \tag{6}$$

Alternatively, for the solute *j*, it is from Eqs. (1), (2), (4), (5):

$$u_M \cdot t_0 = u_{rj} \cdot t_j + u_M \cdot t_M \tag{7}$$

Since the retentive layer is regarded as non-moving in comparison with to the mobile phase velocity, the average elution velocity of the solute *i* retarded by the tail of the retentive layer can be related to the accompanied phase ratio, Φ_i, by the relation

$$u_{ri} = u_M / (K_i \cdot \Phi_i + 1) \tag{8}$$

where K_i is the distribution constant of solute *i* between mobile and the retentive phase. Similarly, we have for the solute *j*:

$$u_{rj} = u_M / (K_j \cdot \Phi_j + 1) \tag{9}$$

With help of Eqs. (6)–(9) we obtain for the retention time of solute *i*:

$$t_{ri} = t_j \cdot [(K_i \cdot \Phi_i)/(K_j \cdot \Phi_j + 1)] + t_0 \tag{10}$$

and alternatively, for solute *j*:

$$t_{rj} = t_j \cdot [(K_j \cdot \Phi_j)/(K_j \cdot \Phi_j + 1)] + t_0 \tag{11}$$

Let us introduce the average retention factor, k^*_{fi}, of the solute *i* on the whole column length, defined as

$$k^*_{fi} = (t_{ri} - t_0)/t_0 \tag{12}$$

By insertion of Eq. (10) into Eq. (12), it can be expressed for solute *i* as

$$k_{fi}^{*} = t_j/t_0\left[(K_i\Phi_i)/(K_j\Phi_j + 1)\right] \quad (13)$$

Now, let us take the retention of solute j as the reference one, to which the retentions of all solutes i are related. The difference in the retention times, Δt_{ij}, is:

$$\Delta t_{i,j} = t_{ri} - t_{rj} \quad (14)$$

The injected volume of the retentive liquid is dissolved in time, t_s, which is proportional to the amount of retentive liquid injected, V_{CHOH}. It is further determined by the volume fraction of retentive phase soluble in water, s, and the ratio of capillary volume V_C to t_0:

$$t_s = V_{CHOH}t_0/(sV_C) \quad (15)$$

Since the rear end of the pulse dissolves in the stream of the mobile phase with finite speed, the tail of phase ratio is generated, see above. However, it is difficult to define the moment at which the film generated by injection of the volume of the retentive liquid can be regarded as dissolved at a certain capillary position. For purposes of this study, we set the moving end of the layer to such thickness (or phase ratio) which is traced by a suitable solute, which indicates that the uniform film thickness starts to decrease and a gradient develops. Thus, we choose the solute j for tracing the moving end of the retentive layer and we set $t_j = t_s$. By insertion of Eqs. (10), (11), (15) into Eq. (14) we obtain:

$$\Delta t_{i,j} = (V_{CHOH}t_0)/(sV_C)\left[(K_i\Phi_i - K_j\Phi_j)/(K_j\Phi_j + 1)\right] \quad (16)$$

For the reference analyte j, $\Delta t_{i,j}$ is zero and Φ_j is from Eqs. (1)–(4), (15)

$$\Phi_j = \left[(t_{rj}/t_0 - 1)/(1 + V_{CHOH}/sV_C - t_{rj}/t_0)\right]/K_j \quad (17)$$

When the injected volume of the cyclohexanol pulse is so large that it dissolves just when it reaches the detector, then $V_{CHOH} = V_{CHOH}^{s}$, and the solute j reaches the maximum retention time, $t_{rj} = t_s = t_{j,max}$. Then we obtain from Eqs. (11) and (16):

$$\Phi_j = 1/K_j(t_{j,max}/t_0 - 1) \quad (18)$$

By insertion of Eq. (18) into Eq. (17) we obtain the relation:

$$V_{CHOH}^{s}/V_C = st_{j,max}/t_0 \quad (19)$$

It follows from the above simple model that the retardation of the analytes increases with the amount of injected volume of the pulse of retentive liquid and with the length of the capillary. Also, the properties of their inner surfaces, flow velocity of the mobile phase and capillary temperature play an important role. The behavior of the system retentive pulse – inner surface of the capillary – stream of the mobile phase in the final phase of dissolution of cyclohexanol in the capillary could be somewhat different from the suggested model description since the retentive layer may be nonhomogeneous.

2. Experimental

2.1. Instrumentation

2.1.1. Preconcentration procedure

Experiments were carried out using laboratory-made apparatus [1]. Mobile phase was delivered by an HPP 5001 high-pressure syringe pump (Laboratory Works, Prague, Czech Republic). A laboratory-made injector consisted of a six-port valve with an external loop (10–500 μl) coupled with a three-way injection block made from PTFE. The sample was injected using a six-port valve. The cyclohexanol was injected with the 100 μl syringe. Fused-silica capillaries of 0.095 and 0.2 mm I.D. were purchased from Lachema Brno, Czech Republic.

The length of the capillaries used was 1500–15 000 mm. The internal surface of the capillaries was persilylated with octamethylcyclotetrasiloxane (D_4 reagent) (VCHZ Synthezia, Kolín, Czech Republic) [9]. The procedure was carried out twice. Some capillaries were hydrothermally modified after persilylation in this procedure and the capillaries were filled with distilled water by 80%, than both ends of the capillaries were closed. The capillaries were thermostatted for 2 h at 250°C.

The modified capillary was connected directly to the injection block and was immersed in a water-bath maintained at 50°C by a U8 thermostat (MLW

Prüfgeräte–Werk, Medingen/Sitz Freital, Germany). On-column UV detection with optical fibers was used. A variable-wavelength UV detector (LCD 2082, ECOM, Prague, Czech Republic) was optically connected by means of spherical quartz lens and optical fibers (Polymicro Technologies, Phoenix, AZ, USA) to the detection cell. Geometry of the detection spot is defined by the dimensions of the fused-silica capillary used (0.36 O.D.×0.2 mm I.D., or 0.21 O.D.×0.095 mm I.D.) and by the core diameter of the optical fiber (0.2 mm). Effective volume of the on-column detection cell is than less than 5 nl so that it does not influence the shape of the monitored peaks. The detector signal was registered with a TZ 4100 line recorder (Laboratory Instruments, Prague, Czech Republic).

2.1.2. Gas chromatography

Laboratory-made SIMAX glass (Kavalier Sázava, Czech Republic) capillary column (26 m×0.2 mm I.D., film thickness, d_f, 0.3 μm) was coated with SE-30 stationary phase (WGA, Dusseldorf, Germany). The inner surface of the capillary was silanized with D_4 reagent. Nitrogen as the carrier gas at linear velocity 25 mm s^{-1} was used. The capillary column was thermostatted and tested in Fractovap model 2300 AC gas chromatograph equipped with flame ionization detection (Carlo Erba, Milan, Italy).

2.2. Chemicals and model analytes

The pumped liquid was 0.01 mol l^{-1} sodium acetate in water. Measurements were carried out at the mobile phase linear velocity 4–42 mm s^{-1}. The model analytes were sodium chromate (marker of the dead time, t_0), phenol, 2,4-dinitroaniline, polynuclear aromatic hydrocarbons (Lachema) and phthalic acid esters (Carlo Erba). The model analytes in gas chromatography, polynuclear aromatic hydrocarbons, phthalic acid esters and cyclohexanol were dissolved in acetone. The test solutes and other chemicals used were purchased from Lachema.

2.3. Determination of the distribution constant, K_i

The solute distribution constant, K_i, between coexisting cyclohexanol and water phases for phenol, $K_i = 16.7$ at 50°C was taken from Ref. [6]. The value for 2,4-dinitroaniline, $K_i = 90.2$ at 50°C was determined by the method described previously [18]. The logarithm of the octanol–water distribution constant of phenanthrene and fluoranthene is reported to be 4.45 [19] and 4.95 [20], respectively; a similar value is expected here for cyclohexanol–water system.

2.4. Calculations

The linear velocity of the mobile phase, u_M, was determined from the dead time of the sodium chromate, (t_0) and the capillary length, L_C. The phase ratio corresponding to solute, *i*, which migrates in tail the pulse of the retentive liquid on the part of the capillary L_P, Φ_i, was calculated from the combination of Eqs. (1)–(4), (15), (17). We can express the phase ratio as

$$\Phi_i = [(t_{ri} - t_0)/(V_{CHOH}/sF_M + t_0 - t_{ri})]/K_i \quad (20)$$

We take the solubility of cyclohexanol in water, $s = 0.03$ (v/v), at the capillary temperature 50°C [6]. When the retention time of phenol on the pulse of injected cyclohexanol is smaller than the time of the dissolution of the retentive liquid, t_s, we can express the phase ratio as

$$\Phi_i = (t_{ri} - t_0)/(t_0 K_i) \quad (21)$$

The difference between the retention time of solute, t_{ri}, (i = fluoranthene, phenanthrene, 2,4-dinitroaniline) and phenol, t_{rp}, in the gradient of the retentive layer, Δt_r, were calculated from Eq. (22)

$$\Delta t_r = t_{ri} - t_{rp} \quad (22)$$

For phenol it is $\Delta t_r = 0$.

The volume fraction of the injected pulse of cyclohexanol, V_{CHOH}, into the capillary volume, V_C, was calculated as the ratio of both values, V_{CHOH}/V_C. The enrichment factor, *F*, was calculated from the time-based standard deviation of the eluted peak of the solute, σ_t^f, and from the time-based length of the volume of the injected sample plug, V_S, which can be focused. The maximal injectable sample volume is $V_S = t_{ri}F_M$, when $t_s \geq t_{ri}$, or $V_S = t_s F_M$, when $t_s < t_{ri}$. With use of the relation for standard deviation of square wave profile [21] we obtain:

$$F = V_S/(\sqrt{12} \cdot \sigma_t^f F_M) \quad (23)$$

3. Results and discussion

3.1. The length of the gradient of the phase ratio

The process of sample focusing according to the suggested model in Fig. 1a,b was examined by focusing of analytes – phenol, 2,4-dinitroaniline (single injected) and phenanthrene and fluoranthene injected together. The analytes are eluted from the capillary in accordance with the increase in their K_i values, see Fig. 1c. The zone of sodium chromate in Fig. 1c is not focused and it is eluted as a nearly rectangular profile. The sample volume of 250 μl is close to the maximum which gives separated zones of the matrix and analyte under the conditions used.

Figs. 2 and 3 show the dependencies of the retention time, t_r, for phenol, 2,4-dinitroaniline, phenanthrene and fluoranthene on the injected volume of cyclohexanol, into the capillaries, length 9500 mm (0.2 mm I.D.) (Fig. 2) and 7000, 15 000 mm (0.095 mm I.D.) (Fig. 3a, Fig. 3b), respectively. Line A shows the course of the dependence t_r on V_{CHOH} (Eq. (11)). Straight line B corresponds to the dependence t_s vs. V_{CHOH} (Eq. (15)). The section of lines A and B corresponds to the value $t_r = t_s$ and $V_{CHOH} = V^s_{CHOH}$. Then the end of the pulse of the

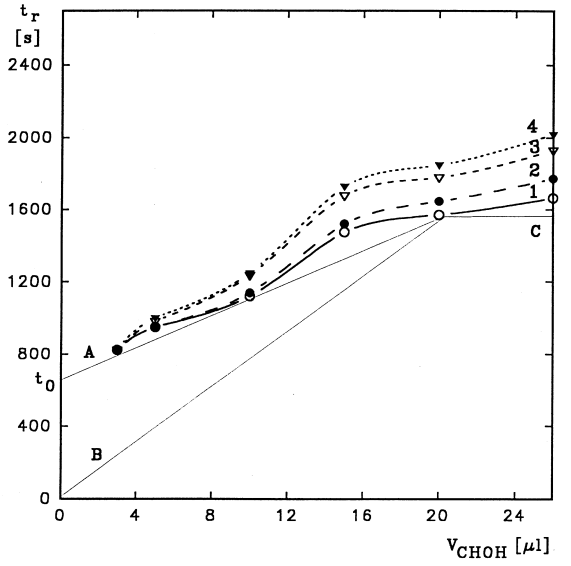

Fig. 2. Dependence of the retention time, t_r, on the injected volume of cyclohexanol pulse into the capillary 0.2 mm I.D.. Conditions and designations as in Fig. 1. Length of the capillary: 9500 mm; linear mobile phase velocity, u_M, 15 mm s^{-1}. Sample volume, V_S, 500 μl. t_r: the retention times of the solutes; analytes: curve 1=phenol (10^{-5} mol l^{-1}, 283 nm), 2=2,4-dinitroaniline (10^{-5} mol l^{-1}, 336 nm), 3=phenanthrene (10^{-7} mol l^{-1}, 235 nm), 4=fluoranthene (10^{-7} mol l^{-1}, 235 nm). Line A: the dependence calculated from Eq. (11); line B: the dependence of t_s on V_{CHOH} (Eq. (15)); line C: $t_{rj} = t_s = t_{j,max}$ = const.; t_0: dead time (s).

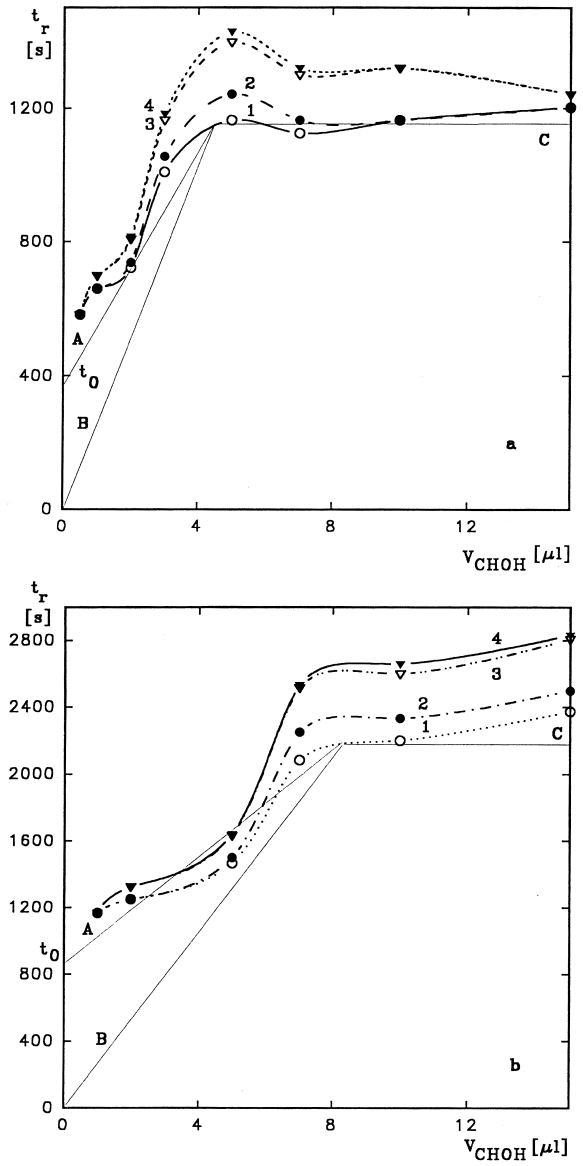

Fig. 3. Dependence of the retention time, t_r, on the injected volume of cyclohexanol pulse into the capillary 0.095 mm I.D.. Conditions and designations as in Figs. 1 and 2. Length of the capillary: (a) 7000 mm; (b) 15 000 mm.

retentive liquid dissolves approximately in the detection cell. When the volume of injected retentive liquid is further increased, ($V_{CHOH} > V^{s}_{CHOH}$), the t_r values are smaller than t_s values (see Figs. 2 and 3; line C and Eq. (19) for $t_{rj} = t_{j,max} = const.$). Of course, the film of the retentive liquid flows through the detection cell under such conditions. The found ratio V^{s}_{CHOH}/V_C is about 0.1 which is in agreement with Eq. (19) which gives $V^{s}_{CHOH}/V_C = 0.1$ for $t_{j,max}/t_0 = 3.3$ (see Fig. 3a, Fig. 3b) and $s = 0.03$. Thus, phenol can be used as indicator of dissolving end of pulse of the retentive liquid.

In Fig. 4a,b, the dependencies of difference between the retention times fluoranthene and phenol, Δt, on the injected volume of cyclohexanol are shown. The magnitude of Δt can be used as indication of the length of the gradient tail of retentive layer expressed in time units. On the capillaries of 0.2 mm I.D., the injected pulses of cyclohexanol are dissolved on the capillaries before the detection cell (Fig. 4a). However, in the range $V_{CHOH} = 16–26$ μl on the capillaries of lengths 3500 and 5500 mm, the cyclohexanol layer reaches the detection cell.

The value of Δt in dependence on the injected volume of cyclohexanol in Fig. 4b (capillary 0.095 mm I.D.) increases with the increase in the capillary length more significantly than on the capillaries of 0.2 mm I.D.. The velocity of rear end of the retentive layer is less on the capillary of 0.095 mm I.D. than on the 0.2 mm I.D. capillary and the length of the gradient thickness increases more significantly (Eq. (16) and Ref. [17]). The influence of different inner surface modification of the capillaries can be seen from comparison of curves 5 and 6; length of this capillaries were 15 000 mm. Curve 6 was obtained using the capillary which was hydrothermally modified after persilylation of the inner surface with D_4 reagent (see Section 2.1.1). As we can see later in Section 3.3 the enrichment is higher for this hydrothermally modified capillary (curve 6).

3.2. The gradient steepness

The dependencies of the calculated average phase ratio, Φ, in the tail of the retentive layer on observed Δt_r are shown in Fig. 5a–c. Different volumes of retentive liquid were injected into the capillaries of 0.095 mm I.D. and lengths 1500–15 000 mm. As can

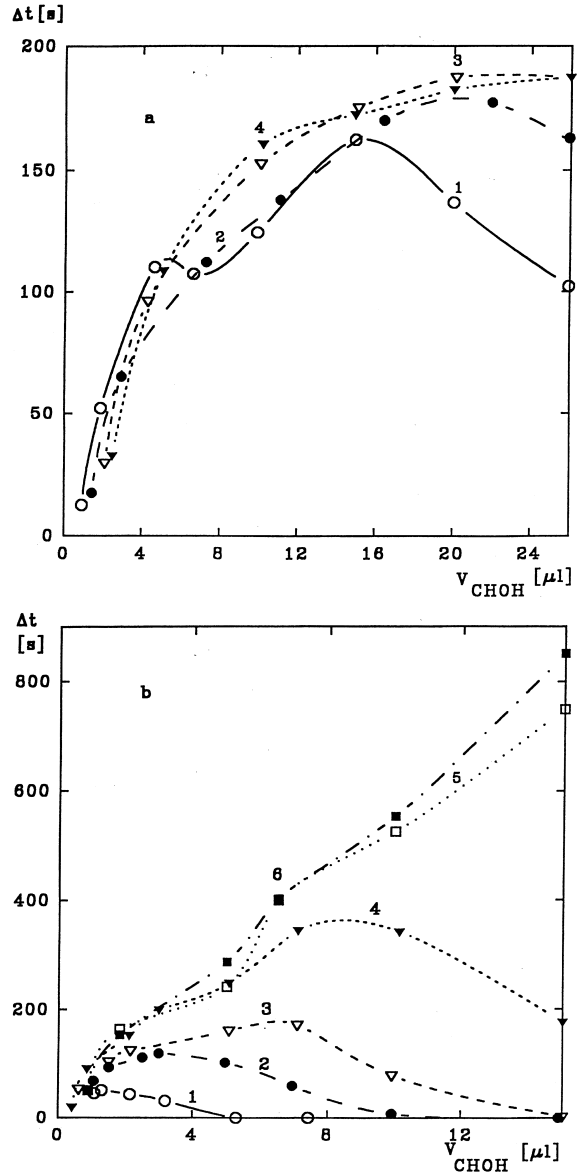

Fig. 4. Dependence of Δt on the volume of cyclohexanol pulse injected into the capillary. Conditions and designations as in Figs. 1 and 2. Capillary I.D.: (a) 0.2 mm, (b) 0.095 mm. Volume of injected cyclohexanol, $V_{CHOH} = 1–26$ μl. Length of the capillaries, (a) curve: 1 = 3500 mm; 2 = 5500 mm; 3 = 8000 mm and 4 = 9500 mm; (b) curve: 1 = 1500 mm; 2 = 3500 mm, 3 = 5000 mm; 4 = 7000 mm and 5 = 15 000 mm, 6 = 15 000 mm (the inner surface of the capillary is after persilylation of D_4 reagent hydrothermally modified). Δt: difference between the retention time fluoranthene and phenol (s).

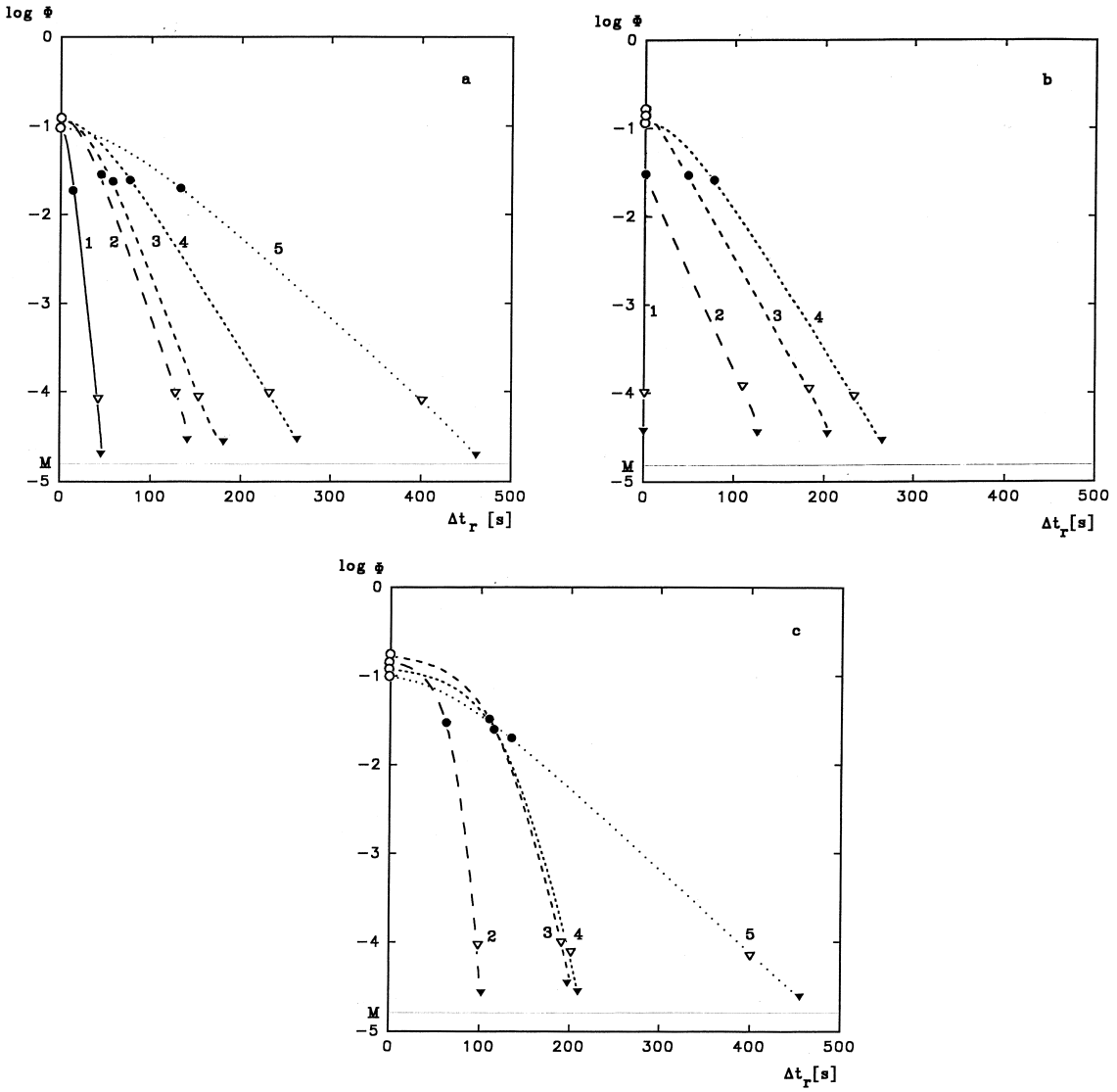

Fig. 5. Gradient steepness. Conditions and designations as in Figs. 1, 2 and 4. Fused-silica capillary 0.095 mm I.D.; length of the capillaries: 1=1500 mm; 2=3500 mm, 3=5000 mm; 4=7000 mm and 5=15 000 mm. Analytes, ○=phenol, ●=2,4-dinitroaniline, ▽=phenanthrene, ▼=fluoranthene. (a) V_{CHOH}/V_C~0.1; (b) V_{CHOH}=5 μl; (c) V_{CHOH}=10 μl. Φ: the phase ratio; Δt_r: difference between the retention times of the analytes, fluoranthene beyond phenol (s). M: the phase ratio for the monolayer of cyclohexanol on the inner surface of the capillary 0.095 mm I.D..

be seen from Fig. 5, the average phase ratio which is bound to a certain analyte were ~ the same on the capillaries of all lengths. It can be also concluded that the steepness of the gradients decreases with increasing length of the capillary. This observation is compatible with a model presented above. The line M corresponds to the phase ratio for monolayer of cyclohexanol calculated for the smooth inner surface of the capillary 0.095 mm I.D. For the calculation of the thickness of the cyclohexanol monolayer, we supposed perfectly smooth inner surface of the capillary and spherical molecules of cyclohexanol. The thickness of the monolayer was than calculated through the Avogadro number $N_A = 6.022 \cdot 10^{23}$, mo-

lecular mass of cyclohexanol, M_w, 100.16 and its density at 50°C, ~0.94 g ml^{-1}, the calculated thickness of the monolayer than equals 0.5 nm and the phase ratio $\Phi = 2.1 \cdot 10^{-5}$. We can see that the phase ratio and the thickness of the retentive layer bound to fluoranthene peak is just above the phase ratio of the monolayer of cyclohexanol. The fluoranthene is indeed on the rear end of the gradient generated by the pulse of the retentive liquid; it indicates also the maximum value of the distribution constant K_i which can be operated under the conditions used.

3.3. Factors influencing the efficiency of enrichment

In Fig. 6 the dependence of enrichment, F, on the injected volume of cyclohexanol is shown for different mobile phase velocities. The capillary used was 7000×0.095 mm I.D.. The maximum of F is around the injected volume of cyclohexanol which corresponds to the ratio $V_{CHOH}/V_C = 0.12$ (see Section 3.1) at the mobile phase velocity, $u_M = 14$ mm s^{-1}. For comparison, the maximum value of enrichment $F \sim 27$, was found previously [1] for the volume fraction of cyclohexanol $V_{CHOH}/V_C = 0.11$ for capillary 0.2 mm I.D.

In Fig. 7, the dependence F on the injected volume of cyclohexanol V_{CHOH} is shown for different lengths of the capillaries 0.095 mm I.D.. The maximum value of enrichment, F, was reached after the injection about 5.5 μl of cyclohexanol into the capillary, see curve 6, $F = 280$, at the capillary length 15 000 mm.

Based on above paragraphs, the useful ratio of retentive phase volume and capillary volume can be estimated. Though the maximum retention is reached when $V^s_{CHOH}/V_C = 0.1$, the optimum enrichment can be reached at $V^s_{CHOH}/V_C = 0.5$, 0.6, see Fig. 7, curves 5 and 6 for the system used.

3.4. Off-line GC analysis of enriched fractions

The application possibilities of suggested enrichment method are demonstrated on water solutions of polynuclear aromatic hydrocarbons and phthalic acid esters, see Figs. 8–10. The injected sample volume

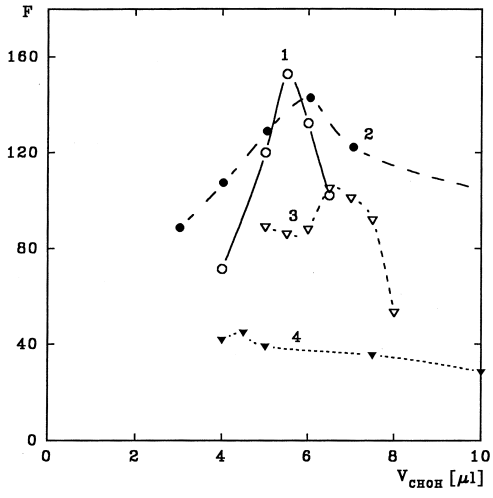

Fig. 6. Dependence of enrichment, F, on V_{CHOH} at different u_M. Conditions and designations as in Fig. 1. Capillary: 7000×0.095 mm I.D.; analyte: 2,4-dinitroaniline; u_M: 1=14 mm s^{-1}; 2=18 mm s^{-1}; 3=21 mm s^{-1}; 4=40.9 mm s^{-1}.

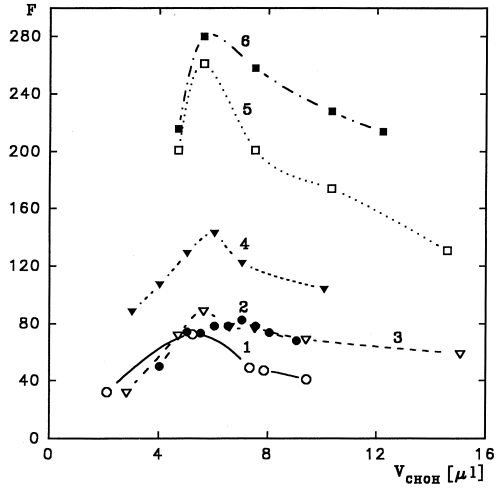

Fig. 7. Dependence of the enrichment, F, for 2,4-dinitroaniline on V_{CHOH} for different length of the capillaries. Conditions and designations as in Figs. 1 and 2. Capillary 0.095 mm I.D., length of the capillaries: 1=1500 mm; 2=3500 mm, 3=5000 mm; 4=7000 mm and 5=15 000 mm; 6=15 000 (the inner surface of this capillary is hydrothermally modified after persilylation by D_4 reagent).

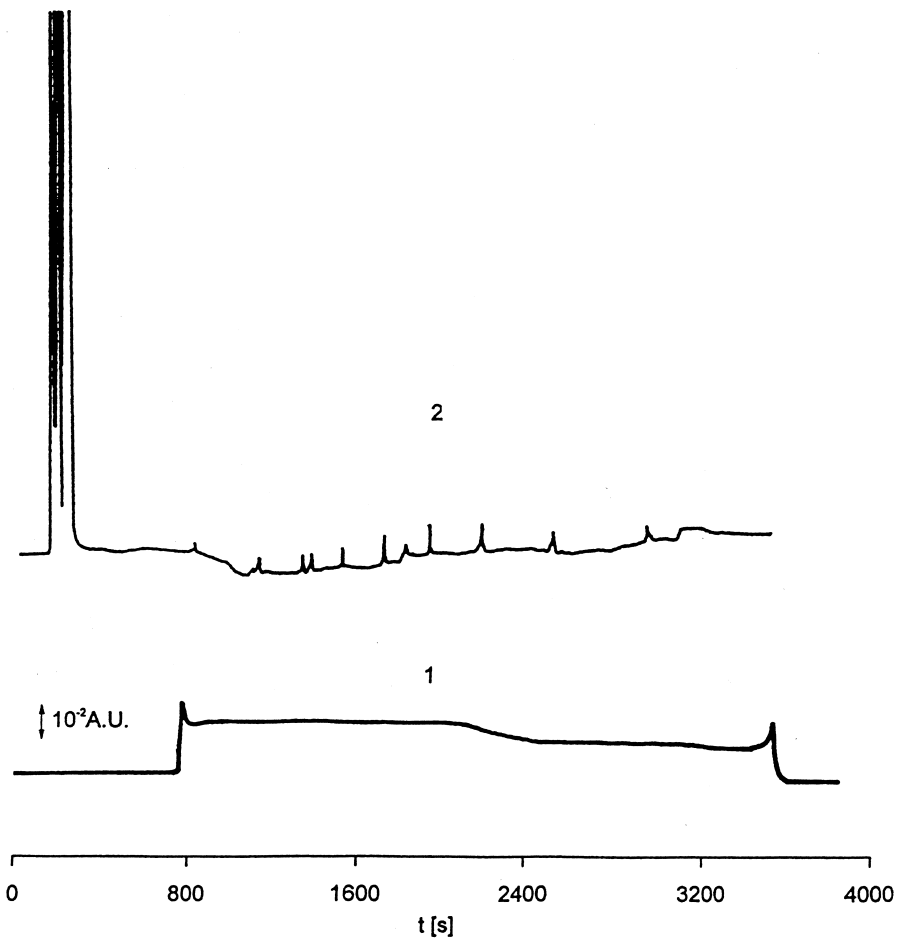

Fig. 8. Chromatograms of cyclohexanol. Chromatogram 1: response on the injection of the segment of the cyclohexanol (254 nm); conditions and designations as in Figs. 1 and 2. Chromatogram 2: gas chromatogram of cyclohexanol used in preconcentration procedure. Conditions: 26 m×0.2 mm I.D. capillary column; d_f=0.3 μm; stationary phase, SE-30; column temperature 120°C programmed at 5°C min^{-1} to 280°C; carrier gas, nitrogen; velocity 25 cm s^{-1}.

250 μl is close to the maximum which gives separated zones of the matrix and analyte under conditions used. Through the injection of cyclohexanol should not give any appreciable response of the UV–Vis detector, an appreciable signal was obtained at 254 nm, see Fig. 8, curve 1, which has been discussed [1] previously. The profile corresponding to the injection of 13 μl of cyclohexanol is shown. In capillary GC on Fig. 8, curve 2 some major impurities are eluted at the dead time.

Figs. 9 and 10 illustrate the potential of suggested method for trace analysis. In Fig. 9 the chromatograms of model mixture of water solution of polynuclear aromatic hydrocarbons are shown. The zones of injected sample of model mixture, see Fig. 9a, analytes 2–6, are preconcentrated in the tail of the retentive pulse. Presence of the analytes in the preconcentrated zone is verified by capillary GC, see Fig. 9b.

In Fig. 10, the chromatograms of model mixture of phthalic acid esters in water are shown. The injected sample is preconcentrated in the gradient tail of the

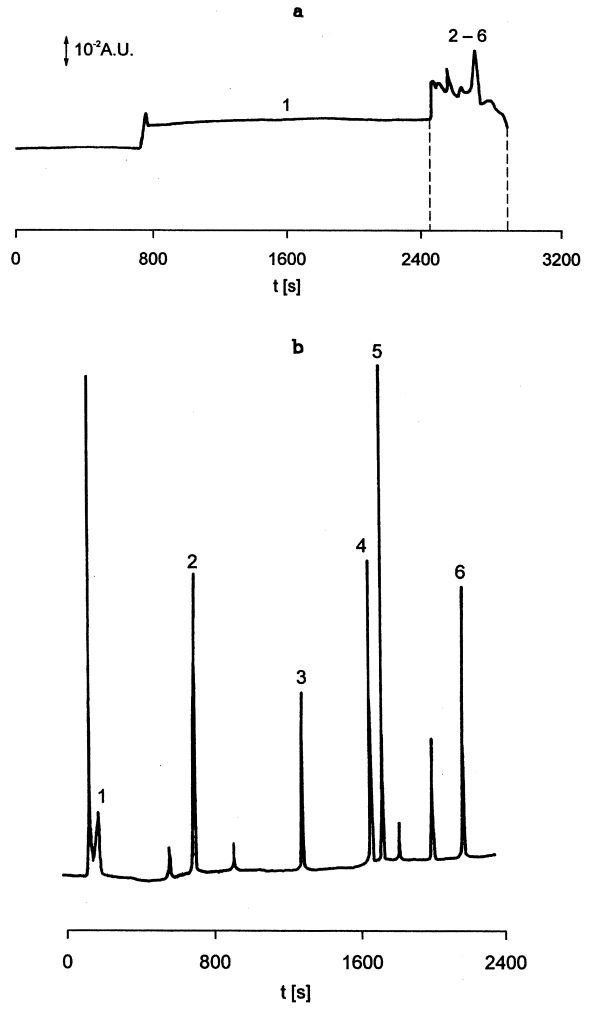

Fig. 9. Chromatograms of model mixture of polynuclear aromatic hydrocarbons. Conditions and designations as in Figs. 1 and 3. (a) Focusing of the model mixture of polynuclear aromatic hydrocarbons in water on the pulse of retentive liquid. Analytes, 1 = cyclohexanol (254 nm), 2–6 = preconcentrated zone of the polynuclear aromatic hydrocarbons (235 nm). (b) Gas chromatogram of the analytes from the preconcentrated zone ($\sim 10^{-3}$ mol l^{-1}). Conditions: 2.5 min isothermal at 120°C then programmed at 5°C min^{-1} to 280°C. Analytes, 2 = diphenyl, 3 = phenanthrene, 4 = fluoranthene, 5 = pyrene, 6 = chrysene.

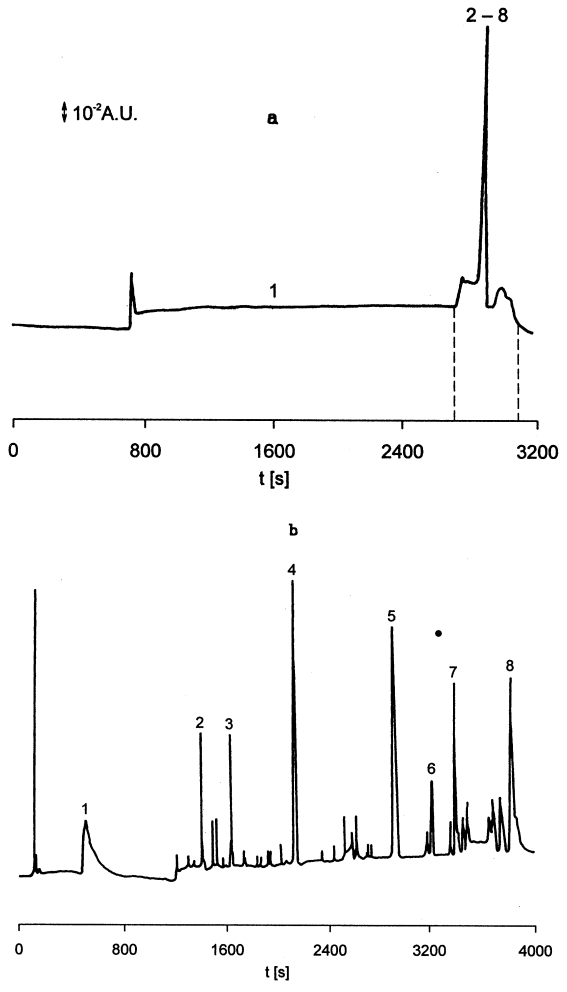

Fig. 10. Chromatograms of phthalic acid esters in water. Conditions and designations as in Figs. 1,3,9. (a) Focusing of the model mixture of phthalic acid esters in water on the pulse of retentive liquid. Analytes, 1 = cyclohexanol (254 nm), 2–8 = preconcentrated zone of the phthalic acid esters (235 nm). (b) Gas chromatogram of the analytes from the preconcentrated zone ($\sim 10^{-3}$ mol l^{-1}). Conditions: 5 min isothermal at 70°C then programmed at 2.5°C min^{-1} to 300°C. Analytes, 2 = dimethylphthalate, 3 = diethylphthalate, 4 = dibuthylphthalate, 5 = diisoamylphthalate, 6 = dioctylphthalate, 7 = dinonylphthalate, 8 = didecylphthalate.

retentive pulse, see Fig. 10a, enriched fraction of analytes 2–8. The preconcentrated zone is collected and injected into the GC system, see Fig. 10b.

The chromatograms show that method permits the group separation of the analytes with broad range of distribution constants. The collected enriched solution can be treated by other off- or on-line analyses. The described preconcentration procedure based on

the parallel current chromatography can handle analytes down to sub-ppb concentrations in injected solution.

4. List of symbols

F	The enrichment factor
F_M	The flow-rate of the mobile phase (μl s^{-1})
i	Solute
j	Solute which indicates the beginning of the tail of the retentive layer
k_f^*	Retentive factor for solute on the capillary length, L_C
K	The distribution constant
L_C	Length of the capillary (mm)
L_0	The length of the capillary from the place of the dissolution of the retentive pulse to the detection cell (mm)
L_P	The average length of the capillary on which dissolves the pulse of the retentive liquid during the preconcentration procedure (mm)
M	the phase ratio, Φ, for the monolayer of cyclohexanol on the inner surface of the capillary (Fig. 5)
p	Phenol
s	The volume fraction of cyclohexanol in water at the capillary temperature 50°C
t_M	The time of the moving of the solute after dissolution of the retentive layer or after leaving from the retentive pulse to the detection cell (s)
t	The retention time of the solute i, j, p, on the capillary length, L_P, (s)
t_0	Dead time (s)
t_r	The retention time of the solute i, j, p on L_C, (s)
$t_{j,max}$	The maximum retention time of the solute j when $V_{CHOH} \geq V^s_{CHOH}$
t_s	Time of the dissolution of the pulse of the retentive liquid (s)
Δt	The difference between the retention times of fluoranthene and phenol in the tail of the retentive layer (s), Fig. 4
Δt_r	Retention time the difference between phenol and the other solutes in the tail of the retentive layer (s)
u_M	The linear velocity of the mobile phase (mm s^{-1})
u_r	The average velocity of the analytes on L_P (mm s^{-1})
V_C	The volume of the capillary (μl)
V_{CHOH}	The volume of the injected pulse of cyclohexanol (μl)
V^s_{CHOH}	The volume of the injected pulse of cyclohexanol (μl) for which the retention time of solute j is $t_{j,max} = t_{rj} = t_s$
V_S	Volume of the injected sample (μl)
Φ	The phase ratio
σ_t^f	The time based standard deviation of the eluted peak of the solute (s)

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