

A study of the gel structure in a nonionic O/W cream by X-ray diffraction and microscopic methods

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Abstract: The structure of a nonionic O/W cream and its separate components were investigated by X-ray diffraction and microscopic methods. The results presented in this paper are combined with the results obtained with DSC in order to elucidate these structures. The very important role of water with respect to the formation of the colloidal structures is discussed. The influence of water on the polymorphic behaviour of cetylstearylalcohol is attributed to a good structural fit of its α -modification and water. Hydration induces a tilt of the alkyl chains of poly(oxyethylene)₂₀glycerylmonostearate (PGM₂₀). For this surfactant an antiparallel arrangement of the molecules is proposed. The lamellar structures present in mixtures of cetylstearylalcohol, PGM₂₀ and water are discussed extensively.

Key words: Gel structure, wide and small angle X-ray diffraction.

Introduction

Creams are frequently used as topical administration vehicles for drugs. Usually, creams contain more than three components, but in their simplest form they consist only of water, oil and surfactant. The surfactant may be ionic or nonionic in nature. The use of nonionic surfactants has considerable advantages. Irritation of the skin is much less severe than with ionic surfactants. Furthermore, the colloidal structure in a nonionic cream will be much less affected by the addition of salts. Therefore, incompatibilities of drugs with a nonionic cream will occur less frequently than with ionic creams. These advantages make a nonionic cream more suitable as a general applicable vehicle for topical administration than a cream containing ionic surfactants.

The colloidal structures in creams determine their viscoelastic properties, which are, among others, important for their practical application. Furthermore, these structures affect the release of water and drugs from creams. Relating the viscoelastic properties of a cream, or the release of water and drugs from a cream,

with its colloidal structure requires characterization of such structures. In order to develop vehicles with optimal properties, the colloidal structures in existing formulations have to be studied first. After that, these formulations can be modified to achieve a general applicable cream with optimal properties.

Considering the above mentioned advantages of nonionic surfactants, we decided to investigate the colloidal structures in a nonionic cream: "Unguentum Hydrophilicum Nonionicum Aquosum, DAC" [1]. The composition of this particular O/W cream is:

– Poly(oxyethylene) ₂₀ glycerylmonostearate (= PGM ₂₀)	5.0% W/W
– Liquid paraffin	7.5% W/W
– Cetylalcohol	5.0% W/W
– Stearylalcohol	5.0% W/W
– Glycerol	8.5% W/W
– White soft paraffin	17.5% W/W
– Water	51.5% W/W

It may be expected that mainly the amphiphilic components PGM₂₀, cetylalcohol and stearylalcohol are responsible for the construction of the colloidal

network. Therefore, we studied systems containing these components and water. Under certain conditions these systems could be regarded as simple creams [2]. This paper is a part of series in which a detailed report of a study on the colloidal structures occurring in these creams is presented. In this study, also the separate components were investigated extensively. Moreover, a series of mixtures of polyoxyethylene 1550 and water has been investigated in order to study the interactions between the polyoxyethylene part of the PGM₂₀ molecule and water more precisely [3].

The results, obtained by several experimental techniques, were combined in order to elucidate the colloidal structures. Results obtained by differential scanning calorimetry (DSC) have been discussed earlier [2, 3]. Here, results obtained by polarization microscopy (PM), transmission electron microscopy (TEM), wide angle X-ray diffraction (WAXD) and small angle X-ray diffraction (SAXD) are presented. Some preliminary results, mainly obtained by SAXD, have been published elsewhere [4].

Measurements in which several experimental techniques were used, were carried out on samples of the same batches simultaneously. Alterations in our samples due to ageing will be discussed in a forthcoming paper.

Experimental

The components cetylalcohol (Lorol C16[®], Henkel KGaA, 90–95% cetylalcohol), stearylalcohol (Lorol C18[®], Henkel KGaA, over 95% stearylalcohol), polyoxyethylene 1550 (Polywachs 1550[®], Huels AG, mean degree of polymerization of 35), PGM₃₀ and PGM₂₀ (Tagat S and Tagat S₂ respectively, Th. Goldschmidt AG) were used without further purification. The water used was demineralized. The fatty alcohols have been derived from natural oils and fats by transesterification and reduction. Therefore, they contained not only homologous compounds, but also small amounts of esters, fatty acids and unsaturated compounds. PGM₂₀ is an industrial product which contains about 3% water. It has a molecular weight distribution, the hydrophilic chain can be branched and moreover, the glyceryl part can be attached to different C atoms in the oxyethylene chain. This is illustrated in Figure 1. Since the stearyl part of PGM₂₀ has been derived from natural fats and oils, the surfactant contains some impurities comparable with those present in cetylalcohol and stearylalcohol.

First, the components were investigated separately. Next, we studied samples containing cetylalcohol, stearylalcohol and PGM₂₀ in the ratio 1:1:1 (W/W), corresponding to the ratio of these components in the DAC formulation, and water. The water concentration was varied between 0 and 90% W/W and was determined thermogravimetrically (TG, Heraeus TGA 500 and Mettler TA 3000 system). The water loss found with TG was in good agreement with the amount of water used in preparing the samples (the maximum

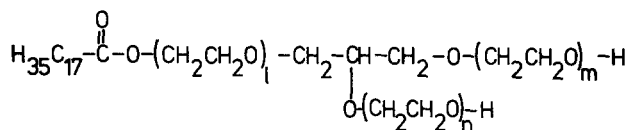


Fig. 1. Schematic representation of PGM₂₀, $l + m + n = z$, $\bar{z} = 20$

deviation was about 1% of the sample weight). Cetylalcohol, stearylalcohol and PGM₂₀ were melted and stirred at 70°C. Subsequently, water with a temperature of 70°C was added and the mixture was cooled down to room temperature under stirring. The samples were stored at room temperature in airtight glass containers. Samples containing only PGM₂₀ and water were prepared and stored in the same way. Samples under investigation were 3 weeks old. Since DSC measurements revealed a transition in PGM₂₀ at ambient temperature (23°C, [2]), samples of this surfactant were stored at –10°C for 7 days prior to experiments whereby adaptation problems could be expected.

During X-ray diffraction experiments the temperature of the samples deviated by maximum 1°C from the adjusted temperature. SAXD experiments were carried out with Ni-filtered CuK α -rays ($\lambda = 0.154$ nm). The camera (Kratky camera, A. Paar) was equipped with a one dimensional position sensitive detector (M. Braun) which was connected to a multi channel analyzer (Canberra S 40). After calibration, each channel on the multi channel analyzer corresponded to a certain diffraction angle. The use of slit collimation implied that principally the obtained SAXD spectra had to be desmeared. However, if one is only interested in the position of the intensity maxima, desmearing is not always required. The necessity of desmearing an individual SAXD spectrum depended on the degree of anisotropy of the studied sample. In turn, the anisotropy depended on the colloidal structure and the visco elastic behaviour of the sample, and therefore, also on the procedure of filling the sample holder (glass capillary, ϕ 1.5 mm). As a criterion for desmearing we used the relative intensity at diffraction angles around the maxima. If the peaks were sharp, i. e. if the intensities at angles near the maxima were relatively high, desmearing had little or no effect. On the contrary, if the intensities around the maxima were relatively high (especially at smaller angles), desmearing was necessary to derive the correct lamellar spacings from the obtained SAXD curves. If necessary, the spectra were desmeared using a method which has been described by Vonk [5]. Examples will be given in the results section.

WAXD spectra were taken with a Philips powder diffractometer (PW 1064, monochromatic CuK α -rays) equipped with a proportional detector, or with a Kiessig camera (Ni-filtered CuK α -rays) using photographic detection.

Samples for TEM were frozen in melting N₂ (–210°C, cooling rate 10⁴ to 3 × 10⁴ Ks^{–1}). The frozen samples were placed in a Balzers BAF 400 freeze-etching apparatus. After breaking (at –100°C) and etching (–100°C, 10^{–9} bar, variable etching time) the samples were shadowed with a Pt/C mixture (angle 45°) and subsequently with pure C (angle 90°). The replica were studied with a Philips EM 300 microscope.

PM studies were performed either with an Olympus BH-2 microscope, which was equipped with a Mettler FP 800 hot stage, or with a Leitz Dialux-Pol microscope.

Results

Cetylstearylalcohol

A mixture of equal weights of cetylalcohol and stearylalcohol was studied. A SAXD curve is shown in Figure 2 a. The intensity maxima have the spacing ratios 1/1 to 1/2 to 1/3, indicating that these maxima correspond to the first, second and third order peaks of a sample with a lamellar structure respectively. The calculated thickness of the lamellae is 4.7 nm. After desmearing the maxima are hardly shifted compared with the smeared curve and the calculated repeating distance is again 4.7 nm. In Figure 2 b the desmeared first order peak is depicted. This case is a typical example illustrating that if one is only interested in the

position of the intensity maxima, desmearing is not always necessary. A PM photograph (Fig. 3 a) illustrates the lamellar nature of cetylstearylalcohol at 20°C. A WAXD spectrum is given in Figure 4 a. The diffraction lines at $2\theta = 24.3^\circ$ and $2\theta = 24.1^\circ$ correspond with Bragg spacings of 0.366 nm and 0.415 nm respectively.

A sample of cetylstearylalcohol containing 20% W/W water showed, at 20°C, only a reflection at $2\theta = 21.6^\circ$.

PGM₂₀

PM showed that at 21°C PGM₂₀ had a lamellar structure (Fig. 3 b). In Figure 2 c a SAXD curve of

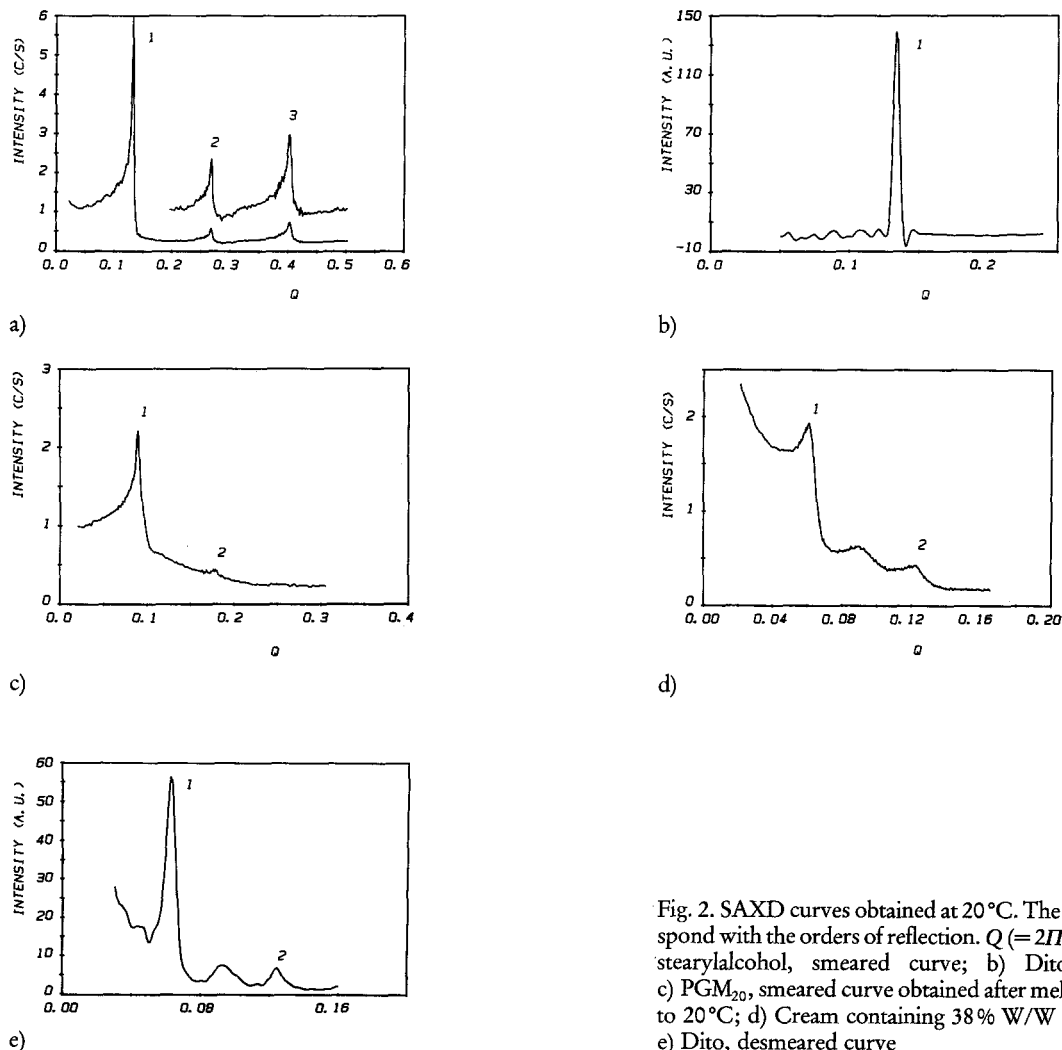


Fig. 2. SAXD curves obtained at 20°C. The arabic numerals correspond with the orders of reflection. $Q (= 2\pi/d)$ in 10/nm. a) Cetylstearylalcohol, smeared curve; b) Dito, desmeared curve; c) PGM₂₀, smeared curve obtained after melting and cooling down to 20°C; d) Cream containing 38% W/W water, smeared curve; e) Dito, desmeared curve

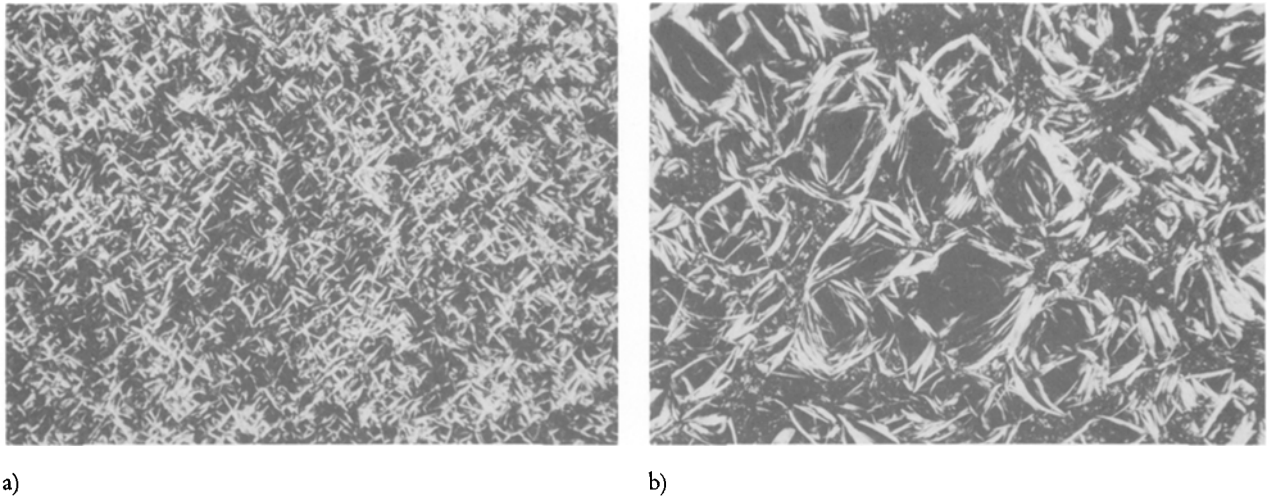


Fig. 3. PM photographs of a) Cetylstearylalcohol at 20°C; b) PGM₂₀ at 21°C

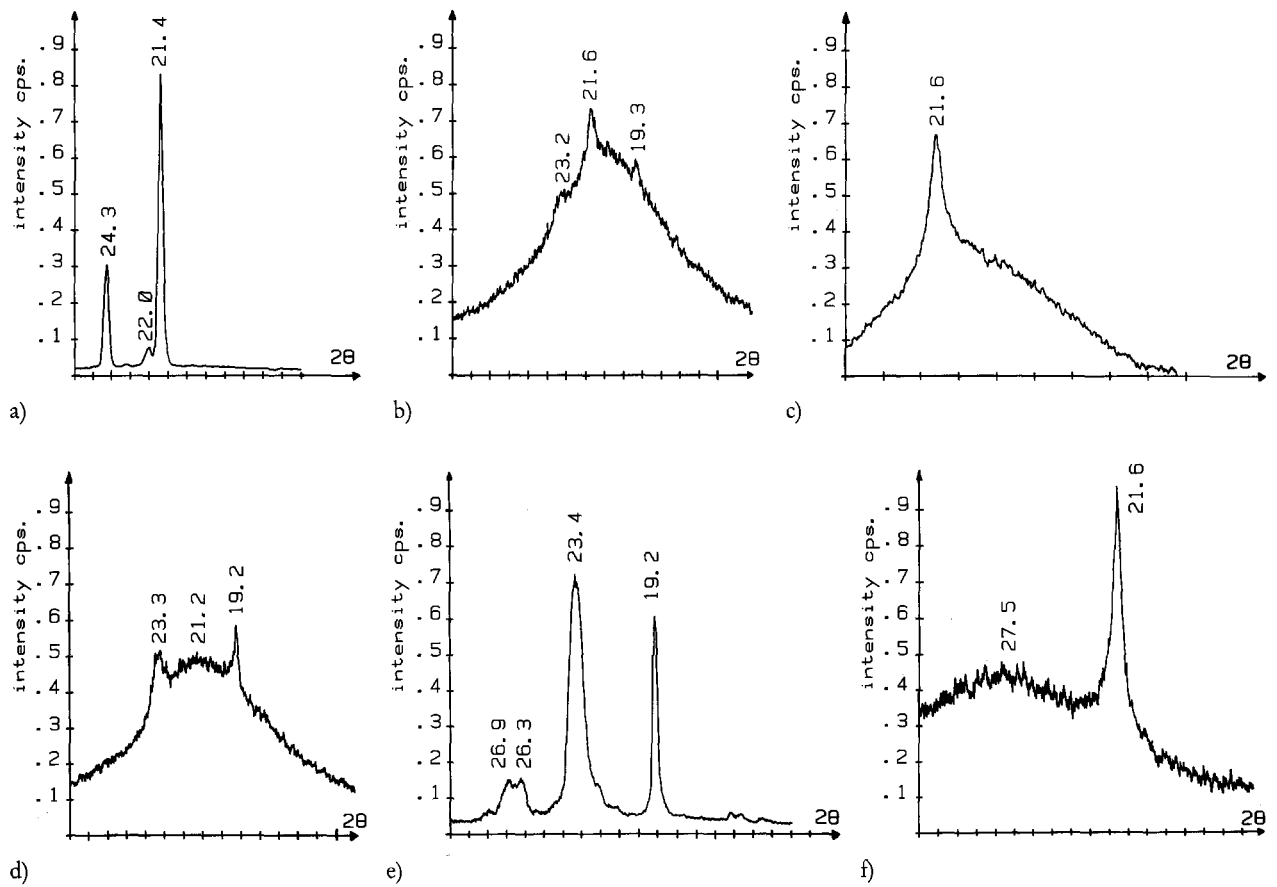


Fig. 4. WAXD curves obtained at 20°C. The numbers at the maxima correspond with the diffraction angles 2θ (in degrees). Full scale intensities (in cps) between parenthesis: a) Cetylstearylalcohol (10^4); b) PGM₂₀, stored at room temperature (10^3); c) Dito, after melting and cooling down (10^3); d) PGM₃₀ (10^3); e) Polyoxyethylene 1550 (2×10^3); f) Cream containing 83% W/W water (4×10^2)

PGM₂₀ is depicted. Prior to this measurement the sample was heated above its melting point (31 °C) and was then allowed to cool down to 20 °C. From the first and second order maxima a lamellar spacing of 7.1 nm is calculated. SAXD at 15 °C, after storage at -10 °C, showed longer distances next to 7.1 nm. WAXD spectra of PGM₂₀ are shown in Figures 4 b and 4 c. Both spectra were taken at 20 °C. Figure 4 b belongs to a sample of PGM₂₀ which was stored at room temperature. Figure 4 c is derived from a sample which was melted first and subsequently cooled down to 20 °C prior to the WAXD experiment. Both spectra show a peak at $2\theta = 21.6^\circ$. However, in Figure 4 b additional peaks (shoulders) are visible at $2\theta = 23.2^\circ$ and $2\theta = 19.3^\circ$. In Figures 4 d and 4 e WAXD spectra of PGM₃₀ (30 oxyethylene units) and polyoxyethylene 1550, both at 20 °C, are depicted. Figure 4 d (PGM₃₀) shows maxima at $2\theta = 23.3^\circ$, $2\theta = 21.2^\circ$ (diffuse) and $2\theta = 19.2^\circ$. In Figure 4 e the positions of the main intensity maxima are at $2\theta = 23.4^\circ$ (0.380 nm) and $2\theta = 19.2^\circ$ (0.462 nm).

Samples containing PGM₂₀ and water can be divided in 3 regions. The classification is based on PM and DSC results [2]. Samples containing 0 to $\approx 37\%$ W/W water (domain I) consist of a lamellar solid phase. In domain II ($\approx 37\%$ to $\approx 82\%$ W/W water) a lamellar solid phase and an optically isotropic liquid phase coexist. The amount of liquid phase increases with increasing water concentration. Samples containing over 82% W/W water (domain III) only consist of optically isotropic liquid. On addition of water the lamellar thickness increases. The calculated repeating

distances are plotted in Figure 5 as a function of the ratio water/surfactant. This way of plotting the long spacings should yield a straight line if all water molecules contribute to swelling of the lamellae and the interfacial area does not change (see Eq. (1) below). Samples containing less than 37% W/W water showed strong reflections. The peaks in SAXD spectra of samples containing over 37% W/W water gradually become more diffuse with increasing water content. In samples containing over 60% W/W water no sharp SAXD reflections are visible. After centrifugation the clear supernatant of a sample originally containing 60% W/W water showed a broad SAXD peak. A repeating distance of about 5.8 nm was calculated from the desmeared curve. Samples of PGM₂₀ containing 13% W/W or more water showed, at 20 °C, only a WAXD peak at $2\theta = 21.6^\circ$.

Mixtures of cetylalcohol, stearylalcohol, PGM₂₀ and water

The studied samples contained equal weight fractions of cetylalcohol, stearylalcohol and PGM₂₀. This ratio corresponds to the ratio of these components in the DAC formulation [1]. The water concentration is varied between 0 and 90% W/W. Below 20% W/W water the samples are solid like [2, 4]. Above 20% W/W water the samples gradually become softer with increasing water concentration. Above 75% W/W water phase separation is perceptible.

PM indicated that samples of this series had a lamellar structure. The lamellar nature is illustrated with an EM photograph of a sample containing 60% W/W water (Fig. 6). In samples containing less than 20% W/W water several lamellar phases coexist. With microscopic methods no inhomogeneities were observed in samples containing 20 to 75% W/W water.

With SAXD several long distances were found in samples containing less than 20% W/W water. At 0.7% W/W water distances of 4.7, 6.4 and 8.9 nm were observed. Increasing the water concentration reduced the relative intensity of the 4.7 nm peak. The distance increased from 4.7 nm to 4.9 nm at 15% W/W water. Between 15 and 25% W/W water the corresponding peak vanished. On addition of water the distance of 6.4 nm increased to ≈ 7.0 nm at 15% W/W water. This distance remained constant between 15 and 50% W/W water. The intensity of the peak was low and between 35 and 50% W/W water the peak was hardly visible. In samples containing over 50% W/W water the 7.0 nm peak was not observed. The

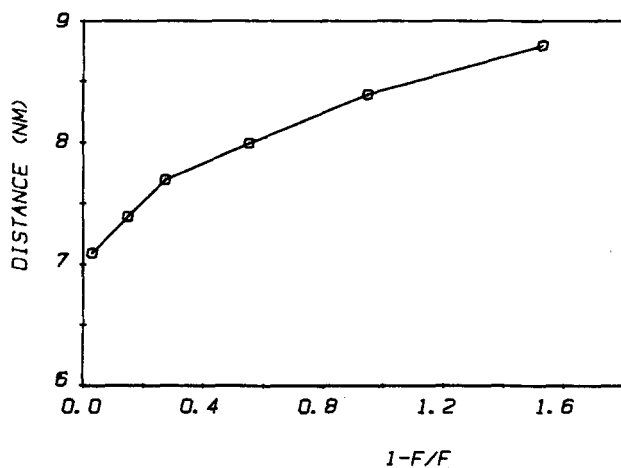


Fig. 5. Long spacings in mixtures of PGM₂₀ and water at 20 °C vs. the ratio water/surfactant. F is the weight fraction of surfactant

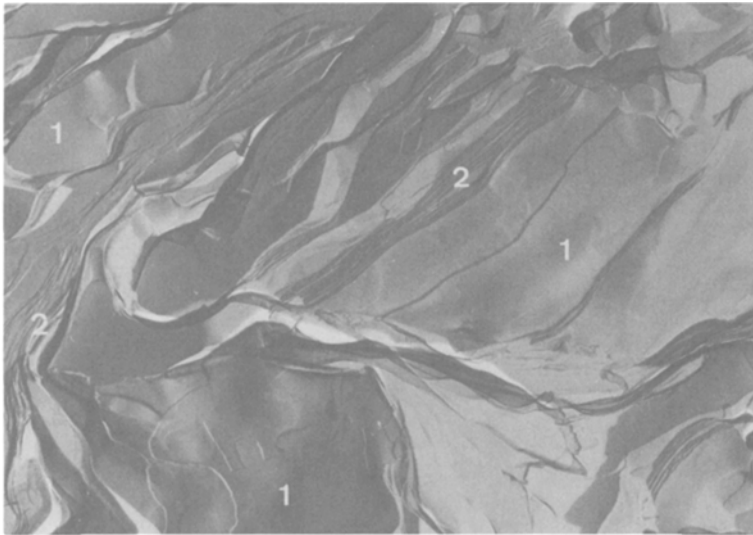


Fig. 6. EM photograph of a cream containing 60% W/W water. The lamellar spacing is about 12 nm; 1 = lipophilic sheet; 2 = section perpendicular to the plane of the lamellae

distance of 8.9 nm varied between 8.9 and 8.2 nm in the range 0–20% W/W water.

Besides the distance of 7.0 nm related to the weak peak mentioned above, only one long distance is found in samples containing 20 to 75% W/W water. The repeating distance can be calculated from a strong first order peak and a weak second order peak. A smeared SAXD curve of a sample containing 38% W/W water is shown in Figure 2 d. The desmeared curve is depicted in Figure 2 e. In this case, in contrast with cetylstearylalcohol (Figs. 2 a and 2 b), desmearing influenced

the position of the maxima. A long distance of 10.4 nm was derived from the smeared curve. After desmearing the calculated distance was 10.0 nm. Other examples of smeared SAXD curves can be found in [4]. In Figure 7 the long spacings derived from the smeared and desmeared curves are compared. Again, the spacings are plotted versus the ratio water/surfactant.

WAXD spectra of samples containing 20 to 75% W/W water showed only a peak at $2\theta = 21.6^\circ$ (0.411 nm). A very diffuse additional peak, at $2\theta = 27.5^\circ$ (0.324 nm), is found in spectra of samples containing over 75% W/W water. An example of these spectra is given in Figure 4 f. A sample containing 1.0% W/W water had a WAXD spectrum comparable with the spectrum of cetylstearylalcohol (Fig. 4 a). The spectrum of a sample containing 13.1% W/W water had maxima at $2\theta = 22.6, 21.6$ and 21.3° .

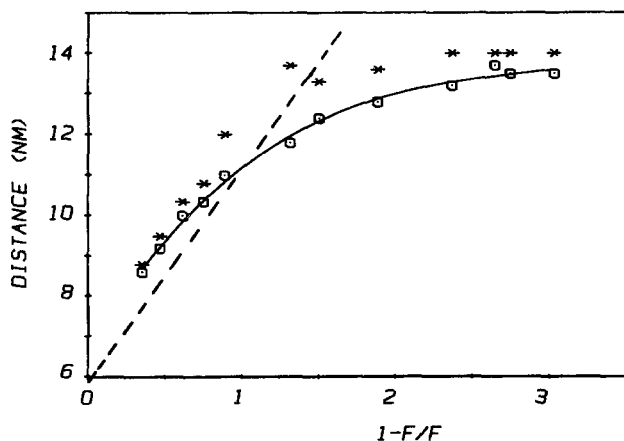


Fig. 7. Longs spacings in mixtures of water and equal weights of cetylalcohol, stearylalcohol and PGM₂₀ at 20 °C, vs. the ratio water/surfactant. F is the weight fraction of surfactant (= dry gel). * = Derived from smeared curve; O = Derived from desmeared curve. The dotted line is explained in the text

Discussion

Cetylstearylalcohol

The polymorphism of cetylalcohol, stearylalcohol and their mixtures has been studied by Beer [6]. DSC experiments showed that the impurities present in the batches of cetylalcohol and stearylalcohol that we used did not influence the polymorphic behaviour of their mixtures in a wide composition range [2]. Two modifications of cetylstearylalcohol are of importance in our studies. According to Precht [7] these modifications are called the β_0 - and the α -modification. In both the

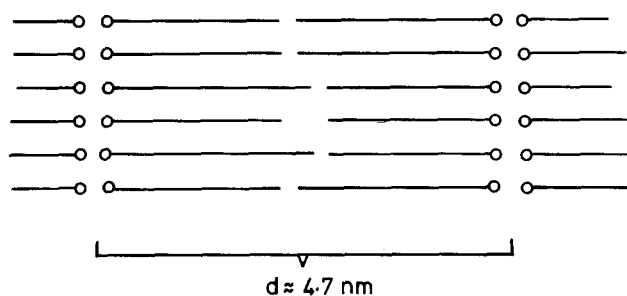


Fig. 8. Lamellar structure of a mixture containing equal weights of cetylalcohol and stearylalcohol. \circ = hydrophilic head; — = hydrocarbon tail

α - and β_0 -modification the hydrocarbon chains are placed perpendicular to the lattice plane, forming bilayers (Fig. 8). The β_0 -modification has a nearly orthorhombic unit cell [7]. The α -modification is a plastic crystal in which the hydrocarbon chains are placed in a hexagonal array. Both phases have a solid character. In the β_0 -modification the alkyl chains are closely packed. The compressibility is low [8]. In the α -modification the hydroxyl groups are hexagonally packed in one plane, resulting in a relatively high compressibility.

At 20°C cetylstearylalcohol possesses the β_0 -modification [2, 9, 10]. The lamellar structure was confirmed by PM and SAXD. The lamellar spacing of 4.7 nm, derived from the SAXD spectra (Figs. 2 a and 2 b), is in good agreement with the values of 4.8–4.9 nm found by Führer et al. [10] and 4.7 nm reported by Junginger et al. [9]. The wide angle reflections at $2\theta = 24.3$ and $2\theta = 21.4^\circ$ belong to the 200 and 110 lattice planes of the orthorhombic unit cell of long chains paraffins [10]. At 31°C and 50°C the structure of cetylstearylalcohol changes from the β_0 - to the α -modification and from the α -modification to the liquid state respectively [2].

Incorporation of water lowers the β_0 to α transition temperature and increases the melting point [2, 11]. At 20°C the hydrocarbon chains of the formed hydrate exhibit the α -modification. This is confirmed by the WAXD results which showed only a reflection at $2\theta = 21.6^\circ$ for hydrated cetylstearylalcohol at 20°C, this reflection angle being characteristic for the α -modification [9]. From the diffraction angle the distance between the hexagonally packed alkyl chains is calculated: $d = \lambda/(\sqrt{3} \sin \theta) = 0.475$ nm. The cross-sectional area of the chains (S), calculated from the reflections, amounts to 0.195 nm². It is known that S is most sensi-

tive to packing [12], and that the packing as well as the orientation of long chain normal aliphatic compounds at interfaces vary with the nature of the polar groups [13].

In this particular case we can easily understand why hydration induces the transition from the β_0 - to α -modification at ambient temperature. The explanation goes as follows: from the density of water at 20°C (0.988 g/cm³) the volume of one water molecule is calculated (30.25×10^{-3} nm³). Considering the water molecule as a cube, the “cross-section area” of this cube is 9.71×10^{-2} nm². Assuming the structure of hydration water to be an ice like structure (Ice I, i. e. hexagonal ice) the cross-sectional area of this cube is 10.18×10^{-2} nm². Since the cross-sectional area of the water cube is about half the S -value of the hydrocarbon chains, it is very well conceivable that the hydroxyl groups at the interfaces become part of the water structure on hydration. On closer examination of the water structure, the compatibility with the hydroxyl groups of cetylstearylalcohol in the α -modification is clear. We know from WAXD experiments that the mean distance between the hydroxyl groups is 0.475 nm. The O—O—O angle of water molecules having a tetrahedral coordination is 109.47° . The calculated O—O distance in the hydroxyl group/water complex, resulting from substitution of part of the water molecules by hydroxyl groups, is 0.291 nm ($= (0.5 \times 0.475 \text{ nm})/\sin(0.5 \times 109.47)$). The O—O distance of liquid water is 0.282 nm at 4°C and 0.294 nm at 200°C [14]. We therefore conclude that the hydroxyl groups of cetylstearylalcohol possessing the α -modification fit very well in the water structure and that this compatibility causes the β_0 to α transition on hydration at ambient temperature. The hydroxyl groups replace half of the water molecules in the first hydration layer.

PGM_{20}

At 20°C wide angle reflections were present at $2\theta = 21.6^\circ$, $2\theta = 23.2^\circ$ and $2\theta = 19.3^\circ$ (Fig. 4 b). From the WAXD spectrum of polyoxyethylene 1550 (Fig. 4 e) it is obvious that the two latter reflections originate from crystalline polyoxyethylene domains in this polydisperse surfactant. These reflections are in agreement with values reported for other surfactants derived from polyoxyethylene [4, 15, 16]. The diffraction angle of $2\theta = 21.6^\circ$ originates from the hydrocarbon chains possessing the α -modification. PM (Fig. 3 b) and SAXD (Fig. 2 c) revealed that PGM_{20} had a lamellar structure. The repeating distance was 7.1 nm. Com-

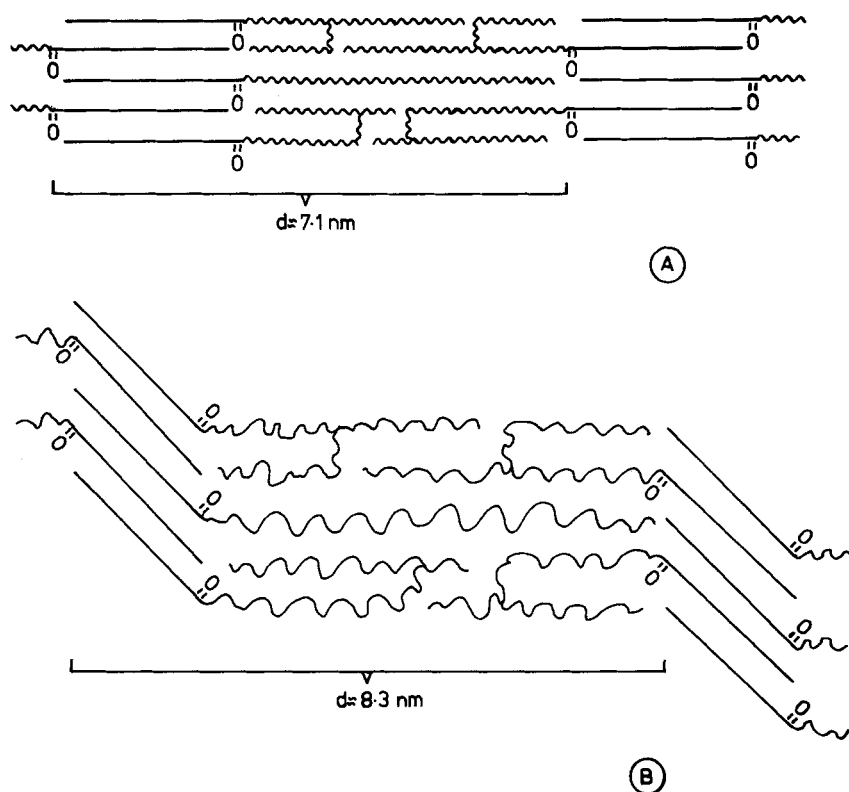


Fig. 9. A) Lamellar structure of PGM₂₀ (20°C); B) Lamellar structure of a mixture containing PGM₂₀ and 37% W/W water (20°C): \sim = polyoxyethylene chain; \sim = hydrated polyoxyethylene chain; — = hydrocarbon chain

binning these findings with commonly observed values for densities and molecular sizes leads to a picture shown in Figure 9 A. This will now be discussed. For a linear PGM₂₀ molecule a length of 8.3 nm is calculated using 0.126 nm, 0.278 nm and 0.5 nm for the lengths of the methylene unit, the oxyethylene unit and the glyceryl unit respectively. The measured repeating distance (7.1 nm), which besides includes the spacings between the terminal groups and the space occupied by the 3% W/W water, is considerably smaller than the calculated surfactant length. However, the surfactant has a molecular weight distribution, the hydrophilic chain can be branched and will be partly in the liquid state [2]. Therefore, 7.1 nm is a realistic value for the average length of a PGM₂₀ molecule. From this value we notice that the lamellar structure can not be composed of bilayers. We suggest an antiparallel arrangement of surfactant molecules, as shown in Figure 9 A. A parallel arrangement of branched surfactant molecules is unlikely, because this would lead to distortion of the hydrocarbon structure, which is crystalline, as shown by DSC measurements [2]. An antiparallel arrangement leaves the possibility for the hydrocarbon

chains to adapt the α -modification. Furthermore, in this arrangement the hydrophilic chains are able to fill up the space between the hydrocarbon sheets in such a way that a most compact hydrophilic layer is formed, whether the chains are branched or unbranched. A quantitative elaboration of the model depicted in Figure 9 A will be given now. From the structural formula of the surfactant (Fig. 1) and the densities of the hydrocarbon chains (ρ_{hc}) and the hydrophilic chains (ρ_{oe}) the lipophilic/hydrophilic volume ratio is calculated. The ratio 1 : 2.26 is derived using 0.8 g/cm^3 for ρ_{hc} and 1.2 g/cm^3 for ρ_{oe} . From this ratio and the repeating distance (7.0 nm for the dry surfactant, derived from Fig. 5) a length of 2.15 nm is calculated for the hydrocarbon chains. The length of a linear chains of 18 methylene units theoretically is 2.27 nm. Neglecting the terminal groups, we may conclude that the agreement between the calculated and the theoretical length is fairly good. We propose that the model of antiparallel arrangement is correct because it fits very well with the experimental findings.

In the WAXD spectrum of PGM₃₀ the polyoxyethylene reflections dominate (Fig 4 d). In the spectrum

of PGM₂₀ the main reflection ($2\theta = 21.6^\circ$) originates from the hydrocarbon chains (Fig. 4 b). The ratio of the intensities at $2\theta = 21.6^\circ$ (Figs. 4 b and 4 d) is equal to the ratio of the hydrocarbon weight fractions of PGM₂₀ and PGM₃₀. The intensities of the oxyethylene reflections strongly depend on the thermal history of PGM₂₀. On cooling molten PGM₂₀ to 20 °C the hydrocarbon chains, which melted at 31 °C, solidify quickly. At 20 °C the crystallization of the oxyethylene chains is slow. This effect can be seen in Figure 4 c. In this spectrum the polyoxyethylene reflections for the most part vanished. This can be understood from DSC measurements, where it was found that the polydisperse polyoxyethylene chains melted over a wide temperature range and on heating the last crystalline polyoxyethylene domains melted at 23 °C [2]. This is a reasonable temperature when compared with the melting points found for pure polyoxyethylenes with molecular weights of about 1000 [17–19]. It also agrees with the fact that in the bulk of surfactants derived from polyoxyethylene, crystallization of the polyoxyethylene chain at ambient temperature is observed at a degree of polymerization of 15 to 20 [16]. SAXD at 15 °C showed longer distances next to 7.1 nm. This is due to crystallization of larger polyoxyethylene domains (see Figs 5 and 6 in [2]) resulting in distortion of the compact hydrophilic layer. At 20 °C PGM₂₀ can be considered as a gel, the liquid parts of the surfactant being held in a network of crystalline hydrocarbon chains.

In the results section we mentioned the division in 3 domains of samples containing PGM₂₀ and water. Now we will discuss the properties of samples in the 3 domains separately. Samples of domain I (0 to $\approx 37\%$ W/W water) had sharp SAXD reflections which showed that the lamellar spacing increased with increasing water concentration (Fig. 5). If all water molecules contribute to swelling of the lamellae and the structure of the lipophilic sheets (Fig. 9 A) does not change, the increase should be directly proportional to the ratio water/surfactant. Obviously, this kind of swelling cannot account for the amount of water incorporated in the samples of domain I. Since no separation of liquid is observed and extreme densities of the hydrophilic layer are excluded, there must be an increase of the lipophilic/hydrophilic interface. From the diffraction angle $2\theta = 21.6^\circ$ the distance between the hydrocarbon chains, which are placed in a hexagonal array, is calculated: $d = \lambda(\sqrt{3} \sin \theta) = 0.475$ nm. WAXD experiments showed that on addition of water this distance remained constant. Therefore, the

increase of the interface is caused by an increasing tilt of the stearyl chains with respect to this interface. This is illustrated in Fig. 9 B. The tilt angle is the angle between the hydrocarbon chain axis and the normal to the interface. The tilt is induced by hydration. Hydration increases the diameter of the hydrophilic chains. The polyoxyethylene chains are forced apart and the lipophilic/hydrophilic interface increases. This leads to a shift of the stearyl chains with respect to each other and therefore, to a tilted structure. For the calculation of the tilt at 37% W/W water the following parameters are used: ρ_{hc} (0.8 g/cm³), ρ_{oe} (1.2 g/cm³), the density of water ρ_w (1.0 g/cm³), the distance between the hydrocarbon chains (0.475 nm), the thickness of the lamellae at 0% and 37% W/W water (7.0 nm and 8.2 nm respectively) and the length of the hydrocarbon chains (2.15 nm). The negative excess volume of mixtures of polyoxyethylene and water is small (maximum < 3% [20]) and is therefore neglected. The calculated tilt of 44° corresponds with a mean shift of the stearyl chains of 3.65 methylene groups. The shift reduces the melting enthalpy of the hydrocarbon sheets. This can be seen from Figure 9 B and Figure 10. Plane A in Figure 10 is comparable with the plane of the paper in Figure 9 B. The central hydrocarbon chain (0) is surrounded by 6 neighbours (1–6). The calculated mean shift of the central hydrocarbon chain with respect to chain 4 was 3.65 methylene groups at 37% W/W water. The shift with respect to chain 1 is –3.65 methylene units. The shifts of chain 0 with respect to chains 3 and 5, respectively 2 and 6, are 0.5×3.65 and 0.5×-3.65 methylene units. We can see from Figure 9 B, that regarding an individual hydrocarbon chain, the situations at the

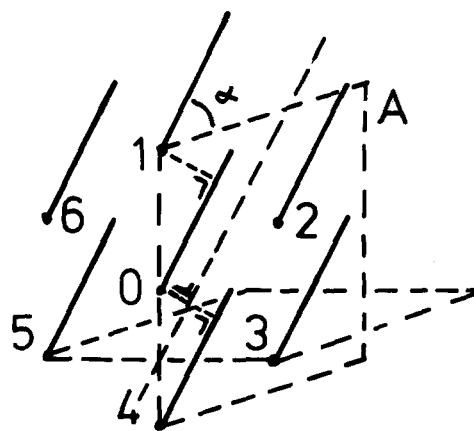


Fig. 10. Tilted hydrocarbon chains in PGM₂₀/water mixtures. α is the tilt angle. A and the numerals 0–6 are explained in the text

two interfaces are complementary. So, if the interactions between the terminal methylene groups have a crystalline character, the melting enthalpy should reduce with about 14% ($((3.65 + 0.5 \times 3.65 + 0.5 \times 3.65)/(3 \times 18)) \times 100\% = 13.5\%$). Performing this calculation we assume that the carboxyl groups lie within the hydrocarbon layer. However, it is likely that the carboxyl groups are hydrated and stick out of the hydrocarbon sheets. If, for instance, in the untilted situation. (Fig. 9 A) only 16 methylene units overlap, the decrease should be 15%. Another assumption is that the water-hydrocarbon interactions do not influence the mutual hydrocarbon interactions and that the latter interactions retained a crystalline character. If, as a result of the shift, the terminal groups are in the molten state, the decrease in the melting enthalpy should be twice the calculated value. Anyway, starting from 16 overlapping methylene units, we should find a decrease of 15–30%. The decrease found with DSC was $21 \pm 5\%$ (see Fig. 8 in [2]). This value agrees well with the value calculated above and therefore, it supports our tilt model.

At 37% W/W water the basic hydration of the hydrophilic chains, 2 water molecules per oxyethylene unit, is completed [2, 3]. The cross-sectional area of the alkyl chains increased from 0.195 nm² to 0.272 nm². However, excess water still contributes to the hydration of the polyoxyethylene chains [2, 21]. So, on further addition of water (domain II) the volume of the hydrophilic groups increases once more. Because at 37% W/W water the maximum tilt is reached, the increase of the interface can not be compensated by a further tilt and the stearyl chains are forced apart. The hydrocarbon chains get into the liquid state and the lamellar structure breaks down. This agrees with the disappearance of the SAXD peak related to this lamellar structure. The decrease of the crystalline fraction is also supported by the findings of Dorset [22], who observed a disappearance of the wide angle X-ray reflections of the alkyl chain moiety on hydration of alkylethyleneoxides. SAXD measurements indicate that the separated optically isotropic liquid phase in samples of domain II is probably a concentrated micellar solution. There is a strong resemblance between the obtained SAXD curves and curves found for samples containing a liquid L_2 -phase (components: water/sodium octanoate/1-decanol, obtained by SAXD [23]) and for samples containing calamitic or discotic aggregates (components: D₂O/sodium decyl sulphate/1-decanol, obtained by X-ray and neutron diffraction [24]). Our results do not permit a statement concern-

ing the shape of the micelles. A repeating distance of 5.8 nm was found. The fraction of micellar solution increases with increasing water concentration. At 82% W/W water only micellar solution is left and the lamellar phase has dissolved completely [2].

Addition of more water (domain III) leads to dilution of the micellar solution and possibly to alterations in the size and the shape of the micelles.

Mixtures of cetylalcohol, stearylalcohol, PGM₂₀ and water

In the results section the samples of this series were divided in 3 categories: samples containing (1) less than 20% W/W water, (2) 20–75% W/W water and (3) over 75% W/W water. Samples of each category contained at least one lamellar phase. Due to the small amount of water present in the PGM₂₀ batch used at the preparation, mixtures of cetylstearylalcohol and PGM₂₀, (to which no water was added), still contained 0.7% W/W water.

At 0.7% W/W water 3 lamellar phases coexist:

(1) A phase containing an "ideal" ratio unhydrated PGM₂₀ and cetylstearylalcohol. This mixture is relatively rich in PGM₂₀ and accounts for the distance of 8.9 nm. For experiments which will be discussed in a forthcoming paper concerning the ageing of the colloidal systems, it followed that the "ideal ratio" depended on the water concentration.

(2) A small amount of a mixture containing hydrated PGM₂₀ molecules and cetylstearylalcohol. This phase is relatively rich with cetylstearylalcohol molecules. The corresponding spacing is 6.4 nm.

(3) Excess cetylstearylalcohol, showing the characteristic distance of 4.7 nm.

By increasing the water concentration, more PGM₂₀ molecules are hydrated. Consequently, a greater amount of cetylstearylalcohol-rich phase is present. Between 0.7 and 20% W/W water this phase is formed at the cost of PGM₂₀-rich phase, water and cetylstearylalcohol. Due to the decrease in the amount of cetylstearylalcohol, the intensity of the 4.7 nm peak is reduced. The increase from 4.7 to 4.9 nm is the result of the formation of a hydrate of cetylstearylalcohol. A water layer of 1 molecule thickness is built in between the hydroxyl groups of the bilayers [11]. On addition of water the cetylstearylalcohol-rich phase swells and the spacing of 6.4 nm increases to 7.0 nm. The swelling is the result of a stronger hydration of the polyoxyethylene chains. At 20% W/W water the basic hydration of all polyoxyethylene chains is completed [2, 3] and

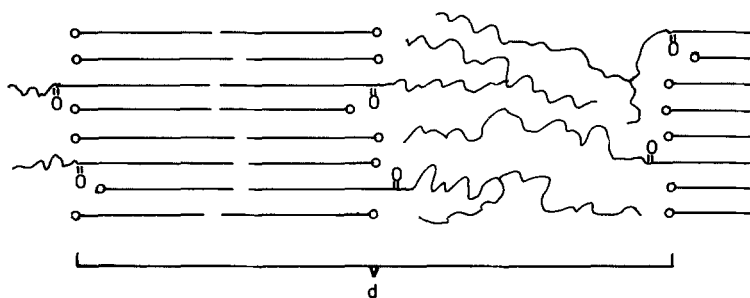


Fig. 11. Lamellar structure of a cream containing cetylstearylalcohol, PGM₂₀ and water. O = hydrophilic head; ⋈ = hydrated polyoxyethylene chain; — = hydrocarbon chain; *d* = lamellar spacing

all cetylstearylalcohol molecules are distributed among the PGM₂₀ molecules, i. e. only one lamellar phase is left. This is in agreement with either DSC heating curves [2] and WAXD results, both showing only one peak in the range 20–75 % W/W water.

The structure of this lamellar phase, present in samples belonging to region II, is depicted in Figure 11. This shows hydrophilic layers consisting of the glycerylpolyoxyethylene chains, water and hydroxyl groups, alternate lipophilic sheets. These lipophilic sheets are bilayers formed by the hydrocarbon chains of PGM₂₀ and cetylstearylalcohol. WAXD results indicate that all hydrocarbon chains of samples containing more than 20 % W/W water exhibit the α -modification. This agrees with DSC results [2] from which it was concluded that these hydrocarbon chains were in the crystalline state, possessed the α -modification and melted at 59 °C.

The density of the gel structure (i. e. bilayers + polyoxyethylene chains) can be calculated from the structural formulas of cetylalcohol, stearylalcohol and PGM₂₀, the ratio of these components in the studied samples and using the before mentioned densities ρ_{hc} and ρ_{oe} . Performing this calculation, a value of 0.9 g/cm³ is obtained for ρ_g , the density of the gel structure. Now having a value for ρ_g , we are able to calculate the overall lamellar spacing *d* at 0 % W/W water (*d*₀). Obviously, this is only a theoretical thickness, since at 0 % W/W water the structure outlined in Figure 11 does not exist. Using a bilayer thickness of 4.8 nm, a value of 5.9 nm is obtained for *d*₀. If all water molecules contribute to the swelling of the gel structure and the interfacial area remains constant, the following equation holds:

$$\frac{d_x - d_0}{(1 - F_x)/F_x} = \frac{\rho_g \times d_0}{\rho_w} \quad (1)$$

where *d*_{*x*} is the distance at *x* % W/W water and *F*_{*x*} is the weight fraction of dry gel. The dotted line in Figure 7

represents Equation (1). According to this equation, at 18 % W/W water, which is the minimum amount of water required to retain the proposed gel structure, the measured distance should be 7.0–7.1 nm. In the results section, it is shown that throughout the range 20–50 % W/W water this spacing is observed.

The lamellar structure shown in Figure 11 is capable of holding more than just 18 % W/W water between the lipophilic sheets. On addition of more water the structure swells and the overall lamellar spacing *d* increases. This spacing was measured with SAXD and the results are plotted in Figure 7. The intensity of the 7.0 nm peak decreased rapidly between 20 and 50 % W/W water, but it indicates that initially a small part of the gel containing 18 % W/W water remains unswollen. Consequently, the other lamellae swell more than predicted by Equation (2) and the measured distances are longer than expected, as can be noticed from Figure 7. The amount of unswollen lamellae rapidly decreases on further addition of water and at 50 % W/W water all lamellae are swollen. This is confirmed not only by SAXD results, but also by DSC results, the melting peak at 59 °C becoming sharper with increasing water concentration. An example of the ratio swollen/unswollen lamellae is given now. From the SAXD data and Equation (1) it is calculated that at an overall water concentration of 31.7 % W/W, 76 % of the lamellae are swollen and only 24 % did not swell. The swollen lamellae contain 36 % W/W water (*d* = 9.2 nm). As mentioned before, the unswollen lamellae contain 18 % W/W water (*d* = 7.0 nm). It is assumed that at this water concentration all water molecules are located between the lamellae.

From Figure 7 it is clear that the increase of *d* is not directly proportional to the ratio of water/(dry gel), but instead the curve reaches a plateau value. In this case, in contrast with PGM₂₀, addition of water does not induce a tilt of the hydrocarbon chains. This was concluded from DSC measurements, which showed

that the crystallinity of the hydrocarbon chains was not affected on addition of water, i. e. the melting enthalpy per unit mass of hydrocarbon chains remained constant [2]. Also lateral swelling is hardly possible because WAXD experiments show only the reflection related to the α -modification of the hydrocarbon chains. Besides, lateral swelling would influence the crystallinity of these chains. This behaviour can be understood by considering the structure of the gel. Here, only 1 out of 11 alkyl chains in the bilayers belong to a PGM₂₀ molecule, leaving enough space between the polyoxyethylene chains for expansion of hydration shells. For this reason the hydrophilic chains are not forced apart and the hydrophilic/lipophilic interface is not increased on addition of water. Since the structure of the lipophilic sheets does not change and the curve in Figure 7 levels off with increasing water concentration, our conclusion must be that part of the added water is situated outside the lamellae, especially at high water concentrations. Between 20 and 75 % W/W water, the excess water could not be visualized with microscopic methods. It probably fills up cavities between stacks of lamellae and is in equilibrium with interlamellar water. The plateau distance that was found with SAXD is 13.6 nm. This spacing corresponds with an interlamellar water content of 59 % W/W. So, at an overall water concentration of 75 % W/W, the amount of extralamellar water is 16 % W/W.

Due to the particular structure of the stacks of lamellae, this is probably the maximum amount of water filling up the cavities, since on further addition of water the lamellar phase disperses in excess water. Phase separation is perceptible and the WAXD patterns show a diffuse peak at $2\theta = 27.5^\circ$ (Fig. 4 f). This reflection originates from bulk water [14, 25]. The absence of this peak at low water concentrations is deceptive. The peak of a small amount of water is buried in the noise caused by the large amount of dry material. The appearance of the water peak at high water concentrations could be strengthened by surface effects, because the obtained WAXD pattern results mainly from the layer near the surface of the sample. This is inherent in the applied method. Perhaps separated water is adsorbed preferentially at the surface, increasing the local water concentration and consequently increasing the intensity of the reflection at $2\theta = 27.5^\circ$.

The cross-sectional area (S) of the hydrocarbon chains in the bilayers of the gel can be calculated from:

$$S = \frac{2 \times \bar{M}_w}{d_0 \times \rho_g \times N_A} \quad (2)$$

The mean molecular weight \bar{M}_w (= 347) is calculated from the molecular weights of cetylalcohol (= 242), stearylalcohol (= 270) and PGM₂₀ (= 1241), and the ratio of these components in the gel. The value found for S is 0.217 nm^2 , a high value compared with the 0.195 nm^2 calculated from WAXD experiments. The latter value is derived more straightforward and therefore more reliable. It is generally accepted that the minimum cross-sectional area of a paraffin chain is 0.185 nm^2 [8, 12, 16, 26, 27]. The cross-sectional area of the alkyl chain in the β_0 -modification of cetylstearylalcohol is 0.187 nm^2 [28]. The S -value in the α -modification found from our WAXD results is 0.195 nm^2 . This value agrees very well with the findings of Schulman and Hughes [8], who observed at 0.198 nm^2 and 0.195 nm^2 a transition from the α to β condensed state for hexadecanol and eicosylalcohol respectively (at 15°C). Various S -values for long chain alcohols can be found in the literature. These values generally are related to monolayers at air-water or oil-water interfaces [8, 13, 26, 29–31]. Because the properties of bilayers differ significantly from monolayer properties, a detailed comparison with these data does not seem very appropriate. Our findings are in agreement with a bilayer study by Weiss [32] showing that bilayer alkyl chains with cross-sectional areas of 0.19 to 0.25 nm^2 are placed perpendicular to the interface and are in the solid state.

Conclusions

In a series of investigations, research on the colloidal structures present in a nonionic O/W cream and its separate components was presented. Elucidation of these structures could only be achieved by a combination of experimental techniques. Especially the combination of WAXD, SAXD and DSC yield valuable information because these techniques provide quantitative results.

Water plays a very important role in the formation of the above mentioned structures. Hydration lowers the β_0 to α transition temperature of cetylstearylalcohol and increases the melting point of its α -modification. The β_0 to α transition induced by addition of water at ambient temperatures, is caused by a good structural fit of water and the hydroxyl groups of the α -modification. The melting enthalpy is not affected by addition of water.

The properties of PGM₂₀ can be related with the properties of its composing parts, the alkyl chain and the glycerylpolyoxyethylene chain moieties. The pro-

posed antiparallel arrangement of surfactant molecules is based on the molecular dimensions and molecular interactions. Hydration induces a tilt of the alkyl chain with respect to the lipophilic/hydrophilic interface. The maximum tilt is reached at the point at which the basic hydration of the polyoxyethylene chains is completed. The basic hydration number (2 water molecules per oxyethylene unit) is equal to the number derived from the non-freezing water fraction that is found for pure polyoxyethylenes. Excess water contributes to further hydration of the polyoxyethylene chains and induces the formation of a micellar solution in which the hydrocarbon chains are in the liquid state.

Mixtures of cetylstearylalcohol, PGM₂₀ and 20 to 75% W/W water can be regarded as simple creams with a lamellar structure. The properties of these creams (crystallinity of the hydrocarbon chains, melting enthalpy per unit mass of the hydrocarbon fraction, hydration effects, nonfreezing water fraction) can be explained considering their structure and the behaviour of the separate components. The fact that only lamellar phases and no hexagonal phases were observed is probably due to the crystalline character of the alkyl chains. Substitution of water by an aqueous drug solution will affect the hydration of the gel structure. Consequently, it can be expected that some of the properties of the gel will change to a certain extent also. In fact, variation of the interlamellar fluid showed a relation between the melting point of the hydrocarbon bilayers and the polarity of the liquid. Changing the polarity or temperature will possibly also affect the maximum swellability. In turn, this will influence the rate at which a drug is related from the hydrophilic layers. The differences between the preparation temperature and the storage temperature underlies a slow ageing process which will be discussed in a forthcoming paper.

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