Base-Pair Formation of Self-Organizing RNA Amphiphiles within Two Dimensions

Ulf Rädler,[†] Cornelia Heiz,[‡] Pier Luigi Luisi,[‡] and Robert Tampé*[,†,§]

Lehrstuhl für Biophysik, Technische Universität München, James-Franck-Strasse, D-85747 Garching, Germany, Institut für Polymere, ETH-Zürich, Universitätstrasse 6, CH-8092 Zürich, Switzerland, and Max-Planck-Institut für Biochemie, Am Klopferspitz 18a, D-82152 Martinsried, Germany

Received May 1, 1998. In Final Form: September 22, 1998

In an attempt to get a better insight in specific base pairing by self-organization of DNA or RNA, the synthesis of a complete set of monoalkyl ribonucleotides is described. The thermodynamic phase and mixing properties of these self-organizing RNA amphiphiles were analyzed within two dimensions using film balance technique. The pure compounds form stable monolayers at the air−water interface on a physiological buffer. In analogy to base-paired RNA or DNA in solution, the monolayers were mostly stabilized by complexing the phosphate groups by calcium or magnesium ions. By use of mixtures of RNA amphiphiles, specific recognition and base-pair formation of monoalkyl phospho adenosine/uridine were demonstrated.

Introduction

The combination of molecular recognition and self-assembly plays a crucial role in biological systems such as RNA, DNA,[1] and biomembranes.[2−4] In these unique systems, molecular interactions are the basis for information storage and transfer of the genetic code, substrate binding to proteins, cell−cell recognition processes, signal transduction, and others.[5]

Hydrogen bonding and Coulomb interactions are major aspects of molecular recognition.[6,7] In aqueous solution hydrogen bonding is less effective for specific recognition since the molecules are surrounded by bulk water.[8] As a result, base pairing of free nucleotides is not observed in water. However, in nature and molecular design, enthalpically bound water (in form of crystal or surface water) is extremely important for structure formation, while the bulk water is excluded from molecular architecture. Therefore, nature is using special geometry and steric hindrance as in the RNA or DNA backbone,[9,10] while in chemistry a hydrophobic cage is build around the recognition units.[11−13] Interestingly, the storage and transfer of information in RNA and DNA (by itself a linear process) is a two-dimensional problem taking place in the plane of the base pairs. The surrounding three-dimensional architecture is needed only to fit the information carrier into the right place and to exclude the water, so that specific hydrogen bonding and therefore specific base pairing is possible.

In chemistry, the concept of self-assembly is used to organize molecules in an architecture depending on energetic and entropic terms as mobility, orientation, and hydrophobicity while the chemical functionality of the molecules remains mainly unchanged.[14,15] Due to the hydrophobic effect[16] amphiphiles self-assemble into supramolecular structures such as vesicles, micelles, or thin films which can be transferred by various techniques without loss of their chemically and laterally encoded information.[4] Of special interest are self-assembled monolayers at the air−water interface, which can be investigated thermodynamically by film balance techniques. These monolayers can serve, e.g., as model membranes[17,18] or as templates for 2D crystallization.[19−21] Hydrogen bond networks at the air−water interface were first described by Kunitake et al.[22,23] Interlayer recognition processes at the air−water interface between nucleobase derivatives and their complementary partners dissolved in the subphase,[24−28] and recently intralayer

* Corresponding author: Dr. Robert Tampé, Max-Planck-Institut für Biochemie, Am Klopferspitz 18a, D-82152 Martinsried, Germany. Fax: ++49-89-8578-2641. E-mail: tampe@biochem.mpg.de.

[†] Technische Universität München.

[‡] Institut für Polymere, ETH-Zürich.

[§] Max-Planck-Institut für Biochemie.

(1) Saenger, W. *Principles of Nucleic Acid Structure*; Springer-Verlag: New York, 1984.

(2) Ringsdorf, H.; Schlarb, B.; Venzmer, J. *Angew. Chem.* **1988**, *100*, 117−162.

(3) Sackmann, E. *Science* **1996**, *271*, 43−48.

(4) Tampé, R.; Dietrich, C.; Gritsch, S.; Elender, G.; Schmitt, L. In *Nanofabrication and biosystems: Integrating materials science, engeneering, and biology*; Hoch, H. C., Jelinski, L. W., Craighead, H. G., Eds.; Cambridge University Press: Cambridge, 1996; pp 201−221.

(5) Lehn, J. M. *Supramolecular chemistry*; VCH: Weinheim, 1995.

(6) Saenger, W.; Jefferey, G. *Hydrogen bonding in biological structures*; Springer-Verlag, Berlin, 1991.

(7) Israelachvili, J. N. *Intermolecular and surface forces*; Academic Press: London, 1991.

(8) Nowick, J. S.; Chen, J. S.; Noronha, G. *J. Am. Chem. Soc.* **1993**, *115*, 7636−7644.

(9) Bolli, M.; Micura, R.; Eschenmoser, A. *Chem. Biol.* **1997**, *4*, 309−320.

(10) Sheppard, T. L.; Breslow, R. C. *J. Am. Chem. Soc.* **1996**, *118*, 9810−9811.

(11) Nowick, J. S.; Chen, J. S. *J. Am. Chem. Soc.* **1992**, *114*, 1107−1108.

(12) Wilcox, C. S.; Adrian, J. C.; Webb, T. H.; Zawacki, F. J. *J. Am. Chem. Soc.* **1992**, *114*, 10189−10197.

(13) Sessler, J. L.; Wang, R. *J. Am. Chem. Soc.* **1996**, *118*, 9808−9809.

(14) Lehn, J. M. *Angew. Chem., Int. Ed. Engl.* **1990**, *29*, 1304−1319.

(15) Conn, M. M.; Rebek, J. *Curr. Opin. Struct. Biol.* **1994**, *4*, 629−635.

(16) Tanford, C. *The hydrophobic effect. Formation of micelles and biological membranes*; John Wiley & Sons: New York, 1980.

(17) McConnell, H. M. *Annu. Rev. Phys. Chem.* **1991**, *42*, 171−195.

(18) Möhwald, H. In *Structure and dynamics of membranes;* Lipowsky, R., Sackmann, E., Eds.; Elsevier: Amsterdam, 1995; Vol. 1A, pp 161−212.

(19) Fromherz, P. *Nature* **1971**, *231*, 267−268.

(20) Uziigiris, E. F.; Kornberg, R. D. *Nature* **1983**, *310*, 134−136.

(21) Jap, B. K.; Zulauf, M.; Scheybani, T.; Hefti, A.; Baumeister, W.; Aebi, U.; Engel, A. *Ultramicroscopy* **1992**, *46*, 45−84.

(22) Kimizuka, N.; Kawasaki, T.; Kunitake, T. *J. Am. Chem. Soc.* **1993**, *115*, 4387−4388.

(23) Lehn, J. M.; Mascal, M.; Decian, A.; Fischer, J. *J. Chem. Soc., Perkin Trans. 2* **1992**, 461−467.

10.1021/la980515s CCC: $15.00
Published on Web 10/17/1998

recognition of dioleoylphosphatidyl nucleosides was reported.[29] Because of their antitumor activity modified nucleotides are a field of great interest. The nucleotides can be derivatized at the phosphate group,[30−32] at the ribose unit,[10] or if the recognition process is not the matter of investigation at the purine or pyrimidine base itself.[33−35] ATP-lipids were synthesized serving as protein anchor and energy source in two dimensions.[35]

In this report, we describe the synthesis of novel monoalkyl ribonucleotides **6a**−**d**. These RNA analogues may lead to new insights into base pair formation induced by self-organization during early steps of evolution in the so-called RNA world.[36] A thermodynamic investigation of these monoalkyl ribonucleotides and their mixtures at the air−water interface is presented.

Results and Discussion

Synthesis of RNA Amphiphiles: Starting from hexadecanol **1**, we received the phosphocholine **4** in a one-pot three-step synthesis in 63% yield.[37] The hexadecanol **1** was converted with ethylene chlorophosphite to the alkyl phosphite **2**. Oxidation with bromine gave under ring opening the alkylphosphonate **3** which was aminated to the phosphocholine **4**. The hexadecylphosphoadenosine (HDP-A, **6a**), the hexadecylphosphouridine (HDP-U, **6b**), the hexadecylphosphoguanosine (HDP-G, **6c**), and the hexadecylphosphocytosine (HDP-C, **6d**) were synthesized via transphosphorylation using Phospholipase D.[38] In this enzymatic reaction only the primary alcohol of the nucleosides is linked to the alkyl phosphate to give the alkyl nucleotides **6a**−**d** in good yields (Figure 1).

Influence of Subphase Conditions. Film balance techniques are most suited to investigate the thermodynamic properties of self-organizing molecules including their mixing behavior. The RNA amphiphiles were spread at the air−water interface, and the influence of different subphase conditions on area−pressure isotherms of HDP-A were investigated (Figure 2a). In analogy to base-paired RNA or DNA in solution, the nucleotide monolayers can be stabilized due to shielding of the electrostatic repulsion of the negatively charged phosphate groups by ion strength and complexing divalent ions such as calcium and magnesium. Compared to 1 mM NaCl, addition of only 1 mM $CaCl_2$ to the subphase changes the π/A isotherm drastically. A plateau region in the range of 38−75 Å^2/molecule and a transition in a solid analogue phase are observed. Addition of 150 mM NaCl raised the plateau about 3 mN/m, and substitution of magnesium ions against calcium ions shifts the isotherm to higher pressure. However, as the plateau phase is steeper, we continued working with a physiological buffer supplemented with 1 mM $CaCl_2$.

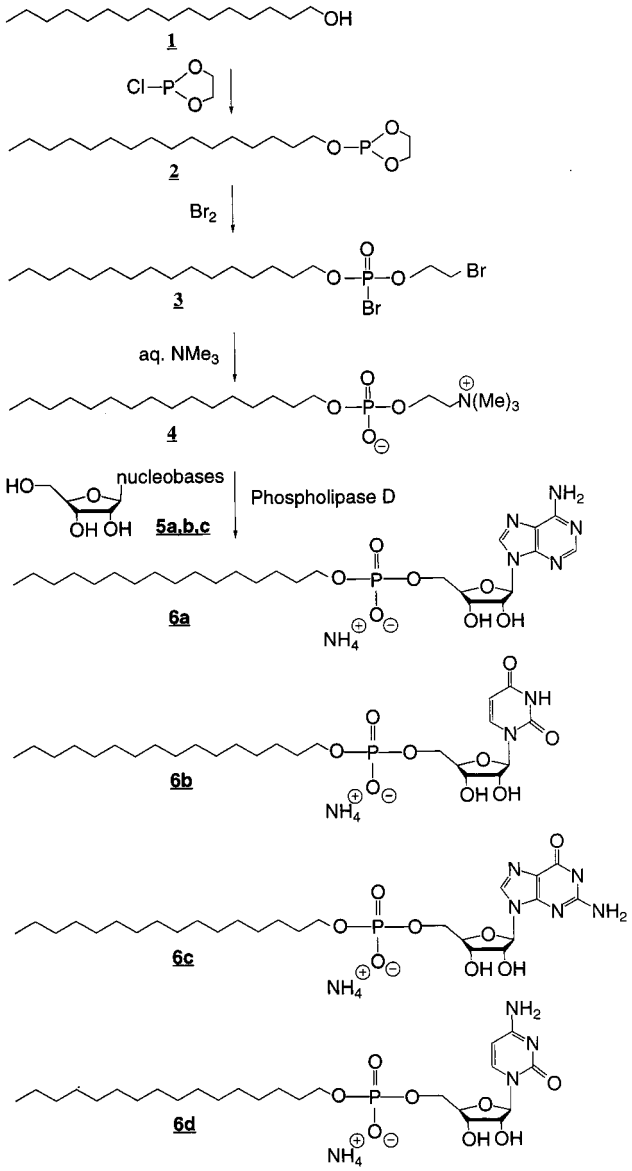

Figure 1. Synthesis of the monoalkyl nucleotides.

Pure Compounds. After spreading onto a physiological buffer (10 mM HEPES, 150 mM NaCl, 1mM $CaCl_2$, pH 7.5), all alkyl nucleotides formed stable monolayers at least up to a lateral pressure of 30 mN/m (Figure 2b). The isotherms of **6a** and **6c** show a transition from an expanded to a condensed phase. Dense packing might be due to stacking of the purine bases. The π/A isotherms of the pyrimidine derivatives HDP-C and HDP-U show no condensed phases, which is characteristic for amphiphiles with large headgroups.[39] We assume that due to the relative hydrophilicity of the pyrimidine bases π stacking is prevented. Because of the additional amino group in the pyrimidine base, the phosphoalkyl cytidine **6d** occupies

(24) Kitano, H.; Ringsdorf, H. *Bull. Chem. Soc. Jpn.* **1985**, *58*, 2826−2828.
(25) Ahlers, M.; Ringsdorf, H.; Rosemeyer, H.; Seela, F. *Colloid Polym. Sci.* **1990**, *268*, 132−142.
(26) Ding, D.; Zhang, Z.; Shi, B.; Luo, X.; Liang, Y. *Colloids Surf.* **1996**, *112*, 25−30.
(27) Oishi, Y.; Torii, Y.; Kato, T.; Kuramori, M.; Suehiro, K.; Ariga, K.; Taguchi, K.; Kamino, A.; Koyano, H.; Kunitake, T. *Langmuir* **1997**, *13*, 519−524.
(28) Shimomura, M.; Nakamura, F.; Ijiro, K.; Taketsuna, H.; Tanaka, M.; Nakamura, H.; Hasebe, K. *J. Am. Chem. Soc.* **1997**, *119*, 2341−2342.
(29) Berti, D.; Franchi, L.; Baglioni, P.; Luisi, P. L. *Langmuir* **1997**, *13*, 3438−3444.
(30) Shuto, S.; Itoh, H.; Ueda, S.; Imamura, S.; Fukukawa, K.; Tsujino, M.; Matsuda, A.; Ueda, T. *Chem. Pharm. Bull.* **1988**, *36*, 209−217.
(31) Bonaccio, S.; Wessicken, M.; Berti, D.; Walde, P.; Luisi, P. L. *Langmuir* **1996**, *12*, 4976−4978.
(32) Wang, P.; Schuster, M.; Wang, Y. F.; Wong, C. H. *J. Am. Chem. Soc.* **1993**, *115*, 10487−10491.
(33) Limbach, P. A.; Crain, P. F.; McCloskey, J. A. *Nucleic Acids Res.* **1994**, *22*, 2183-2196.
(34) Robertson, M. P.; Miller, S. L. *Science* **1995**, *268*, 702−705.
(35) Schmitt, L.; Tampé, R. *J. Am. Chem. Soc.* **1996**, *118*, 5532−5543.
(36) Gilbert, W. *Nature* **1986**, *319*, 618−618.
(37) Erukulla, R. K.; Byun, H. S.; Bittman, R. *Tetrahedron Lett.* **1994**, *35*, 5783−5784.
(38) Shuto, S.; Imamura, S.; Fukukawa, K.; Sakakibara, H.; Murase, J. *Chem. Pharm. Bull.* **1987**, *35*, 447−449.
(39) Gaines, G. L. *Insoluble Monolayers at liquid gas interfaces*; John Wiley & Sons: New York, 1966.

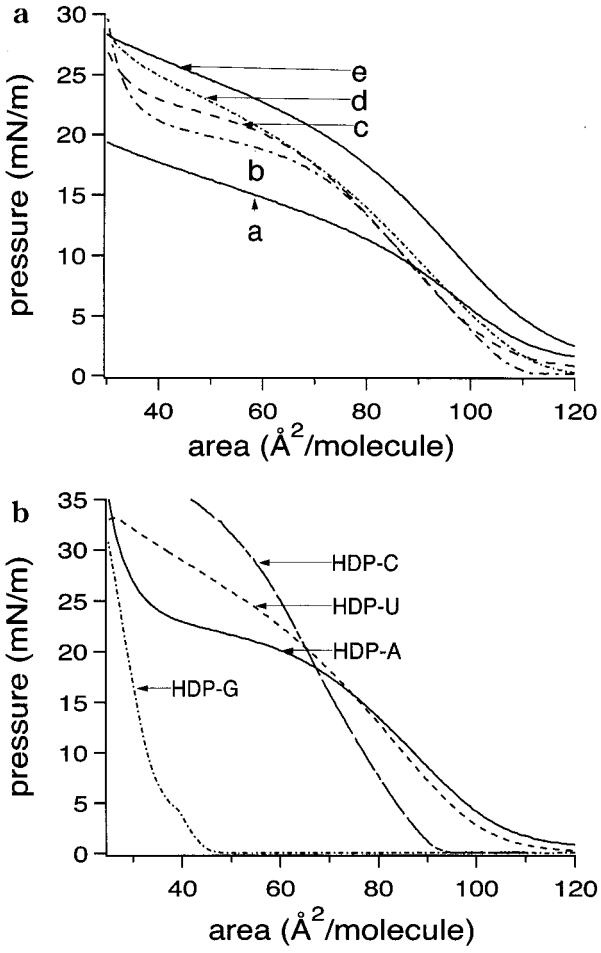

Figure 2. (a) Influence of subphase conditions on the π/A isotherms of **6a**. For these measurements the standard buffer (10 mM HEPES, pH 7.5, 5 °C) was supplemented with (a) 1 mM NaCl, b) 1 mM $CaCl_2$, (c) 150 mM NaCl, 1 mM $CaCl_2$, (d) 150 mM NaCl, 1 mM $MgCl_2$, or (e) 150 mM NaCl. (b) Comparison of the complete set of monoalkyl ribonucleotides **6a**, **6b**, **6c**, and **6d**. Subphase conditions: 10 mM HEPES, 150 mM NaCl, 1 mM $CaCl_2$, pH 7.5, 5 °C.

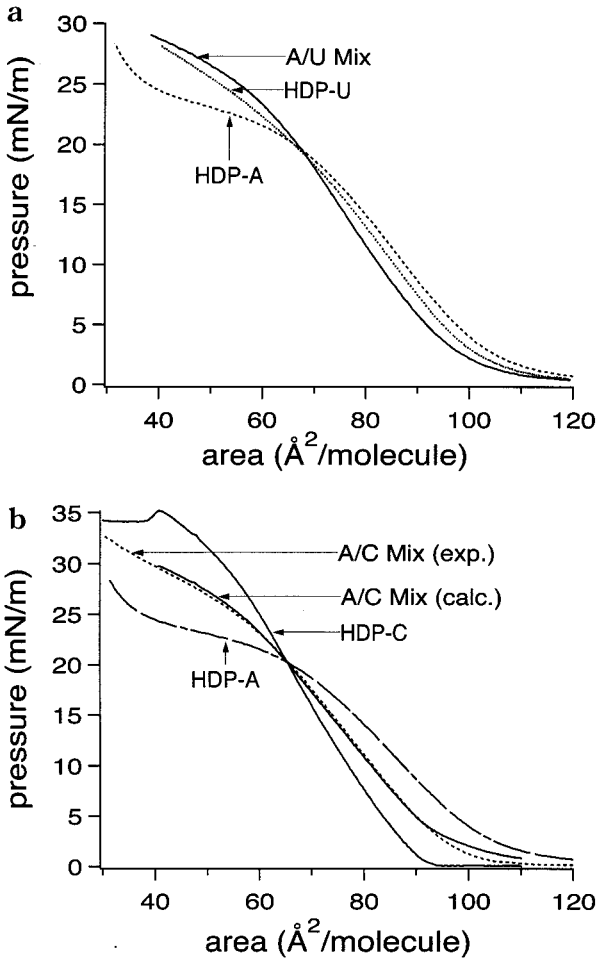

Figure 3. Area−pressure isotherms of an equimolar mixture of (a) **6a/b** and (b) **6a/d**. For comparison, the isotherms of the pure compounds are given. Subphase conditions: 10 mM HEPES, 150 mM NaCl, 1 mM $CaCl_2$, pH 7.5, 5 °C.

a larger specific area than the uridine analogue **6b** in the high-pressure region, indicating that the phase behavior is strongly influenced by the headgroup. The large headgroups of the adenosine and guanosine derivatives might explain why the specific area is shifted to approximately 27 Å²/molecule at the collapse pressure compared to roughly 20 Å²/molecule for a single alkyl chain.

The isotherms of the guanosine analogue **6c** show a very unique phase behavior. Here, the transition from the gas- to the solid-analogue phase occurred in a narrow range at approximately 40 Å²/molecule. It is known that guanosine can form strong hydrogen bonds with itself,[40] leading to dimers and tetramers.[41] Closely packed oligomers explain the area−pressure isotherms of HDP-G. The recognition units of such an oligomeric complex would be located in the center, while the outer shell is formed by the ribose and phosphate units. Such an outer sphere is more hydrophilic, and the stronger hydration could immerse the molecules deeper in the subphase region, making the recognition process to all other nucleobases almost impossible.

Mixtures of the Monoalkyl Ribonucleotides. The isotherm of an equimolar mixture of the adenosine and uridine compound is shown in Figure 3a. Here, we can clearly demonstrate that the phosphoalkyl adenosine and phosphoalkyl uridine interact. At low pressure, the preorganization of the system shifts the mixing isotherm to smaller area/molecule values reflecting a closer packing of the alkyl chains due to the base pairing. In this range (up to 15 mN/m) no phase transition is observed. Above a certain point, the situation is inverted and the values are shifted to larger areas. This indicates that base pairs are not as compressible as single molecules; however, phase transition above a lateral pressure of 15 mN/m cannot be excluded.

To investigate whether this recognition process is specific, cross experiments were performed using the HDP-A/HDP-C and the HDP-U/HDP-C system. As an example, the experimental isotherms of HDP-A, HDP-C and the equimolar mixture of both are given in Figure 3b. Additionally the calculated isotherm based on the partial pressure and an ideal mixing behavior of the pure compounds is included.

In contrast to the HDP-A/HDP-U system, we did not find an indication for a strong interaction, which is also demonstrated by the pressure difference of the experimental and calculated ideal mixing isotherms (Figure 4a). In the case of the HDP-A/HDP-U pair, a specific interaction is reflected by the strong deviation from the ideal mixing behavior. By contrast, the mismatch pairs interact only

(40) Diederichsen, U.; Schmitt, H. W. *Tetrahedron Lett.* **1996**, *37*, 475−478.

(41) Kurz, M.; Göbel, K.; Hartel, C.; Göbel, M. W. *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 842−845.

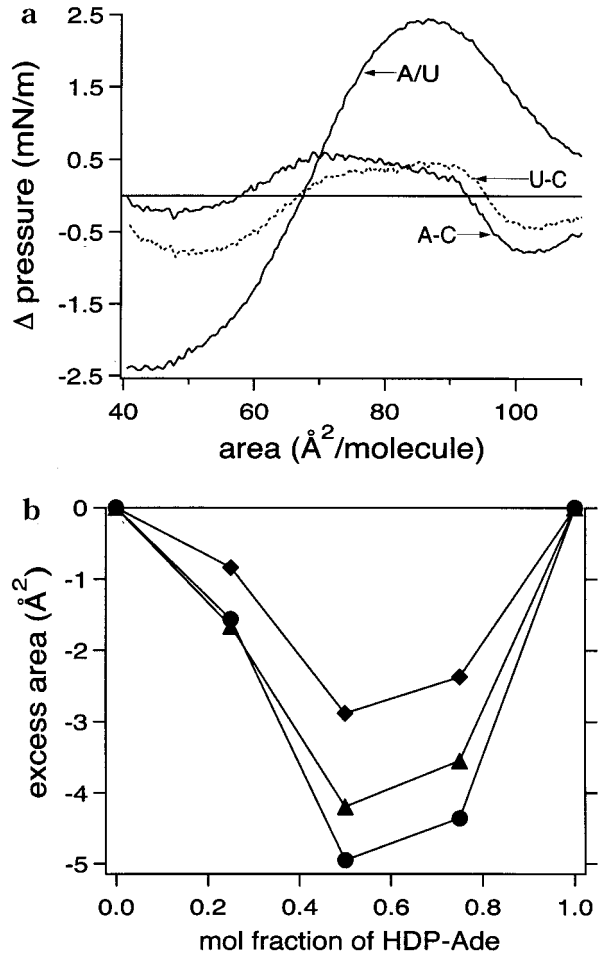

Figure 4. (a) The deviation from the ideal mixing behavior as a function of the specific area for HDP-A/HDP-U, HDP-A/HDP-C, and HDP-C/HDP-U is shown. (b) The excess area is given as a function of the mole fraction of HDP-A for the mixture of HDP-A/HDP-U at lateral pressure of 4 mN/m (●), 10 mN/m (▼), and 14 mN/m (◆).

to a very small extent. An alternative presentation is based on the excess area values describing the difference in packing density of the blended components (Figure 4b). The excess area is given by

$$\Delta A_e = A - (c_1 A_1 + c_2 A_2) \quad (1)$$

where A is the area of the blend while c_1 and c_2 are the mole fractions and A_1 and A_2 are the areas of the single components. So ΔA_e is simply the area difference between the mixture and the ideal behavior. Therefore lowering the excess area indicates a denser packing of the RNA amphiphiles. Interestingly, the HDP-A/HDP-U pair shows the most dense packing for a 1:1 mixture, which is surprising since Watson−Crick or Hoogsteen pairing rules are strongly related to stereochemical order which drastically differs from the situation at the air−water interface. Only a negligible difference from the ideal behavior is observed for the mismatch pairs. According to the Goodrich method[42] we have calculated the mixing energy following

$$\Delta G_{Mix} = \int_0^{\pi} (A - c_1 A_1 - c_2 A_2)\, d\pi \quad (2)$$

(42) Goodrich, F. C. Proceedings of the 2nd International Congress on Surface Activity; Butterworth: London, 1957.

The calculated excess energy of mixing for the 1:1 mixture is (in a pressure range of 4 mN/m) around 0.1 kJ/mol. This is far below a binding energy of a hydrogen bond. Since the RNA amphiphiles are already hydrogen bonded to the surface water and base pairing releases that water, we believe that the process is mostly entropically driven. Specific base-pair formation of the self-organizing RNA amphiphiles is induced due to the high surface concentration, the entropically fixed water at the interface, and steric requests.

The mixing isotherms for HDP-G with the adenosine, uridine, and cytosine analogue show a very different behavior. The isotherms are characteristic for pure HDP-A, HDP-U, or HDP-C, which are shifted only to smaller specific area due to the dilution. This further indicates that the guanosine compound **6c** forms self-aggregates preventing the interaction with the other compounds (data not shown).

Conclusions

In this report, the synthesis of novel RNA amphiphiles is described. In the presence of divalent ions, the nucleotide amphiphiles self-organize into stable monolayers at the air−water interface, allowing an investigation of the recognition process in two dimensions. Specific base pairing within self-assembled monolayers of adenosine/uridine analogues was demonstrated by their mixing π/A isotherms. No indications for a mismatch pair were found. The system has the great advantage that the alkyl chain does not interfere with the nucleotide headgroup and the π/A isotherms reflect mainly the interactions of the nucleotide compounds. However, although studies at the air−water interface are most suited for the investigation of specific interactions between self-organizing amphiphiles, they lack sensitivity by combining those directly with well-known techniques for RNA or DNA investigation in solution such as UV or IR spectroscopy. Studies on solid-supported or multilayer systems will bypass the problem. However, in this case interactions between different layers or the solid support disturbing base-pair formation must be taken into account.

In the future, transferring the self-assembled base pairs to solid substrates might allow the building of devices using RNA or DNA analogues as information storage. Experiments have to focus on preserving the specific RNA−RNA contacts after transfer onto solid substrates, on the preorganization of other sequences, and on a tighter anchoring of the monoalkyl nucleotides. Further, we will focus on interlayer recognition processes of RNA amphiphiles.

Experimental Section

The hexadecylphosphocholine **4** was synthesized with slight modifications as described by Erukulla et al.[37] The nucleosides were linked to the hexadecylphosphocholine **4** in an enzymatic transphosphorylation[38] as shown in Figure 1. To a solution of 8 mmol of nucleoside in 20 mL of acetate buffer (200 mM NaOAc, 250 mM $CaCl_2$, pH 6.0) 900 units of Phospholipase D and 700 mg (1.6 mmol) of choline **4** in 20 mL of $CHCl_3$ were added, while the solution was stirred at 40 °C under argon. After 10 min the solution turns turbid. The reaction was monitored by thin-layer chromatography and stopped after 5 h by adding 5 mL of 0.1 M HCl. The mixture was extracted five times with $CHCl_3$/MeOH (2:1), and half of the organic phase was removed in a vacuum.

Hexadecylphosphoadenosine (6a). To purify **6a**, the mixture was cooled to −10 °C and the adenine product **6a** was precipitated as ammonia salt by adding NH_3/H_2O. The yield was 421 mg (43%). R_f (MeOH/$CHCl_3$/NH_3H_2O 3:11:0.1) = 0.15. ^{1}H NMR ($CDCl_3$/MeOH 1:3): 8.32 (s; H−C(8)), 8.15 (s; H−C(2)), 5.91 (d; J = 5.5 Hz; H−C(1′)), 4.59 (dd; J = 5.2 Hz; 1H), 4.20 (dd;

J = 4.2 Hz; 1H), 4.08 (m; 2H), 3.98 (m; 1H), 3.80 (dd; J = 6.8 Hz; 2H), 1.49 (m; 2H, H−C(2″)), 1.22 (m; 26H), 0.84 (t; J = 6.7 Hz; 3H; Me). ^{31}P NMR ($CDCl_3$/MeOH): 0.13. MS-FAB: 572 [80; M + 2], 178 [9; M − 392], 136 [100; M − 434].

Hexadecylphosphouridine (6b). The crude mixture from the reaction for **6b** was poorly soluble in a mixture of MeOH/$CHCl_3$/NH_3H_2O 3:11:0.1. So the material was divided in three parts and subjected to silica chromatography (MeOH/$CHCl_3$/NH_3H_2O 3:11:0.1). Due to the chromatography we have lost $^2/_3$ of the material. Starting from 700 mg of choline **4** the yield was 253 mg (27%). R_f($CHCl_3$/MeOH/H_2O 65:25:4) = 0.33. ^{1}H NMR (d_6-DMSO): 11.29 (s, N−H), 7.77 (d; J = 8.1 Hz; H−C(5)), 7.37−7.07 (br 4H; NH_4^+), 5.78 (d; J = 5.5 Hz; H−C(1′)), 5.59 (d; J = 8.0 Hz; H−C(6)), 4.05 (t; J = 5.1 Hz; 1H), 3.99−3.93 (br m; 4H), 3.75 (m; 2H; H−C(1″), 1.50 (m; 2H; H−C(2″)), 1.23 (m; 26H), 0.85 (t; J = 6.67 Hz; 3H; Me). ^{31}P NMR (d_6-DMSO): −1.18. MS-FAB: 593 [17; M + 2Na], 571 [21; M + 1 + Na], 549 [8; M + 2], 176 [79; M − 371], 136 [100; M − 411].

Hexadecylphosphoguanosine (6c). The guanosine compound **6c** is hardly soluble in common solvents. Therefore chromatography was impossible. So the crude mixture was washed 30 times with HCl/H_2O (pH 3) and extracted with $CHCl_3$/MeOH 2:1. The solution turns milky, and phase separation takes approximately 20 min. Then the solvent was evaporated, and the crude mixture is washed several times with $CHCl_3$/MeOH/H_2O 65:25:4. The insoluble portion is filtered off and dried in a vacuum. Yield: 125 mg (15%). ^{1}H NMR (d_6-DMSO): 10.61 (s; N−H), 7.91 (s; H−C(8)), 6.53 (br 2H, NH_2), 5.68 (d; J = 5.8 Hz; H−C(1′)), 5.49−5.41 (br 2H; OH), 4.46 (br m; 1H), 4.16 (br m; 1H), 3.96 (br m; 1H), 3.85 (br m; 2H, H−C(5′)), 3.63 (m; 2H; H−C(1″)); 1.44 (m; 2H; H−C(2″)), 1.20 (m; 26H), 0.84 (t; J = 6.2 Hz; 3H; Me). MS-FAB: 610 [85; M + 1 + Na], 588 [25; M + 2], 560 [10; M − 26]; 174 [20; M − 412], 152 [100; M − 434], 136 [70; M − 450].

Hexadecylphosphocytidine (6d). The product was purified by chromatography on silica gel ($CHCl_3$/MeOH/NH_4OH 65:80:8). R_f($CHCl_3$/MeOH/NH_4OH = 68:80:8) = 0.36. ^{1}H NMR ($CDCl_3$/MeOD 1:1): 8.1 (d, J = 7.5 Hz, H−C(6)), 6.01 (1H, d, J = 7.5 Hz, H−C(5)), 5.96 (d, J = 5.2 Hz, H−C(1′)), 4.23 (m, 2H), 4.16 (m, 2H), 4.12 (br m, 2H), 3.87 (d, J = 6.4 Hz, 1H), 1.63 (m, 2H), 1.26 (m, 26H), 0.88 (t, J = 6.7 Hz, 3H; Me). ^{31}P NMR ($CDCl_3$/MeOD) = 0.7235. MS-FAB: 592 [8; M + 2Na], 570 [24; M + 1 + Na], 548 [52; M + 2], 174 [11; M − 372], 136 [100; M-410].

Langmuir−Blodgett (LB) measurements were recorded on a self-built LB trough (420 × 90 mm) with a Wilhelmy system.[43] Measurements were performed on HEPES-buffered saline (10 mM HEPES, 150 mM NaCl, pH 7.5). All isotherms were reproduced at least three times within a range of 0.1 mN/m. Films were stable up to 30 mN/m.

Acknowledgment. We thank Ingmar Dorn, Rudolf Merkel, Klaus Neumaier, and Erich Sackmann for helpful discussions as well as Wolfram Schäfer and Isolde Sonnenbichler for MS and NMR measurements. This work was supported by the Deutsche Forschungsgemeinschaft (DFG) and the European Commission (BIOTECH program).

LA980515S

(43) Schmitt, L.; Dietrich, C.; Tampé, R.; *J. Am. Chem. Soc.* **1994**, *116*, 8485−8491.