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Chromatographic behaviour of oligoethylene glycol nonylphenyl ether anionic surfactants in normal-phase high-performance liquid chromatography

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Abstract

The retention behaviour of sulphated oligoethylene glycol nonylphenyl ether anionic surfactants was investigated in normal-phase systems with nitrile and amino chemically bonded stationary phases and with organic mobile phases containing tetraalkylammonium additives. Non-linear plots of the logarithms of capacity factors versus the number of oxyethylene units in sulphated oligomers were observed on the bonded amino column in mobile phases consisting of cetyltrimethylammonium bromide, 2-propanol and *n*-heptane. It is possible to separate sulphated oligomers and their parent non-sulphated analogues in a single run in these systems using gradient elution.

Keywords: Mobile phase composition; Surfactants; Oligoethyleneglycol nonylphenyl ethers

1. Introduction

Oxyethylenated derivatives of aliphatic alcohols, carboxylic acids and phenols are common non-ionic surfactants and sulphated ethoxylates are also widely used as anionic surfactants in various industrial and household applications. Efficient separations of oxyethylenated non-ionic surfactants can be achieved by high-performance liquid chromatography (HPLC) both in normal-phase [1–11] and in reversed-phase systems [12–18]. Little work has been published on the systematic investigation of their retention behaviour [19,20] and much less on the chromato-

graphic separation of anionic sulphated derivatives than that of their non-sulphated parent compounds. In addition to the separation of the sulphated from the non-sulphated fraction [21], ion-pair reversed-phase chromatography has been used to separate the individual oligomers in the sulphated and non-sulphated fractions [22–25]. Both non-sulphated and sulphated oligomers are eluted in order of increasing number of oligomeric oxyethylene units in normal-phase systems. In reversed-phase systems, the order of elution of the oligomers depends on the length of the bonded alkyl chains and on the end-groups in the oligomeric series. For example, oligoethylene glycol phenylethers are eluted in order of increasing numbers of oxyethylene units on an

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octadecylsilica column [25] and so are oligoethylene glycol octylphenyl ethers on a trimethylsilyl column in mobile phases consisting of water and methanol [18], while oligoethylene glycol nonylphenyl ethers co-elute on a C_{18} column in these mobile phases and their order of elution is reversed in aqueous propanol or dioxane [23], as in chromatography on a mixed-mode reversed-phase–ion-exchange column [26]. Separation of sulphated oxyethylenated surfactants was also attempted by ion-exchange chromatography on silica gel-based stationary phases [27]. Mixtures containing oxyethylenated alkylphenols and carboxylic acids formed by their biodegradation were separated in reversed-phase systems with aqueous–organic mobile phases containing ionic additives [28].

We have suggested equations for the description of the simultaneous effects of the degree of polymerization, n , and of the concentration of the stronger solvent in binary mobile phases on the retention in oligomeric series, in both reversed-phase [23,29], and normal-phase [30] systems. Recently, we have extended the description of the retention behaviour to anionic oligomeric surfactants in reversed-phase ion-pair systems with aqueous–organic mobile phases containing cationic additives with long alkyl chains [31].

In this work, the retention behaviour of anionic surfactants of the type of sulphated oligoethylene glycol nonylphenyl ethers was studied in normal-phase systems with organic quaternary ammonium salts as additives to the mobile phases and conditions were established for the separation of both non-ionic oligoethylene glycol nonylphenyl ethers (OEGNPEs) and their anionic sulphated derivatives in a single run on amino- and nitrile-bonded columns.

2. Theoretical

Sulphated OEGNPEs can be separated by HPLC in reversed-phase systems with non-polar column packing materials and aqueous–organic mobile phases containing ion-pairing reagents [32]. We were interested in the possibilities of separating these compounds in normal-phase

systems with silica gel or polar bonded phases as column packing materials and organic mobile phases, which to our knowledge have not been investigated so far.

We have recently presented a model of retention for oligomeric series in normal-phase systems with binary mixtures of organic solvents as the mobile phases. The treatment departs from Snyder's model of competitive adsorption and, with some simplification, it yields a simple equation for the capacity factors ($k' = V_R/V_M - 1$, where V_R is the elution volume and V_M is the column dead volume) depending on the number of repeat structural units in the oligomers, n , and on the concentration φ of a polar solvent in binary organic mobile phases [19,30]:

$$\begin{aligned} \log k' &= \log \beta + n \log \alpha = a - m \log \varphi \\ &= a_0 - m_0 \log \varphi + (a_1 - m_1 \log \varphi)n \end{aligned} \quad (1)$$

$\log \alpha$ characterizes the selectivity of separation between the oligomers differing by one repeat structural unit and $\log \beta$ represents the contribution to the retention by the end group(s) in a given oligomeric series and chromatographic system. The constants a_{0P} , a_{1P} , m_{0P} and m_{1P} are related to the adsorption energy of the repeat structural unit, Q_i , and of the end group(s), Q_0 , and to the surface area of the adsorbent occupied by an adsorbed oligomeric unit A_i , and by the end group(s), A_0 :

$$a_0 = K + \alpha'(Q_0 - A_0 \varepsilon_b) \quad (2)$$

$$a_1 = \alpha'(Q_i - A_i \varepsilon_b) \quad (3)$$

$$m_0 = \frac{A_0}{n_b} \quad (4)$$

$$m_1 = \frac{A_i}{n_b} \quad (5)$$

where K is an auxiliary adsorbent-dependent constant, ε_b is the solvent strength (a measure of polarity) and n_b is the surface area of the adsorbent occupied by an adsorbed molecule of the more polar solvent in the binary mobile phase.

It has been found that the constants a and m in Eq. 1 are strongly correlated for a given

oligomeric series, stationary phase and components of the mobile phase [30]:

$$m = q + pa \quad (6)$$

This model was used to explain the behaviour of OEGNPE non-ionic surfactants on unmodified silica gel and on polar diol-, nitrile- and amino-bonded phases [19,20].

The retention of non-ionic OEGNPEs on a polar bonded phase decreases significantly if a cationic additive with a long alkyl chain is added to organic mobile phases. Possibly the additive competes with the analytes for the adsorption sites much like a polar organic solvent does in mixed mobile phases without additives and the separation of non-ionic surfactants can be assumed to be based principally on the adsorption mechanism, regardless of the degree of ionization of the tetraalkylammonium additive. In this case, Eq. 1 can be expected to describe suitably the effect of the concentration of the additive on the retention.

In normal-phase chromatographic systems, anionic surfactants such as sulphated OEGNPEs are too strongly retained even in pure polar organic solvents such as propanol or acetonitrile. We found that their elution is possible with organic mobile phases containing an ionic modifier. Because of good solubility in organic solvents, quaternary tetraalkylammonium salts, widely used as ion-pairing reagents in reversed-phase chromatography, are well suited for this purpose. These additives do not form micelles in media containing more than 20–25% of the organic solvent [32].

In normal-phase systems containing tetraalkylammonium additives, it is possible to accomplish the separation of individual sulphated OEGNPEs on various polar bonded phases. On a nitrile-bonded phase, the separation is probably based on the same principle as the separation of the parent non-sulphated oligomers. The principle of separation on a bonded amino phase can be more complex. The amino bonded phase has the properties of a weak anion exchanger and ion-exchange forces can possibly participate in the retention mechanism of sulphated OEGNPEs containing strongly acidic $-\text{O}-\text{SO}_3^-$

groups. This seems to be supported by much stronger retention of the sulphated OEGNPEs in comparison with their behaviour on the bonded nitrile phase. In mobile phases containing a tetraalkylammonium additive, the counter-anions of the additive (such as bromide anions in a tetraalkylammonium bromide) can compete with the sulphated anions for ion-exchange groups and speed up their elution. The elution can be enhanced also by the possible formation of ion pairs between the anionic analytes and tetraalkylammonium cations.

It was not the objective of the present work to investigate the exact retention mechanism in detail, as we were interested mainly in the effect of the components of the mobile phase on the retention. It was shown that, to a first approximation, the dependence of the capacity factor k' on the concentration x of the efficient eluting component in a binary mobile phase can be formally described by the same equation:

$$\log k' = a - m \log x \quad (7)$$

in normal-phase adsorption chromatography, ion-exchange chromatography and ion-pair chromatography [33]. Consequently, it is not possible to distinguish the retention mechanism only on the basis of the retention data. On the other hand, we can expect that Eq. 7 can describe the effect of the concentration of the tetraalkylammonium additive, x , on the retention in the systems studied, even if a possible combination of the above mechanisms controls the retention.

3. Experimental

Some experiments were performed using an M6000A pump, a U6K injector and an M440 UV detector operated at 254 nm (all from Waters, Milford, MA, USA), connected to a TZ 4241 line recorder and a CI 100 integrator (both from Laboratory Instrument Works, Prague, Czech Republic). An HP 1090M liquid chromatograph equipped with a diode-array UV detector, operated at 230 nm, an automatic sample injector, a 3DR solvent-delivery system, a thermostatted

column compartment and a Series 7994A workstation was used for other experiments.

Stainless-steel columns (300×4.2 mm I.D.) were packed in the laboratory with spherical amino- and nitrile-bonded phases, Silasorb SPH Amine and Silasorb SPH Nitrile, both of $7.5 \mu\text{m}$ particle size (Lachema, Brno, Czech Republic), using a high-pressure slurry packing technique.

The sample of OEGNPEs was obtained from Servo (Delden, Netherlands) under the commercial name Serdcox NNP 4. Oligomeric anionic surfactants were prepared by sulphation of this sample and the products contained both sulphated and non-sulphated oligomers. The samples were dissolved in the mobile phase at appropriate concentrations to yield good responses of the UV detector.

2-Propanol and *n*-hexane of spectroscopic grade were obtained from Lachema and cetyltrimethylammonium bromide (CTAB) and tetrabutylammonium bromide (TBAB) from Serva (Heidelberg, Germany). Water was doubly distilled in glass with addition of potassium permanganate. The mobile phases were prepared by mixing the components in the required ratios, filtered using a Millipore $0.45\text{-}\mu\text{m}$ filter and degassed by ultrasonication before use, or were prepared directly in the HP 1090M instrument from the filtered components continuously stripped by a stream of helium.

Column dead volumes, V_M , were determined using *n*-hexane as the dead volume marker and refractometric detection with an R 401 differential refractometer (Waters–Millipore).

The retention volumes, V_R , of the members of oligomeric series were measured at different mobile phase compositions. The mean values of V_R from three repeated experiments were used for calculations of the capacity factors, $k' = (V_R/V_M - 1)$, of the individual oligomers. The identification of the peaks was performed as discussed previously [19,22]. An Adstat software package and a 386 AT personal computer were used for linear and non-linear single- and multiple-parameter regression of the experimental sets of retention data as a function of the concentrations of the mobile phase components and of the number of oligomeric units.

4. Results and discussion

4.1. Influence of the mobile phase on the retention

Some results of our earlier study of the retention behaviour of non-sulphated OEGNPEs in various normal-phase systems [20] are compared here with the data measured for their sulphated derivatives. Sulphated OEGNPEs are anionic compounds that are retained so strongly on polar adsorbents in mobile phases containing only organic solvents that their elution is not possible even with pure acetonitrile or 2-propanol. To make possible the separation of the individual sulphated oligomers, it is necessary to decrease their retention by using mobile phases containing an ionic additive. We found cationic tetraalkylammonium salts, such as CTAB or TBAB, to be suitable for this purpose because of good solubility in non-aqueous media.

Figs. 1 and 2 illustrate the linear decrease in $\log k'$ with increasing logarithms of molar concentration, x , of CTAB in the mobile phase with a constant concentration of 2-propanol on the Silasorb SPH Amine and Nitrile columns, respectively, in agreement with Eq. 7. Also in agreement with Eq. 1, the dependence of $\log k'$ on the logarithm of the volume concentration, φ , of 2-propanol in the mobile phase is characterized

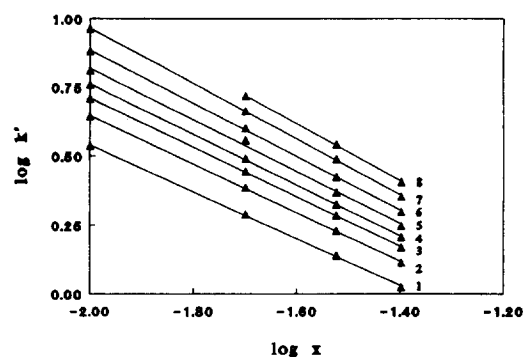


Fig. 1. Dependence of retention (capacity factors, k') of sulphated OEGNPEs with various numbers of oxyethylene units (numbers on the plots) on the concentration x (mol/l) of CTAB in mobile phases containing 13.75% (v/v) 2-propanol in *n*-heptane. Column: Silasorb SPH Nitrile, $7.5 \mu\text{m}$ (300×4.2 mm I.D.).

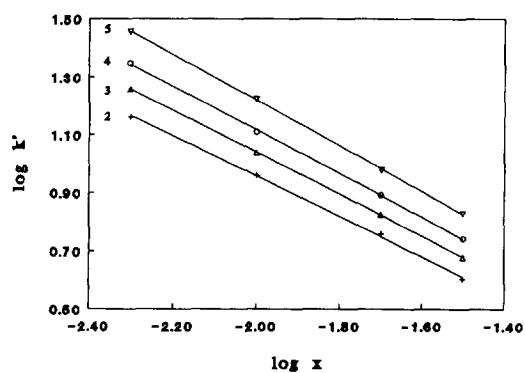


Fig. 2. Dependence of retention (capacity factors, k') of sulphated OEGNPEs with various numbers of oxyethylene units (numbers on the plots) on the concentration x (mol/l) of CTAB in mobile phases containing 50% (v/v) 2-propanol in n -heptane. Column: Silasorb SPH Amine, 7.5 μ m (300 \times 4.2 mm I.D.).

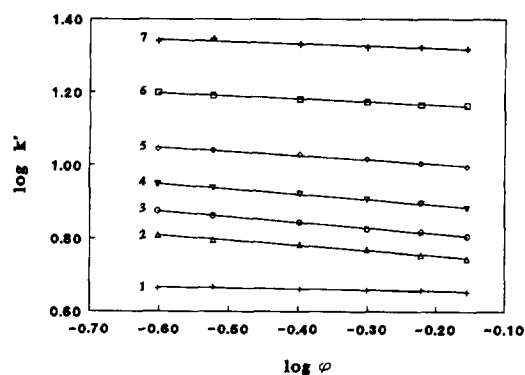


Fig. 3. Dependence of retention (capacity factors, k') of sulphated OEGNPEs with various numbers of oxyethylene units (numbers on the plots) on the concentration φ [% (v/v) $\times 10^{-2}$] of 2-propanol in n -heptane at a constant concentration of 0.02 mol/l CTAB in the mobile phases. Column as in Fig. 2.

Table 1

Experimental capacity factors, k' , of sulphated OEGNPEs on (I) a Silasorb SPH Nitrile and (II) a Silasorb SPH Amine column with a mobile phase consisting of 0.02 mol/l CTAB in 25% (v/v) 2-propanol in n -heptane.

Column	n^a						
	2	3	4	5	6	7	
I	1.71	1.94	2.13	2.33	2.70	3.12	
II	6.44	7.50	8.87	11.09	15.81	21.88	

^a Number of oxyethylene units in the individual oligomers.

by straight lines in the concentration range of 2-propanol investigated, as shown in Fig. 3 for the amino-bonded column. Similar plots can be constructed also from the data measured on the nitrile-bonded column, but because of the more limited concentration range of 2-propanol investigated, the plots are not shown here.

Sulphated OEGNPEs are more strongly retained on the Silasorb SPH Amine column than on the nitrile column (see Table 1) with a mobile phase of the same composition, probably because of strong interactions of the $-\text{OSO}_3^- \text{H}^+$ end-groups with the bonded amino groups (the bonded amino phase has weak anion-exchange properties). The retention can be much more strongly affected by changing the concentration of the tetraalkylammonium salt than that of 2-propanol, especially on the amino-bonded column (compare Figs. 2 and 3).

Table 2 compares the experimental capacity

Table 2

Experimental capacity factors, k' , of sulphated OEGNPEs on a Silasorb SPH Amine column with mobile phases consisting of (I) 0.01 mol/l CTAB, (II) 0.01 mol/l TBAB, (III) 0.04 mol/l CTAB and (IV) 0.04 mol/l TBAB in 50% (v/v) 2-propanol in n -heptane.

Mobile phase	n^a			
	2	3	4	5
I	9.12	10.91	12.88	16.71
II	7.21	9.18	12.16	16.79
III	2.84	3.29	3.86	4.66
IV	2.55	3.19	4.31	6.00

^a Number of oxyethylene units in the individual oligomers.

factors of sulphated OEGNPEs on a Silasorb SPH Amine column in mobile phases containing CTAB and TBAB. As in chromatographic systems with CTAB, the dependences of k' on the molar concentration of TBAB, x , can be described by straight lines, in agreement with Eq. 7, as illustrated in Fig. 4. CTAB is a stronger eluent than TBAB and increasing the concentration of CTAB in the mobile phase results in a more significant decrease in retention than increasing the concentration of TBAB (Table 2).

4.2. Influence of the number of oxyethylene units on the retention

In contrast to the behaviour in reversed-phase systems, where both sulphated and non-sulphated OEGNPEs are eluted in order of decreasing molecular masses, the k' values of sulphated OEGNPEs increase with increasing number of oxyethylene repeat groups, n , in all the normal-phase systems investigated in this work, like the k' of non-sulphated OEGNPEs studied earlier [20]. However, the plots of $\log k'$ versus n are sigmoidal rather than linear (see Figs. 5 and 6). This means that the structural contribution of an oxyethylene group to the retention is not constant for the oligomeric series in the normal-phase systems studied and that Eq. 1 can describe the retention only for a very

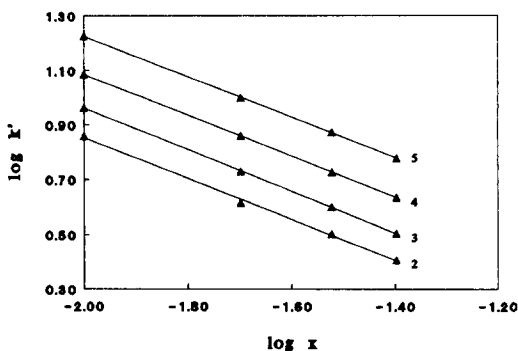


Fig. 4. Dependence of retention (capacity factors, k') of sulphated OEGNPEs with various numbers of oxyethylene units (numbers on the plots) on the concentration x (mol/l) of TBAB in mobile phases containing 50% (v/v) 2-propanol in n -heptane. Column as in Fig. 2.

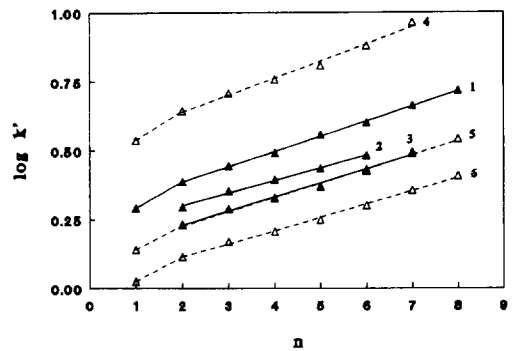


Fig. 5. Dependence of retention (capacity factors, k') of sulphated OEGNPEs in the sulphated sample of Serdiox NNP 4 on the number of oxyethylene units n on a Silasorb SPH Nitrile ($7.5 \mu\text{m}$) column ($300 \times 4.2 \text{ mm I.D.}$) in mobile phases consisting of 0.02 mol/l CTAB in (1) 13.7, (2) 20 and (3) 25% (v/v) 2-propanol in n -heptane and in (4) 0.01, (5) 0.03 and (6) 0.04 mol/l CTAB in 13.7% (v/v) 2-propanol in n -heptane.

limited range of oligomers. The deviations from the expected straight-line dependences measured on the Silasorb SPH Nitrile column are most apparent for the first member of the sulphated OEGNPE series, whereas the plots are almost linear for the oligomers with 2–8 oxyethylene units (Fig. 5).

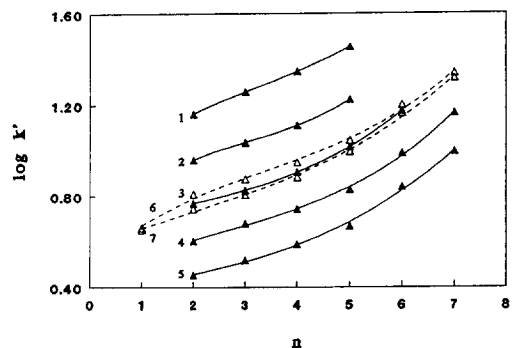


Fig. 6. Dependence of retention (capacity factors, k') of sulphated OEGNPEs in the sulphated sample of Serdiox NNP 4 on the number of oxyethylene units n on a Silasorb SPH Amine ($7.5 \mu\text{m}$) column ($300 \times 4.2 \text{ mm I.D.}$) in mobile phases consisting of (1) 0.005, (2) 0.01, (3) 0.02, (4) 0.03 and (5) 0.04 mol/l CTAB in 50% (v/v) 2-propanol in n -heptane and in (6) 25 and (7) 70% (v/v) 2-propanol in n -heptane.

The plots of $\log k'$ versus n of sulphated OEGNPEs measured on the Silasorb SPH Amine column in mobile phases containing CTAB are significantly curved and can be approximated by straight lines only for the oligomers with 2–5 oxyethylene units (see Fig. 6). The $\log k'$ versus n plots differ from analogous dependences found earlier for non-sulphated OEGNPEs in normal-phase systems [20], which indicates the important role of the $-\text{OSO}_3^-$ end-groups in the retention mechanism. With non-sulphated OEGNPEs, the slopes of these dependences are less steep for higher than for lower oligomers [20], whereas the opposite holds true for the sulphated derivatives investigated here. This means that an oxyethylene group of a higher oligomer contributes more to the retention than an oxyethylene group in a sulphated OEGNPE series member with fewer than five oxyethylene units. As the contribution to the retention of an oxyethylene group is greater for lower than for higher non-sulphated OEGNPE oligomers, the change in the retention behaviour is connected with the presence of the $-\text{OSO}_3^-$ end-group. This anionic group can be attracted to the bonded amino group of the adsorbent with weak anion-exchange properties by electrostatic forces, whereas no such attraction of the end-groups occurs with non-sulphated oligomers. Because a larger contribution to $\log k'$ of an oxyethylene group in higher sulphated oligomers means a higher energy of adsorption, more molecules of the mobile phase additive per oxyethylene unit are necessary to compete for the adsorption centers with longer than with shorter oligomers, which results in a larger area A_i and a higher value of the constant m_1 in Eq. 1 for longer oligomers. On the nitrile-bonded phase the $-\text{OSO}_3^-$ end-groups are not retained by ionic interactions, which can possibly explain larger linearity range of the dependences of $\log k'$ versus n .

The slopes m of the $\log k'$ versus x plots are higher when TBAB is used instead of CTAB in the mobile phases, possibly because a bulkier molecule of CTAB occupies a larger surface of the adsorbent than a molecule of TBAB. This indicates a higher constant n_b and lower con-

Table 3

Best-fit parameters of the Eq. 1 for the dependence of k' on the number of oxyethylene units, n , in sulphated OEGNPEs and on the concentration of 2-propanol, φ [% (v/v) $\times 10^{-2}$] in *n*-heptane with 0.02 mol/l CTAB on (I) Silasorb SPH Nitrile and (II) Silasorb SPH Amine columns.

Parameter	Column I	Column II
a_0	-0.196	0.532
a_1	0.039	0.082
m_0	0.554	0.173
m_1	0.016	0.010
Range of n	2–7	2–5
φ	0.137–0.250	0.3–0.7

stants m_0 and m_1 in Eqs. 4 and 5 for the systems with CTAB.

4.3. Selectivity of separation

Tables 3 and 4 list the constants a_0 , a_1 , m_0 and m_1 of Eq. 1 for sulphated OEGNPEs in the linear range of the $\log k'$ versus n dependences.

The selectivity of separation of the individual oligomers (α in Eq. 1) in the studied range of mobile phases with CTAB is higher on the bonded amino phase (1.19–1.26 for sulphated and 1.82–1.91 for non-sulphated OEGNPEs) than on the nitrile phase (1.11–1.16 for sulphated and 1.38–1.48 for non-sulphated OEGNPEs), probably because of a stronger energy of ad-

Table 4

Best-fit parameters of the Eq. 1 for the dependence of k' on the number of oxyethylene units, n , in sulphated OEGNPEs and on the concentration of (I, II) CTAB and (III) TBAB, φ (mol/l) in (I) 13.7% (v/v) and (II, III) 50% (v/v) 2-propanol in *n*-heptane on (I) Silasorb SPH Nitrile and (II, III) Silasorb SPH Amine columns

Parameter	System I	System II	System III
a_0	-1.136	-0.502	-0.950
a_1	0.015	0.021	0.136
m_0	0.816	0.641	0.757
m_1	0.026	0.033	-0.002
Range of n	1–8	2–5	2–5
φ	0.01–0.04	0.005–0.04	0.01–0.04

sorption of the repeat oxyethylene unit on the former adsorbent. The value of α decreases very slightly as the concentration of CTAB increases (positive, but very low values of the parameter m_1 , which is a measure of the dependence of α on the concentration of the mobile phase additive). On the nitrile column the selectivity decreases as the concentration of propanol, φ , increases, whereas it is independent of the concentration of AB ($m_1 = 0$, $\alpha = 1.3$). On the amino column α is virtually independent of the concentration of propanol, φ ($m_1 = 0.01$). Consequently, better separations of sulphated OEGNPEs are expected on the amino- than on the nitrile-bonded phase using mobile phases with lower concentrations of propanol and higher concentrations of CTAB.

The constants a_1 and m_1 in Eq. 1, describing the effect of 2-propanol on the retention, are larger for non-sulphated ($a_1 = 0.06$ and $m_1 = 0.14$) than for sulphated ($a_1 = 0.039$ and $m_1 = 0.016$) OEGNPEs on the nitrile-bonded column. Therefore, the selectivity of separation of the sulphated oligomers, α (Eq. 1), is lower and less influenced by the concentration of 2-propanol than that of the non-sulphated oligomers. On the other hand, there are no significant differences in the dependences of the selectivity of separation on the concentration of 2-propanol on the bonded amino phase for the two groups of oligomers, as demonstrated by the equal values of the constants $m_1 = 0.01$. Low values of the constants m_1 , especially for the Silasorb SPH Amine column and mobile phases containing TBAB (Table 4), mean that the contribution of one oxyethylene unit to the retention (and to the selectivity of separation) does not depend significantly on the concentration of CTAB and is virtually independent of the concentration of TBAB in the mobile phase. Hence the concentration of TBAB in the mobile phase controls only the retention, and not the selectivity, of separation of sulphated OEGNPEs. Adjusting the concentration of CTAB or of 2-propanol also has little effect on the selectivity of separation in the series of sulphated OEGNPEs. Further, the elution of higher oligomers in mobile phases with TBAB takes a longer time than in the systems

with CTAB and, in spite of the slightly lower selectivity, CTAB is to be preferred.

Sulphated oligomers are usually present in mixtures with non-sulphated oligomers. To accomplish the separation of the individual oligomers in the two groups in a single run, the retention of the earliest eluted sulphated oligomer should be higher than that of the most strongly retained non-sulphated oligomer. This can be achieved by adjusting simultaneously the concentrations of 2-propanol to affect preferentially the retention of non-sulphated OEGNPEs and of the tetraalkylammonium salt (CTAB), which controls mainly the capacity factors of sulphated oligomers, even though the retention of non-sulphated OEGNPEs decreases to some extent with increasing concentration of the cationic additive in the mobile phase. To separate such mixtures in a reasonable time, the application of gradient elution is necessary. It is possible to accomplish the separation in the two groups of oligomers with up to fifteen oxyethylene units in ca. 35 min on the nitrile column, as illustrated in Fig. 7, but the resolution of the sulphated oligomers is incomplete. Better selectivity and significantly improved separation of sulphated oligomers than on the nitrile column can be achieved on a Silasorb SPH Amine column, but at a cost of impaired resolution of the non-sulphated OEGNPEs and longer separation times. Anyway, the overall resolution is better than that achieved previously in reversed-phase systems [31].

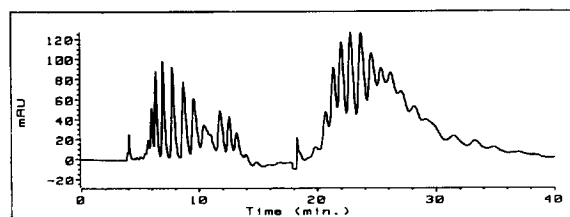


Fig. 7. Separation of non-sulphated (first group) and sulphated (second group) OEGNPEs in a partially sulphated sample of Serdox NNP 4 on a Silasorb SPH Nitrile ($7.5 \mu\text{m}$) column ($300 \times 4.2 \text{ mm}$ I.D.) using elution with a linear gradient from 0.02 mol/l CTAB in *n*-heptane to 0.02 mol/l CTAB in 2-propanol-*n*-heptane (7:93%, v/v) in 42 min at 1 ml/min. Detection, UV at 230 nm.

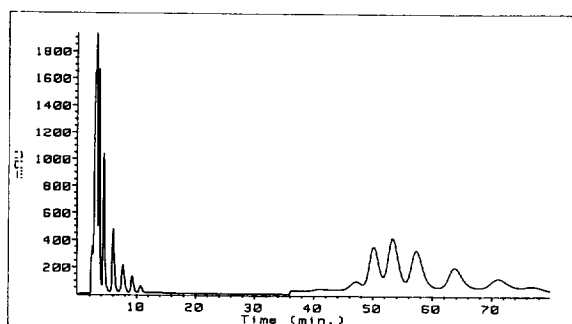


Fig. 8. Separation of non-sulphated (first group) and sulphated (second group) OEGNPEs in a partially sulphated sample of Serdox NNP 4 on a Silasorb SPH Amine (7.5 μm) column (300 \times 4.2 mm I.D.) using elution with a ternary linear gradient from 5% (v/v) 2-propanol in *n*-heptane to 0.02 mol/l CTAB in 20% (v/v) 2-propanol in *n*-heptane in 90 min at 1 ml/min. Detection, UV at 230 nm.

When binary gradient elution with increasing concentration of CTAB and a constant concentration of 2-propanol in heptane (1:1) is used, the separation of the sulphated oligomers is satisfactory, but the non-sulphated fraction is practically unresolved. To accomplish at least a partial separation of the individual oligomers in both the sulphated and the non-sulphated fractions, a ternary gradient with simultaneously increasing concentrations of propanol and cetyltrimethylammonium ions is necessary. Fig. 8 shows the separation of a partially sulphated product with four nominal oxyethylene units on a Silasorb SPH Amine column using elution with such a ternary gradient, where the individual oligomers are separated with significantly improved resolution of the non-sulphated fraction.

5. Conclusions

The effect of tetraalkylammonium salts on the retention of non-ionic oligomers such as OEGNPE surfactants in non-aqueous mobile phases is similar to that of the concentration of the more polar solvent in the mobile phase.

Simultaneous separation of the individual oligomers in the non-sulphated and sulphated fractions of partially sulphated samples of OEGNPEs can be accomplished on both the

nitrile- and the amino-bonded phases. The selectivity of separation and the resolution of the oligomers in the sulphated fraction are better on the Silasorb SPH Amine column, but ternary gradients of simultaneously increasing concentrations of the tetraalkylammonium salt and of the polar solvent in a non-polar solvent are required for this purpose.

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