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Solubilization of liposomes formed by lipids modeling the stratum corneum caused by alkyl pyridinium surfactants

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Abstract

The interactions of a series of alkyl pyridinium surfactants (alkyl chain lengths C-10 (DePB), C-12 (DoPB) and C-14 (TePB)) with liposomes modeling the stratum corneum (SC) lipid composition (40% ceramides, 25% cholesterol, 25% palmitic acid and 10% cholesteryl sulfate) were investigated. The surfactant:lipid molar ratios (Re) and the bilayer–aqueous phase partition coefficients (K) were determined by monitoring the changes in the static light scattering of the system during solubilization. The fact that the free concentration was always similar to the surfactant critical micelle concentration (CMC) indicates that liposome solubilization was mainly ruled by formation of mixed micelles. The Re and K values fell as the surfactant alkyl chain length decreased or their CMC increased. Thus, the higher the surfactant CMC the higher the surfactant ability to saturate or solubilize SC liposomes and the lower its degree of partitioning into liposomes. The balance of these two tendencies shows that the TePB and DoPB had respectively the highest power of saturation and solubilization of SC structures in terms of total surfactant amounts needed to produce these effects. Different trends in the interaction of these surfactants with SC liposomes were observed when comparing the Re and *K* values with those reported for PC ones. Thus, whereas SC liposomes were more resistant to the surfactant action, the degree of partitioning of these surfactants into these liposomes was higher in all cases. © 1997 Elsevier Science Ireland Ltd.

Abbreviations: SC. stratum corneum; Cer. ceramides type III; Chol, cholesterol; PA, palmitic acid: Chol-sulf, cholesteryl sulfate; DePB, decyl-pyridinium bromide; DoPB, dodecyl-pyridinium bromide; TePB, tetradecyl-pyridinium bromide; PIPES. piperazine-1.4 bis(2-ethanesulphonic acid); SLS, static light-scattering; PI. polydispersity index; Re. effective surfactant:lipid molar ratio: Re_{sat}, effective surfactant:lipid molar ratio for liposome saturation: Re_{sol}, effective surfactant:lipid molar ratio for liposome solubilization; K, bilayer–aqueous phase surfactant partition coefficient: K_{SAT}, bilayer–aqueous phase surfactant partition coefficient for liposome saturation; K_{SOL}, bilayer–aqueous phase surfactant partition coefficient for liposome solubilization; S_W, surfactant concentration in the aqueous medium; $S_{W,SAT}$, surfactant concentration in the aqueous medium for liposome saturation; $S_{W,SOL}$, surfactant concentration in the aqueous medium for liposome solubilization; S_B, surfactant concentration in the bilayers; r^2, regression coefficient; TLC-FID. thin-layer chromatography–flame ionization detection system: CMC, critical micellar concentration.

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1. Introduction

The permeability barrier of the skin, which prevents penetration of substances from the environment is localized in the horny layer (stratum corneum), which is a compact mass of metabolically inactive cells. embedded in an intracellular matrix of non-polar continuous lamellar lipid layers (Friberg et al., 1991: Bouwstra et al., 1995). In all cellular and intercellular membranes, such bilayer-forming lipids consist predominantly of phospholipids. However. stratum corneum (SC) has been shown to be virtually devoid of phospholipids. as a result of which its ability to form bilayers has proved to be somewhat surprising (Ranasingle et al.. 1986; Wertz and Downing, 1989; Imokawa et al., 1991). In order to find out whether SC lipids could form bilayers, Wertz et al. (1986). Wertz (1992) and Abraham et al. (1987) prepared liposomes from lipid mixtures approximating the composition of SC lipids at physiological pH and investigated the interaction of these liposomes with the anionic surfactant sodium dodecyl sulfate to study its deleterious effect on human skin (Downing et al., 1993). Furthermore, Blume et al. (1993) reported the permeability of the skin to phospholipid vesicles.

Cationic alkyl pyridinium surfactants are widely used as disinfectants and germicides in pharmacological applications, where they come in contact with mucous membranes (Cutler and Drobeck, 1986). Thus, they are used to prevent the initiation and accumulation of cariogenic dental plaque and gingivitis and also as sporicidals and decontaminants in microbiological processes (Sato et al., 1991; Sugumaran and Nellaiappan, 1991; Rosenberg et al., 1992; Williams and Russell, 1992: Binney et al., 1993: Kozlovsky et al.. 1994; Russell et al.. 1994). In order to understand the principles governing the interaction of these surfactants with biological membranes. a number of investigations have been carried out using simplified membrane models as phospholipid bilayers (Yaroslavov et al., 1994; Das et al.. 1992: Suntres et al.. 1992; Togashi et al., 1991; Bhuyan et al., 1991; Urbaneja et al., 1990). This interaction leads to the breakdown of lamellar structures and the formation of lipid-surfactant mixed micelles. A significant contribution has been made by Lichtenberg (1985) who postulated that the effective surfactant to lipid ratio (Re) producing liposome saturation and solubilization depends on the surfactant critical micellar concentration (CMC) and on the bilayer–aqueous medium surfactant distribution coefficients (K).

We previously studied the interaction of different cationic surfactants with PC liposomes (de la Maza and Parra, 1995a,b) and the formation and characterization of liposomes formed with mixtures of four commercially available synthetic lipids approximating the SC composition (de la Maza et al., 1995: 1996). In this work we seek to extend these investigations by characterizing the Re and K parameters of a series of alkyl pyridinium surfactants (alkyl chain lengths C-10, C-12 and C-14) when saturated or solubilized by SC lipid liposomes. Comparison of this information with that reported for the interaction of these surfactants with PC liposomes could be useful in order to establish a criterion for the evaluation of their activity in different biological membranes.

2. Experimental

2.1. Materials

Decyl, dodecyl and tetradecyl pyridinium bromide surfactants (DePB, DoPB and TePB respectively) were purchased from Sigma and further purified by recrystallization from acetone three times. Piperazine-1,4 bis(2-ethanesulphonic acid) (Pipes) was obtained from Merck. Pipes buffer was prepared as 10 mM Pipes containing 110 mM Na_2SO_4 and adjusted to pH 7.20 with NaOH (de

la Maza and Parra, 1995a). Polycarbonate membranes and membrane holders were purchased from Nucleopore (Pleasanton, CA).

Reagent grade organic solvents, ceramides type III (Cer). cholesterol (Chol) and palmitic acid (PA) (reagent grade) were supplied by Sigma. Cholesteryl sulfate (Chol-sulf) was prepared by reaction of cholesterol with excess chlorosulphonic acid in pyridine and purified chromatographically. The molecular weight of ceremide type III used in the lipid mixture was determined by low resolution fast atom bombardment mass spectrometry (FAB-MS) using a Fisons VG Auto Spec Q (Manchester, UK) with a caesium gun operating at 20 kV. From this analysis a molecular weight of 671 g was obtained for the main component of ceremides type III used. This value was similar to the molecular weight of ceramide 3 (667 g) calculated from the structure of this compound reported by Wertz (1992), despite the fact that the ceramide type III used was a mixture of ceramides of different chain length (purity ~ 99%). As a consequence, we used the molecular weight obtained to calculate the molarity of the SC lipid mixture investigated. The lipids of the highest purity grade available were stored in chloroform–methanol (2:1) under nitrogen at −20°C until use.

2.2. Methods

We previously reported the formation and characterization of unilamellar liposomes formed by a mixture of lipids mimicking the composition of the SC (40 Cer, 25 Chol, 25 PA and 10% Chol-sulf) (de la Maza et al., 1995), which were prepared following the method described by Wertz et al. (1986). Individual lipids were dissolved in chloroform–methanol (2:1) and appropriate volumes were combined to obtain the aforementioned mixture. The lipid mixture was then placed in culture tubes and the solvent was removed with a stream of nitrogen and then under high vacuum at room temperature. Aqueous dispersions of the lipid mixture in Pipes buffer were then prepared to provide a final lipid concentration ranging frorn 1.0 to 10.0 mM. To this end. the lipids were left to hydrate for 30 min under nitrogen with occasional shaking. The resulting aggregates were then sonicated in a bath sonicator (514 ECT Selecta) at 60°C for about 15 min until the suspensions became clear. Vesicles of about 200 nm were obtained by extrusion through 800, 400 and 200 nm polycarbonate membranes at 60°C using a thermobarrel extruder equipped with a thermoregulated cell compartment (Lipex, Biomembranes, Vancouver, Canada). The preparations were then annealed at the same temperature for 30 min and incubated at 25°C under nitrogen atmosphere.

The vesicle size distribution and the polydispersity index (PI) of SC lipid liposomes after preparation was determined by dynamic light-scattering measurements using a photon correlator spectrometer (Malvern Autosizer 4700c PS/MV; Malvern, UK). The studies were made by particle number measurement (de la Maza and Parra, 1996), according to the model proposed by Rayleigh-Gans-Debye. The sample was adjusted to the appropriate concentration range with Pipes buffer and the measurements were taken at 25°C at a reading angle of 90". The unilamellar architecture of these bilayer structures was determined by measuring the internal volume of these vesicles (spectrofluorophotometric analysis of the fluorescent dye trapped into vesicles), the mean vesicle size (dynamic light-scattering techniques) and by TEM analysis of these structures (de la Maza et al., 1995).

The lipid composition and concentration of SC liposomes after preparation were determined using thin-layer chromatography coupled to an automated flame ionization detection system (TLC-FID) (Iatroscan MK-5, Iatron, Tokyo, Japan) (de la Maza et al.. 1995; Ackman et al., 1990).

In order to find out whether all the components of the SC lipid mixture formed liposomes, vesicular dispersions were analyzed for these lipids (Ackman et al.. 1990). The dispersions were then spun at 140 000 x g at 25°C for 4 h to remove the vesicles (Almog et al., 1990). The supernatants were tested again for these components. No lipids were detected in any of the supernatants.

Analyses of proton magnetic resonance ('H NMR) were carried out at temperatures ranging from 25 to 90°C to determine the phase transition

temperature of the SC lipid mixture forming liposomes. The ^{1}H NMR spectra were recorded on a Varian Unity of 300 MHz (Palo Alto, CA). The NMR spectra were measured at intervals of 5°C. The line widths of the CH, band at 1.3 ppm were measured and 1024 scans were accumulated each time. The different line widths were plotted versus the temperature, and inflexion point of the curve was taken as a phase transition temperature, which showed a value of 55–56°C.

2.3. Parameters involved in the interaction of surfactants with SC liposomes

In the analysis of the equilibrium partition model proposed by Schurtenberger et al. (1985) for bile salt–lecithin systems, Lichtenberg (1985) and Almog et al. (1990) have shown that for a mixing of lipids (at a lipid concentration L (mM)) and surfactant (at a concentration S_T (mM)), in dilute aqueous media, the distribution of surfactant between lipid bilayers and aqueous media obeys a partition coefficient K, given (in mM^{-1}) by

$$K = S_B/[(L + S_B) \cdot S_W] \quad (1)$$

where S_B is the concentration of surfactant in the bilayers (mM) and S_W is the surfactant concentration in the aqueous medium (mM). For $L \gg S_B$, the definition of K, as given by Schurtenberger, applies:

$$K = S_B/(L \cdot S_W) = Re/S_W \quad (2)$$

where Re is the effective molar ratio of surfactant to lipid in the bilayers ($Re = S_B/L$). Under other conditions, Eq. (1) has to be employed to define K and this yields:

$$K = Re/S_W[1 + Re] \quad (3)$$

This approach is consistent with the experimental data offered by Lichtenberg (1985) and Almog et al. (1990) for different surfactant lipid mixtures over wide ranges of Re values. Given that the lipid concentration range used in SC liposomes is similar to that used by Almog to test his equilibrium partition model, the K parameter has been determined using this equation.

The solubilization of SC liposomes was characterized by two parameters termed Re_{sat} and Re_{sol}, according to the nomenclature adopted by Lichtenberg (1985) corresponding to the Re ratios at which static light-scattering (SLS) starts to decrease with respect to the original value and shows no further decrease, respectively. These parameters corresponded to the surfactant:lipid molar ratios at which the surfactant: (a) saturated liposomes, and (b) led to a complete solubilization of these structures.

SC liposomes were adjusted to the appropriate lipid concentration. Equal volumes of the appropriate surfactant solutions were added to the liposomes and the resulting mixtures were left to equilibrate for 24 h. This time was chosen as the optimum period needed to achieve a complete equilibrium surfactant–liposome for the lipid concentration range used (Urbaneja et al., 1990; Partearroyo et al., 1996). SLS measurements were made with a spectrofluorophotometer Shimadzu RF-540 (Kioto Japan) with both monochromators adjusted to 500 nm at 25°C (de la Maza and Parra, 1995a). The assays were carried out in triplicate and the results given are the average of those obtained.

The determination of Re and S_W parameters was carried out on the basis of the linear dependence existing between the surfactant concentrations required to saturate and solubilize liposomes and the SC lipid concentration (L), which can be described by the equations:

$$S_{SAT} = S_{W,SAT} + Re_{SAT} \cdot [L] \quad (4)$$

$$S_{SOL} = S_{W,SOL} + Re_{SOL} \cdot [L] \quad (5)$$

where S_{SAT} and S_{SOL} are the total surfactant concentrations. The surfactant:lipid molar ratios Re_{sat} and Re_{sol} and the aqueous concentration of surfactant $S_{W.SAT}$ and $S_{W.SOL}$ are in each curve respectively the slope and the ordinate at the origin (zero lipid concentration). The K_{SAT} and K_{SOL} parameters (bilayer–aqueous phase surfactant partition coefficient for saturation and complete liposome solubilization) were determined from Eq. (3).

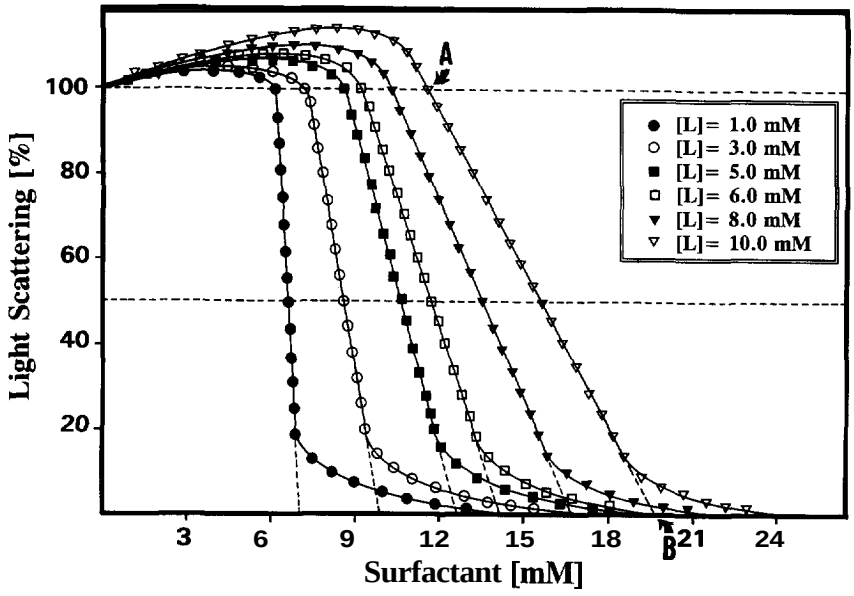

Fig. 1. Percentage changes in static light-scattering of SC liposomes. the bilayer lipid concentration ranging from 1.0 to 10.0 mM, vs. DoPB surfactant concentration. (●) [L] = 1.0 mM; (○) [L] = 3.0 mM; (■) [L] = 5.0 mM; (□) [L] = 6.0 mM; (▼) [L] = 8.0 mM; (▽) [L] = 10.0 mM.

3. Results and discussion

3.1. Stability of SC liposomes

The vesicle size distribution of SC liposomes after preparation varied little (monomodal distribution of about 100 nm in radii) and the PI was in all cases lower than 0.1 indicating that the liposomes showed a narrow particle size distribution in all cases. The vesicle size after the addition of equal volumes of Pipes buffer and equilibration for 24 h at 25°C showed in all cases values similar to those obtained after preparation, with a slight rise in the PI (between 0.12 and 0.14). Hence, SC liposome preparations appeared to be reasonably stable in the absence of surfactants under the experimental conditions used.

3.2. Parameters involved in the surfactant–liposomes interaction

The ability of the SC lipids to form bilayers has been reported by Wertz et al. (1986), who demonstrated that these lipids form liposomes when hydrated at 80°C. The Cer type III used in this work is composed primarily of simple sphingosines linked to largely monounsaturated fatty acids. It, therefore, has a much lower bulk melting temperature than SC ceramides, which contain only saturated fatty acids including hydroxyacids. In preliminary experiments we determined the suitable sonication temperature of the lipid mixture investigated by preparing liposomes at temperatures approximating its phase transition temperature *(55–56°C)*. It was found that temperatures exceeding this temperature by more than 10°C caused noticeable alterations in Cer and Cholsulf. As a consequence, the lipid mixture was sonicated at 60°C.

To determine the Re and S_W parameters, a systematic investigation of SLS variations in SC liposomes caused by the addition of different concentrations of DePB, DoPB and TePB was carried out for various lipid concentrations. The curves obtained for DoPB (SC lipid concentrations ranging from 1.0 to 10.0 mM) are given in Fig. 1. The addition of surfactant led to an initial increase and a subsequent fall in the SLS intensity

of the system until it achieved a low constant value, which corresponded to the complete bilayer solubilization via mixed micelle formation. The curves obtained for DePB and TePB showed similar trends (results not shown). This SLS behaviour is in accordance with that reported for the interaction of these surfactant with neutral or electrically charged PC liposomes (de la Maza and Parra, 1995a).

The surfactant concentrations producing 100% (S_{SAT}) and 0% (S_{SOL}) of SLS in the system were obtained for each lipid concentration by graphical methods. The arrows A and B (curve for PC concentration 10.0 mM, Fig. 1) correspond to these parameters. When plotting the surfactant concentrations thus obtained versus lipid concentration, curves were obtained in which an acceptable linear relationship was established in each case. The straight lines obtained corresponded to Eqs. (4) and (5) from which the Re and S, parameters were determined. The results obtained for each surfactant tested including the regression coefficients (r^2) of the straight lines are given in Table 1.

The Re values clearly increased from saturation to complete solubilization of SC liposomes, regardless of the alkyl chain length of the surfactant tested. In addition, the lower the surfactant hydrophobic moiety the lower the Re parameters both for saturation and solubilization of these bilayer structures. Given that the surfactant capacity to saturate or solubilize bilayers is inversely related to the Re parameter, the maximum activity at these two interaction levels corresponded to the DePB (lowest Re values), whereas the minimum corresponded to the TePB (highest Re values).

Comparison of the Re values obtained with those reported for the interaction of the same surfactants with neutral and electrically charged PC liposomes reveals that the ability of these surfactants to saturate or solubilize SC liposomes was less (higher Re values) than that reported for PC ones in all cases (de la Maza and Parra, 1995a), although showing similar tendencies with respect to the influence of the surfactant hydrophobie tail. As a consequence, SC liposomes exhibited more resistance to the surfactant perturbations than PC ones at the interaction levels investigated. These differences could be explained bearing in mind the more hydrophilic nature of PC, which could facilitate the initial association of surfactant molecules with PC bilayer structures through the formation of short-lived complexes surfactants–PC polar heads and the subsequent bilayer solubilization via mixed micelle formation (Lasic, 1994). The different gel–liquid crystal phase transition temperatures of lipids building these two liposome structures could also affect both the positional organization of lipid molecules and their polar heads as well as their mobility (SC and PC vesicles in the 'gel' and 'fluid' state, respectively at 25°C), also affecting the aforementioned processes.

The free surfactant concentration for each surfactant tested (S_{W} ... S Table 1) was always comparable to its surfactant CMC although showing slightly reduced values with respect to those reported for the interaction of these surfactants with PC liposomes (de la Maza and Parra, 1995a). These findings extend to the SC liposomes investigated the generally admitted assumption for PC ones that the concentration of free surfactant must reach its CMC for solubilization to start occurring (Lichtenberg, 1985; de la Maza and Parra, 1995a).

The K parameters (Table 1) also increased from bilayer saturation to complete solubilization of these bilayer structures regardless of the surfactant alkyl chain length. This means that the affinity of surfactant molecules with the lipids building SC liposomes was greater in bilayer solubilization (micellization process) than during the previous step of bilayer saturation (formation of mixed vesicles). In addition, the decrease in the surfactant alkyl chain length also resulted in a fall in K both for bilayer saturation (K_{SAT}) and solubilization (K_{SOL}). As a consequence, the degree of partitioning of these surfactants into liposomes (or affinity with these structures) decreased as the surfactant hydrophobic moiety decreased. Thus, the DePB molecules had the lowest affinity with SC bilayers (minimum K values), whereas the TePB showed the highest (maximum K values).

Comparison of the corresponding K_{SAT} and K_{SOL} values for each surfactant tested also reveals

Table 1
Solubilizing parametcrs of SC liposomes (treated with DcPB, DoPB and TePB cationic surfactants)

Surfactant	CMC (mM)	$S_{W,SAT}$ (mM)	$S_{W,SOL}$ (mM)	Re_m (mol/mol)	Re_m (mol/mol)	K_{SAT} (mM^{-1})	K_{SOL} (mM^{-1})	r^2 (SAT)	r^2 (SOL)
DcPB	23.5	22.8	23.1	0.46	1.27	0.014	0.024	0.998	0.997
DoPB	6.1	5.5	5.7	0.62	1.40	0.069	0.102	0.994	0.997
TePB	1.70	1.56	1.64	0.88	1.84	0.300	0.395	0.996	0.995

The CMC values of the surfactants in Pipes buffer and the regression coefficients of the straight lines obtained arc also included.

that the higher the surfactant alkyl chain length the lower the quotient between both parameters (K_{SOL}/K_{SAT}). Thus, at the interaction level for complete solubilization the degree of partitioning of surfactants into SC liposomes relatively decreased with respect to that for saturation as the alkyl chain length rose (K_{SOL}/K_{SAT} value from 1.71 for DePB to 1.31 for TePB). As a consequence, we may assume that the increase in the surfactant alkyl chain length in addition to improving the partitioning of surfactant molecules into bilayers also resulted in a relative decrease in its ability to be associated with the lipid molecules building SC liposomes to form mixed micelles. Possibly, the first order phase transition from mixed liposomes into mixed micelles appears to be relatively hampered by the increasing surfactant hydrophobic tail.

In parallel, comparison of the present K values with those reported for the interaction of the same surfactants with neutral and electrically charged PC liposomes shows that the degree of partitioning of these surfactants into SC bilayers (or bilayer affinity) was always greater (higher K values) than that reported for PC ones. However, the influence of the surfactant hydrophobic moiety in this affinity was also similar in both cases in spite of the different compositions and properties of these two bilayer structures (de la Maza and Parra, 1995a).

In general terms, different trends in the interaction of these surfactants with SC liposomes may be observed when comparing the present Re and K parameters with those reported for PC liposomes. Thus, whereas SC liposomes appeared to be more resistant to the action of surfactant monomers the surfactant partitioning into SC structures was always greater than that reported for PC ones. Thus, although a greater number of surfactant molecules were needed to produce the same alterations in SC liposomes, these molecules showed increased affinity with these bilayer structures. However, a similar influence of the surfactant hydrophobic tails on the Re and K parameters was observed for both cases.

If the Re and K values obtained for each surfactant tested are plotted as a function of its CMC's the graphs shown in Figs. 2 and 3 are obtained. A decrease in Re_{sol} and Re_{sat} occurred as the surfactant CMC rose (or the surfactant alkyl chain length decreased), this fall being more pronounced at low CMC values for both parameters.

The rise in the surfactant CMC also resulted in an abrupt fall in K both for bilayer saturation (K_{SAT}) and solubilization (K_{SOL}) especially at low CMC values (Fig. 3). As a consequence, in this CMC range the degree of partitioning of these surfactants into SC liposomes (or its affinity with these bilayer structures) drastically decreased as the surfactant CMC increased.

As a consequence, two opposite trends in the interaction of these surfactants with SC liposomes may be observed when comparing the variation of Re and K versus surfactant CMC. The increase in the surfactant CMC resulted in a rise in the ability of these surfactants to saturate or solubilize SC liposomes and inversely in an abrupt decrease in their degree of partitioning into liposomes.

Fig. 4 shows for each surfactant tested the total surfactant concentration (mM) needed to saturate (S_{SAT}, Eq. (4)) or solubilize (S_{SOL}, Eq. (5)) SC

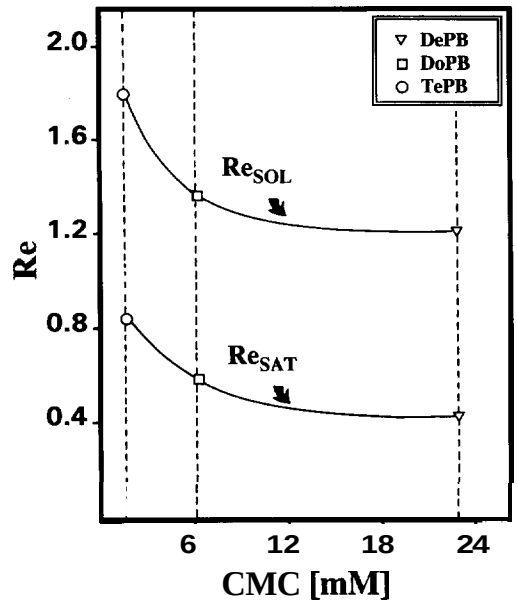

Fig. 2. Effective surfactant:lipid molar ratios Re_{sol} and Re_{sat} of [▽] DePB, [O] DoPB and [○] TePB surfactants vs. CMC of surfactants for SC liposomes.

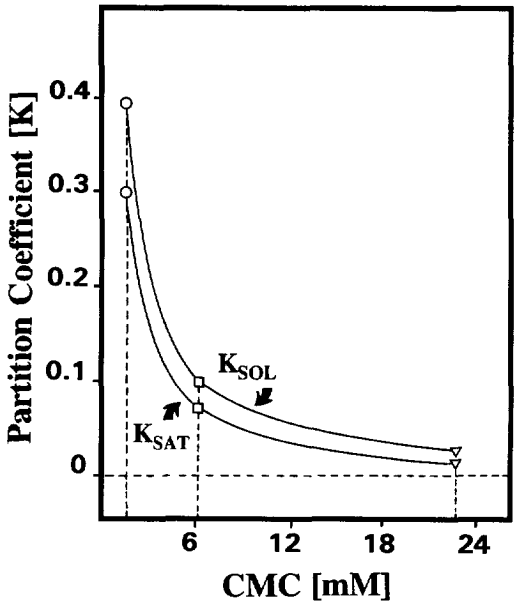

Fig. 3. Partition coefficients K_{SAT} and K_{SOL} of [▽] DePB, [□] DoPB and [○] TePB surfactants vs. CMC of surfactants for SC liposomes.

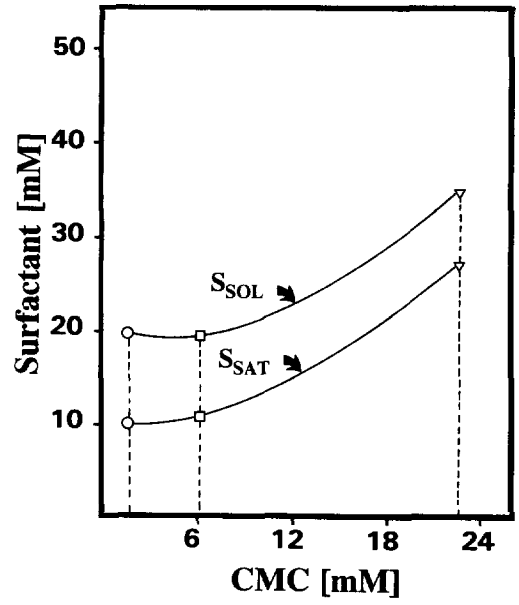

Fig. 4. Plots the total surfactant concentrations S_{SAT} and S_{SOL} of [▽], DePB, [□] DoPB and [○] TePB surfactants vs. CMC of surfactants for SC liposomes, the bilayer lipid concentration remaining constant (10.0 mM).

liposomes versus the surfactant CMC (lipid concentration 10.0 mM). It is noteworthy that although the lowest surfactant concentration for liposome saturation corresponded to the TePB, the DoPB exhibited the lowest value for solubilization of these SC bilayer structures. Thus, the overall balance of the surfactant activity on SC liposomes shows that TePB and the DoPB had respectively the highest power with respect to the saturation and solubilization of these bilayer structures. Bearing in mind these findings we may assume that the hydrophilic–lipophilic balance of each surfactants tested play an important role in its associative capacity with the lipid molecules building SC liposomes and its solubilizing power of these bilayer structures via mixed micelle formation.

The result obtained for liposome solubilization contrasts with that reported for the interaction of these susfactants with neutral PC liposomes, in which the highest power corresponded to the TePB in all cases. As a consequence, the physicochemical characteristics and composition of each liposome structure may be considered as important factors in the selective capacity of these surfactant to saturate and solubilize different bilayer structures.

We are aware of the fact that the lipids used in this work are not exactly the same as those existing in the stratum corneum. Nevertheless, the comparison of the present Re and K parameters with those reported for the same surfactants with PC liposomes may be useful in establishing a criterion for the evaluation of the activity of these surfactants in a simplified membrane model devoid of phospholipids. Our approach may be also useful in the study of the specific surfactant interactions with SC structured lipids, given the growing use of the cationic surfactants in pharmacological applications.

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