Monovalent Cations Trigger Inverted Bilayer Formation of Surfactant Films

Nicolas R. Chevalier,* Corinne Chevallard, and Patrick Guenoun
IRAMIS, LIONS, UMR SIS2M 3299 CEA-CNRS, CEA-Saclay, F-91191 Gif-sur-Yvette Cedex, France

Received July 27, 2010. Revised Manuscript Received September 2, 2010

We monitored single-layer Langmuir—Blodgett films of behenic acid deposited on silanized glass or silicon substrates by atomic force microscopy (AFM) in air. We observed the in situ transformation of the monolayer to a bilayer when the surrounding solution was NaOH or KOH with pH > 8.3. The final state is that of an inverted bilayer, in which both the hydrophobic OTS (octadecyltrichlorosilane) and the alkane chains are exposed to the surrounding solution, defying common intuition based on hydrophobic—hydrophilic energy considerations. Strong sodium-containing carboxylic dimers formed between the headgroups are shown to be responsible for the stabilization of this configuration; calcium ions slow down/inhibit the transformation.

Introduction

Langmuir—Blodgett (LB) films have been used in a wide variety of works to functionalize surfaces.1 The technique relies on non-covalent adsorption of amphiphilic molecules to hydrophilic (mica, oxidized silicon) or hydrophobic (silanized) substrates. Among other functionalizing techniques such as self-assembled monolayers (SAMs) on gold and silver substrates or grafting, LB is the most straightforward and versatile one since it does not require modifying the end groups of the molecules to be deposited (such as thiol or alkylchlorosilane groups).2 In addition, the density of the deposit can be easily tuned by changing the surface pressure of the film.

The stability of LB films has been questioned in several works. It has been found that monolayers of dimethylidioctadecylammonium bromide (DODA) and cetyltrimethylammonium bromide (CTAB) deposited on a mica substrate transformed to bilayers when samples were kept in water. Schwartz et al.3 have similarly shown by atomic force microscopy (AFM) imaging in air that multilayer arachidic acid films (deposited on mica or silicon) which were allowed to stay for a prolonged time in solution during the LB transfer were prone to rearrange to higher order multilayers. In these works, the surfactant always transformed to a multilayer configuration in which the hydrophobic end-group of the surfactant ended up in contact with the solution. The only exception to this rule we know of can be found in a work by Vaknin et al.,4 who ended up in contact with the solution. The only exception to this rule we know of can be found in a work by Vaknin et al.,4 who

among surface probe microscopies, allows for direct imaging of LB films of behenic acid (BA) CH₃(CH₂)₂₀COOH deposited on OTS (octadecyltrichlorosilane) silanized glass or silicon substrates by liquid tapping AFM. We were able to follow the in situ transformation of the initial film to an inverted bilayer when the film was immersed in NaOH or KOH; Ca²⁺ ions were found to slow down and/or inhibit the transformation to a bilayer. The surprising aspect of this phenomenon is that it results in both the hydrophobic OTS and the alkane chains being exposed to the surrounding solution, a situation which seems energetically unfavorable. We show that Na⁺/K⁺ promotes this configuration by forming very stable carboxylic dimers between the behenic acid headgroups. It is generally believed that a divalent cation is required to bridge two carboxylic groups and stabilize inverted bilayer structures: the novelty of our work is to show both experimentally and theoretically that two monovalent cations can fulfill the same task. In addition, our work draws attention to the fact that LB-functionalized hydrophobic substrates are prone to reconfigure depending on solution conditions: care should be taken when using them as functionalized substrates, for example, in the area of templated crystal growth.5–10

Materials and Methods

Silanation. Glass microscope slides (18 × 18 mm², Prolabo, rms roughness ≈ 0.2–0.3 nm measured by AFM) were first thoroughly washed with soap, rinsed with DI water, sonicated for 10 min in DI water (18.2 MΩ-cm, pH 5.5), isopropanol (Merck, 99%), and CHCl₃ (SDS, 99.9%), and dried in an oven (80 °C). Silicon wafers (Siltronix, [111] oriented, rms roughness = 0.14 nm) were only sonicated in CHCl₃. The glass slides and Si wafers were then placed under UV-light (Novascan, PSD Pro Series) for 20 min on each face. At this point, a drop of water completely spreads on the substrate surface. The substrates were then moved to a glovebox under N₂ atmosphere and immersed in

DOI: 10.1021/la102976e
a 1 mM solution of OTS (C18H37Cl3Si) in heptane (SDS, 99%) for 24 h. The excess OTS was thereafter removed by thoroughly washing with heptane; the slides were then taken out of the glove-box and dried. They were finally sonicated in the reverse order in CHCl3, isopropanol, and water and kept in air in a sealed container for further use. The resulting average water contact angles were 103° for silanized silicon wafers (SiOTS) and 97° for silanized glass (GOTS); the latter had a slightly higher drop tilt hysteresis than SiOTS.

**LB Deposit, Isotherms.** Langmuir films of behenic (docosanoic) acid (BA; Fluka, 99%) were spread from a 0.73 g L⁻¹ CHCl3 solution on top of an ultrapure DI water (18.2 MΩ·cm) subphase. The films were allowed to relax for 10 min before they were compressed (Riegler & Kirsten trough, Germany) at a speed of 1.9 Å² s⁻¹ and further equilibrated at the aimed pressure Π for 5–10 min. The silanized substrates were then dipped vertically from the air into the water at a speed of 60 μm s⁻¹; during transfer, the water meniscus pointed downward, and the BA molecules were thus dragged along with the substrate; transfer rates were always 100% (note that this configuration is only possible with substrates having contact angles bigger than 90°; HMDS covered slides, for example, with a water contact angle of ~75° will not give any appreciable transfer ratio). The functionalized substrates are then released in an AFM Teflon fluid cell (home-built) at the bottom of the trough. The remaining BA is sucked from the surface, and the whole cell is transferred to a glass jar containing 400 mL of conservation solution: salts used in this work include NaOH, NaHCO₃ (Normapur, 99.5%), CaCl₂ (Sigma, 99.5%), KOH (Sigma, 80%), and Ca(OH)₂ (Sigma, 96%). Samples were usually kept for times ranging from 1 h to 2 days. Care is taken that the dipped substrate always stays immersed during the whole process.

**AFM.** The monolayers were imaged in the fluid cell in their conservation solution with a Veeco Dimension V atomic force microscope in amplitude modulated tapping mode with DNP (Veeco) 0.58 N/m silicon nitride tips at a frequency of ~15 kHz. The resolution in the z-direction is 0.6 Å. The LB film features had characteristic length scales of ~1 μm such that 10 × 10 μm² images yielded the overall morphology of the films. Images were taken of at least two different regions of the sample. Possible dragging effects of BA molecules by the AFM tip were checked by scanning an area several times and then zooming out; the scanned area did not show any measurable contrast with its surroundings, precluding any tip-related artifacts.

**Contact Angle Measurements.** Contact angle measurements of water and NaOH pH 13 on GOTS were taken in a saturated humidity cell to prevent drop evaporation (when a salt solution slowly evaporates, it crystallizes at the contact line and pins the substrate; transfer rates were always 100% (note that this configuration is only possible with substrates having contact angles bigger than 90°; HMDS covered slides, for example, with a water contact angle of ~75° will not give any appreciable transfer ratio). The functionalized substrates are then released in an AFM Teflon fluid cell (home-built) at the bottom of the trough. The remaining BA is sucked from the surface, and the whole cell is transferred to a glass jar containing 400 mL of conservation solution: salts used in this work include NaOH, NaHCO₃ (Normapur, 99.5%), CaCl₂ (Sigma, 99.5%), KOH (Sigma, 80%), and Ca(OH)₂ (Sigma, 96%). Samples were usually kept for times ranging from 1 h to 2 days. Care is taken that the dipped substrate always stays immersed during the whole process.

**Results**

Figure 1a shows an image of the BA monolayer morphology in pure water (pH 5.6); the monolayer on GOTS appears as a somewhat loose structure, while it is more ordered and dense on SiOTS (Supporting Information). Film pressure (10, 20, or 32 mN/m) had no visible effects on monolayer morphology on GOTS. The monolayers in pure water were stable over time (no noticeable morphology change 2 days after deposit).

We next replaced the fluid in the cell with NaOH pH 11. Figure 1b–d depicts the evolution in time of the monolayer. After 30 min, aggregate structures are seen to have nucleated (nucleation densities were on the order of 1 nucleus/10 μm²); they are surrounded by a halo depleted of BA, indicating that the aggregates result from the surfactant diffusing and folding to a thicker structure.

![Figure 1. Liquid AFM height images, XY full scale 10 × 10 μm: (a) behenic acid monolayer at Π = 10 mN/m on GOTS substrate in DI water; (b) 30 min after replacement of the liquid in the fluid cell by NaOH 1 mM, bilayer islands appear (bright spots); (c) 1 h later, bilayer islands are seen to grow at the expense of the monolayer (gray); the silane substrate appears (dark) where the monolayer has been depleted (d) 1 day later, and the initial monolayer has completely transformed to a bilayer (bright streaks). Bilayer coverage on this image can be computed to be 48.6%.

In the following, the word “folding” will be used to describe the transition from a mono- to a multilayer state (see Figure 5). The aggregates often grew along one particular edge. Typical growth speeds of the edges at this early stage were 100–500 nm h⁻¹. Imaging of the monolayer after 24 h reveals that the monolayer has completely folded (Figure 1d). Folding also occurred on SiOTS substrates (Supporting Information) and was independent of the pressure at which the film was deposited (10, 20, or 35 mN/m) (Supporting Information).

Height histogram analysis performed on partially (Figure 2b) or fully folded films and averaged over 21 images of 10 different samples reveals that the average aggregate height is 5.65 ± 0.36 nm, while that of the monolayer is 1.72 ± 0.13 nm and 2.56 ± 0.32 nm on GOTS and SiOTS, respectively. These values are to be compared with the fully stretched length of an equivalent linear alkane C₂₂H₄₆, computed according to Tanford. From these values, we can first conclude that the BA molecules in the monolayer configuration have a tilt angle η = a cos(measured/3 / l_stretched) = 54 ± 3° on GOTS and θ = 36 ± 12° on SiOTS; the former value seems to indicate that the monolayer is disordered on GOTS. The aggregate height indicates a bilayer with a tilt angle in the range 0–25°. The thickness we find is in good agreement with wide-angle X-ray scattering (WAXS) diffraction results on sodium behenate soap crystals and reflectivity measurements on 30 layer thick sodium behenate films on Si wafers; they find bilayer thicknesses of 5.5 and 5.6 nm respectively, and tilt angles in the range 0–26°. As they assemble to a bilayer, the chains straighten up,

(11) http://bigwww.epfl.ch/demo/dropanalysis.


forming a more regular, compact structure; in fact, all the bilayer regions we observed were almost free of defects (holes) and were less rough than the monolayer. Trilayer folding was observed on only one sample and represented only a minor fraction of the surface coverage. Analysis (with the GIMP software) of the multilayers after complete folding (Figure 1d and Supporting Information) shows that the surface coverage is close to 50%. It could be less if the BA molecules were to desorb or form higher order multilayers. The fact that we find nearly \( \sim 50\% \) coverage gives further, independent proof that the aggregate structure is really a bilayer.

To understand whether the bilayer exposed its hydrophobic tails or its carboxylic headgroups to the surrounding solution (see Figure 5), we analyzed the phase images taken by tapping mode AFM of partially folded BA films, shown in Figure 3b. In amplitude modulated tapping mode, greater phase lag (corresponding to darker regions in Figure 3b) has been associated with greater dissipation resulting from inelastic interactions between the tip and the sample.\(^{15,16}\) SAMs bearing regions differing only by their surface chemical functionalities (COOH or CH\(_3\)), and not by their local stiffness, have been used to demonstrate that phase contrast images taken in liquid can be used to distinguish chemical groups.\(^{17}\)

On partially folded monolayers, the monolayer regions always stood out as bright regions, whereas contrast between the bilayer and OTS was in most cases (3 out of 4) weak, or even nonexistent; in the fourth case, the bilayer appeared darker than the OTS. This indicates that the bilayer regions have a surface chemistry similar to that of the OTS substrate: the alkane chains are exposed to water and not the headgroups. Care should however be taken in interpreting phase contrast images: a recent paper\(^{18}\) has shown that, under certain conditions, the dominant contribution to phase contrast in liquid amplitude modulated tapping AFM is the local variation of Young’s modulus. In our case, following this interpretation of phase contrast would lead to the conclusion that the monolayer is softer (smaller phase lag) than both the silane and the bilayer, which seems reasonable, but it does not enable us to conclude on whether the bilayer is an inverted one or not. This theory however clearly does not apply to the study of Noy et al.,\(^{17}\) where stiffness is uniform and the phase contrast observed can only be due to differences in chemical surface groups. In our case, a further experiment dispels any ambiguity arising from the interpretation of phase images: we removed a GOTS substrate covered with a fully folded LB film from its conservation solution and imaged it in air after washing and drying. The bilayer regions were stable and had a thickness of \( \sim 5.6 \) nm. The contact angle of water on the substrate was 98 \( ^\circ \) (2\(^\circ\)), that is, equal to that of water on bare GOTS, confirming the hydrophobic nature of the bilayer surface.

We investigated the influence of pH and ions on the LB film in solution. Folding occurred at pH 8.3 (NaHCO\(_3\) 10 mM), phase contrast in liquid amplitude modulated tapping AFM is the local variation of Young’s modulus. In our case, following this interpretation of phase contrast would lead to the conclusion that the monolayer is softer (smaller phase lag) than both the silane and the bilayer, which seems reasonable, but it does not enable us to conclude on whether the bilayer is an inverted one or not. This theory however clearly does not apply to the study of Noy et al.,\(^{17}\) where stiffness is uniform and the phase contrast observed can only be due to differences in chemical surface groups. In our case, a further experiment dispels any ambiguity arising from the interpretation of phase images: we removed a GOTS substrate covered with a fully folded LB film from its conservation solution and imaged it in air after washing and drying. The bilayer regions were stable and had a thickness of \( \sim 5.6 \) nm. The contact angle of water on the substrate was 98 \( ^\circ \) (2\(^\circ\)), that is, equal to that of water on bare GOTS, confirming the hydrophobic nature of the bilayer surface.

We investigated the influence of pH and ions on the LB film in solution. Folding occurred at pH 8.3 (NaHCO\(_3\) 10 mM),
although its kinetics was slower than that in more alkaline solutions (the bilayer was found to be only half-folded after 24 h, see the Supporting Information). This narrows the pH range at which the transformation takes place to between 5.6 (no reversal) and 8.3. KOH (1 mM, pH 11) solutions and 5 mM Na₂CO₃ (pH 10.7) also induced folding (Supporting Information). In 10 mM CaCl₂ (pH 5.6), the monolayer was stable (Supporting Information) and had the same appearance as that in water. In 0.5 mM Ca(OH)₂ (pH 11), small bilayer islands had nucleated after 2 days, but would barely grow, even after having spent 1 week in solution (Figure 4). The same behavior was observed when the monolayer was dipped in a NaOH 1 mM + CaCl₂ 5 mM solution (Supporting Information), indicating that Ca²⁺ ions inhibited folding in this case.

Next, we questioned the reversibility of the transformation by transferring completely folded bilayers in mildly acidic HCl solutions (pH 4): after 7 h, large bilayer regions were still found (Supporting Information), indicating that the transformation to a bilayer state was mostly irreversible.

Does bilayer formation also occur at the air–water interface? Film collapse pressures, measured at a compression speed of 1.9 Å/min, were smaller on NaOH than on H₂O: we find \( \Pi_{\text{NaOH}} = 45.4 \pm 1.3 \text{ mN m}^{-1} \) and \( \Pi_{\text{H₂O}} = 54.6 \pm 3.5 \text{ mN m}^{-1} \) (Supporting Information); this latter value is in agreement with the literature.⁶ The pressure rise on NaOH pH 11 occurred at molecular areas ~15% smaller than those on H₂O. Compared Brewster angle microscopy images of the LB films on H₂O and on NaOH pH 11 at surface pressures \( \Pi \) ranging from 0 to 32 mN m⁻¹ did not however reveal any difference in film morphology (Supporting Information). A constant pressure (10 mN m⁻¹) can be kept on the NaOH pH 11 solution for 4 h without any measurable area change. This indicates that, at the air–water interface, bilayer formation does not occur spontaneously at pressures \( \Pi < 32 \text{ mN m}^{-1} \).

We wondered whether the OTS layer might become altered when kept for prolonged time in solution, especially at high pH. Maccarini et al.¹⁹ have shown OTS monolayers to be stable in pure water for at least 24 h. Concerning the effect of alkali, a recent paper by Tian and Shen²⁰ showed using phase-sensitive sum-frequency generation spectroscopy that OH⁻ tends to adsorb to OTS monolayers more than other ions (H₂O⁻ or Cl⁻); the authors could not however measure absolute binding constants. The association of hydroxide ions to the OTS could lower the water–OTS interfacial tension. In order to test this idea, we measured contact angles of NaOH pH 13 drops on OTS substrates in a saturated humidity cell. Contact angles were 97.5 ± 2°, almost identical to those found using pure water drops, and did not evolve over the course of 2 h. We also tracked possible morphological changes of the OTS layer in NaOH by liquid AFM: no changes were observed after the substrate was kept for 1 day in NaOH pH 11. Although these results do not in any way contradict the findings of Tian and Shen,²⁰ they show that OH⁻ ions do not exert any significant influence on the OTS layer and the water–OTS interfacial energy.

Discussion

When forming a bilayer, the behenic acid molecules either form tail–tail contacts and expose their hydrophilic heads to the surrounding solution (path 1 in Figure 5), or form a head–head interface and end up with the alkane chain exposed to the solution (inverted bilayer, path 2). Our experiments indicate that the outermost layer is hydrophobic and thus clearly cut in favor of scenario number 2.

It should be noted that folding schemes where monolayers would be stacked on top of each other (tail on head) are not in agreement with WAXS results,¹⁵ that find the unit cell of sodium benenate to always consist of a bilayer.

Exposure of the OTS substrate and the alkane tails to the surrounding solution comes at an energetic cost. This energy (per unit surface) can be estimated as \( \Delta G_{\text{adhesion}} = (\gamma_{\text{OTS/water}} + \gamma_{\text{tail/water}} - \gamma_{\text{tail/OTS}}) \), where we denote by \( \gamma_{X/Y} \) the interfacial tensions between \( X \) and \( Y \). Given that \( \gamma_{\text{OTS/water}} \approx \gamma_{\text{tail/water}} \approx \gamma_{\text{oil/water}} = 48 \text{ mJ m}^{-2} \), and that \( \gamma_{\text{tail/OTS}} \approx 0 \), we find \( \Delta G_{\text{adhesion}} = 96 \text{ mJ m}^{-2} \). This value is in good agreement with work of adhesion measurements of hydrophobic surfaces in water.²² As bilayer formation is seen to happen spontaneously in alkaline solutions of NaOH and KOH, this energetic cost Has to be counterbalanced by a favorable contribution from the head–head contacts in the inverted bilayer.

It is well-known that in-plane repulsive electrostatic interactions shift the pKa of the monolayer to higher values than the

---

(21) http://www.kayelaby.npl.co.uk/.
single-molecule one (typically pK_a ∼ 5 for carboxylic groups). 23–25 Goddard et al. 13 have investigated the surface potential of behenic acid monolayers on alkaline solutions and found that a sharp break in the surface potential versus pH curve occurs at pH 9; Le Calvez et al. 26 measured by using PM-IRRAS a pK_a of 9.0 for arachidic acid on a NaOH subphase at the air–water interface. On solid substrates, SAMS of 3-mercaptopropionic acid on gold were found to have pK_a values of 7.4 and 7.7. 24,25 In our system, the minimal pH value as from which folding develops was found to lie in the range 5.6–8.3, which is consistent with the pK_a values we just mentioned. This is suggestive of the fact that bilayer formation is linked to the deprotonation of the behenic acid headgroups.

Counterions subsequently condense on the deprotonated monolayer. In 1 mM NaOH, the only counterions left in the bulk solution are Na^+ ions. To describe the extent of screening/counterion association on the monolayer just after it has been transferred to NaOH pH 11 solution, we have solved the nonlinear Poisson–Boltzmann (PB) equation, \( \phi_j = -e\varepsilon_\varepsilon_0 \kappa \sum \Delta \beta_j \) the squared inverse screening length, \( \kappa = (2\pi n_0 e^2/\varepsilon_0 k_B T) \) the inverse square of the elementary electric charge, and \( \varepsilon_\varepsilon_0 \) the dielectric constant. \( \varepsilon_\varepsilon_0 \) is the dimensionless potential, \( \kappa^2 = (2\pi n_0 e^2/\varepsilon_0 k_B T) \) the inverse square of the elementary electric charge, and \( \varepsilon_\varepsilon_0 \) the dielectric constant. \( \varepsilon_\varepsilon_0 \) is the dimensionless potential, \( \kappa \) the Boltzmann temperature, \( k_B \) the Boltzmann constant, \( e \) the elementary electric charge, and \( e_\varepsilon_0 \) the water electrical permittivity. We used charge-regulating boundary conditions to describe the local association equilibrium COO^− + Na^+ → COONa (the so-called Stern layer) with a binding constant \( K = 0.54 \text{ M}^{-1} \), which is the binding constant of Na^+ to acetic acid. 27 We solved the equation for the fraction of dissociated groups \( \alpha = [\text{COO}^-]/([\text{COO}^-] + [\text{COONa}^-]) \). For a density of behenic molecules equal to 1/20 Å^−2 in 1 mM NaOH, computation yields that \( \alpha = 20\% \); that is, 80% of the surface groups are complexed by Na^+; the reduced potential at the boundary is high, \( \psi_0 \approx -8.9 < \gamma \). This shows that, in NaOH pH 11, (1) sodium ions are strongly condensed on the monolayer and (2) the local [H^+] concentrations [H^+] = 10^{-10} \exp(-\psi_0) = 7.3 \times 10^{-6} \text{ M} can be neglected. In contrast, in H_2O pH 5.6, the only cation present is H^+.

The above considerations and the requirement of charge neutrality suggest a chemical description for the head–head contact in the bilayer state, depicted in Figure 6a. Similarly, if a bilayer were to form in H_2O pH 5.6 (which we did not observe, the monolayer was stable in pure water), the head–head contact could be that depicted in Figure 6b.

Figure 6b represents the well-known 29 carboxylic dimer, stabilized by hydrogen bonds; Figure 6a is a sodium carbonate dimer: the existence of this dimeric form has been evidenced experimentally in perfluoropolyethers and investigated computationally by Doan et al. 28 They computed heats of dimerization in vacuum of, respectively, -19.5 and -53.1 kcal mol^{-1} for the hydrogen and sodium dimers. The much lower heat of dimerization of the sodium dimer is due to the attractive Coulombic interactions between the ionic constituents.

Thus, the headgroup–headgroup interactions in the bilayer could be a factor 53.1/19.5 ≈ 2.7 more attractive after ionization and replacement of the counterion by Na^+. Further support for the notion that the bilayer could be stabilized by sodium carboxylate dimers (this structure is also referred to in the literature as a “fatty acid soap”) can be found in the bulk phase diagram established by Cistola et al. 20 who found that it is the dominant form for fatty acids in potash at pH > 9 below their melting point.

We shall now give an estimate of the Gibbs free energy change (per unit surface) brought about by dimer formation in water, \( \Delta G_{X,dim} \). For hydrogen carboxylic dimers, \( \Delta G_{H,dim} \) can be estimated from measurements by various experimenters of the association constant \( K_a \) of formic acid dimers in water, 28 through the relation \( \Delta G_{H,dim} = \sigma F RT \ln(K_a) \), where \( \sigma F = 8.3 \times 10^{-6} \text{ mol m}^{-2} \) is a typical value for the surface density of the behenic acid monolayers we considered. Formic acid was chosen over other carboxylic acids because the contribution of its alkane part to the surface density is negligible. Therefore, the minimum and maximum values of \( K_a \) that can be found in the literature, 20 we find \( \Delta G_{H,dim} \) in the range -60 to -100 mJ m^{-2}. Assuming that the ratio of the heats of formation of a sodium dimer to that of a hydrogen dimer is approximately the same in water as in vacuum, we find \( \Delta G_{Na,dim} \) in the range -160 to -270 mJ m^{-2}. A further, positive contribution to \( \Delta G_{Na,dim} \) comes from the fact that Na^+ counterions must first be completely condensed on the monolayer. This contribution is the inverse of the Gibbs free energy of electric double layer formation: it amounts to \( \sim 10 \text{ mJ m}^{-2} \) and can thus be neglected. This estimate thus shows that \( \Delta G_{H,dim} < \Delta G_{Na,dim} \), suggesting that the formation of sodium dimers in NaOH could trigger the transformation to an inverted bilayer. The same mechanism is likely to hold true for KOH.

The measurements we performed at the air–water interface show that the pressure rise on NaOH pH 11 occurs at lower molecular areas than on H_2O. 31 Moreover, induced collapse happens on NaOH at lower pressures than on H_2O. This indicates that attractive interactions are higher on NaOH and that multilayer stacks form more readily, which is in agreement with the overall fact that it leads to aggregation (we have not however analyzed in detail the nature of the collapsed films on NaOH). The work of adhesion \( \gamma'_{air/water} + \gamma'_{NaOH/water} - \gamma'_{NaOH/air} \) at the air–water interface works out to be approximately the same as that of H_2O.

(31) This indicates that NaOH induces attractive interactions between the headgroups of the monolayer, contrary to intuitive belief based on considering only charge–charge repulsions. Goddard et al. 25 find a decreasing surface per molecule area with increasing alkalinity in the pH range 2–10. Our result at pH 11 goes along this trend. An expansion of the film occurs as from pH 12.
on OTS (since $\gamma_{\text{air/water}} = 72 \text{ mJ m}^{-2}$ but $\gamma_{\text{tail/air}} \sim 20-25 \text{ mJ m}^{-2}$) so that bilayer formation could also in theory occur spontaneously at the surface of NaOH. We believe the difference might lie in the fact that the BA molecules at the air–water interface are kept under pressure by the barriers; in contrast, after being deposited on a substrate, no lateral pressure is exerted anymore. This could give the film more conformational freedom and lower the activation barrier to bilayer formation. We also note that Johann and Vollhardt\(^\text{32}\) have observed on NaOH pH 13 solutions condensed domains of behenic acid at the air–water interface which bear resemblance to the ones we observed on substrates.

We can only speculate as to why inverted bilayer formation is incomplete in the presence of Ca\(^{2+}\). It could be due to the formation of a calcium carboxylate complex where the two alkane chains are still in the monolayer state, as depicted in Figure 7. The existence of this complex has been confirmed\(^\text{26}\) by PM-IRRAS on arachidic acid monolayers; the authors find that, as from pH 6.2, the carboxylic headgroups are all in this calcium-complexed form. The stability of this complex could be high enough that bilayer formation would result in little or no energetic gain. This aspect is not taken into account in our estimate of the Gibbs energy of dimer formation. It could lead to very slow or even inhibit bilayer formation, as observed in Figure 4. Vaknin et al.\(^\text{6}\) found that arachidic acid on 1 mM CaCl\(_2\) pH 6.5 subphases form inverted bilayers when compressed above a collapse pressure of $\Pi_{\text{collapse}} = 55 \text{ mN m}^{-1}$. This collapse pressure is higher that the one we find on NaOH, which agrees with the fact that Na\(^+\) seems to be more efficient at promoting the inverted bilayer configuration than Ca\(^{2+}\). Vaknin et al.\(^\text{6}\) moreover note that inverted bilayer formation never happens spontaneously at pressures below $\Pi_{\text{collapse}}$ although computations\(^\text{33}\) predict that this configuration is energetically favorable.

To summarize, we have found that behenic acid adsorbed on OTS monolayers forms an inverted bilayer when exposed to alkali solution, resulting in the formation of a two-dimensional sodium or potassium behenate bilayer, also called soap. This folding appears counterintuitive as the monolayer switches from a presumably stable situation to one where both the hydrophobic OTS substrates and the hydrophobic alkane chains are exposed to water. This instability is triggered by the charging of the headgroups and subsequent replacement of the counterions by Na\(^+\) (or K\(^+\)). We suggest that its driving force lies in the formation of strong, Coulomb stabilized carboxylic dimers, that overcome the adhesion energy of the fatty acid alkane tail to the substrate. It is generally thought that cations of valency > 2 are required to bind different carboxylic groups; this work illustrates how two monovalent cations, given the right pH conditions, can perform the same task. This work points at the ambivalence of using Langmuir–Blodgett technique to functionalize surfaces: since the method relies on noncovalent adsorption, it enables a wide variety of amphiphilic molecules to be deposited but sacrifices monolayer stability.

Acknowledgment. We thank Christian Blot for building the fluid cell, and Michel Goldmann and Luc Belloni for helpful discussions.

Supporting Information Available: Bilayer formation on SiOTS, folding at different film deposition pressures, surface coverage of bilayer after complete folding, images of bilayer in air and contact angle, isotherms and BAM images of behenic on H\(_2\)O and NaOH, irreversibility of monolayer folding, images of behenic on GOTS in CaCl\(_2\) 5 mM, KOH 1 mM, and CaCl\(_2\) 5 mM + NaOH 1 mM. This material is available free of charge via the Internet at http://pubs.acs.org.

---

Figure 7. Calcium behenate complex; the two alkane chains are depicted in the monolayer state, lying side by side.

---
