enzymes that catalyze hydrolytic or reversible hydration–dehydration reactions, such as enolase[18] and leucine aminopeptidase.[19] For each of these enzymes the multinuclear metal center functions in substrate binding and in catalysis, suggesting that the binuclear Mn(II) center of arginase may have an analogous function. In addition, it is noteworthy that the X-band EPR spectrum for arginase shown in Figure 1A is remarkably similar to that reported for the Mn(II)–Mn(II) oxidation state of the Mn-catalase from *Thermus thermophilus*.[20] X-ray diffraction studies of the Mn-catalase indicate that the two metal ions are separated by 3.6 Å; however, protein ligands to the metal ions have not been identified.[21] Structure–function analysis of the binuclear Mn(II) center of arginase is the focus of our ongoing spectroscopic and crystallographic studies.

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## Self-Assembled Multifunctional Receptors for Nucleotides at the Air–Water Interface

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We report here that multifunctional receptors specific for mononucleotides are spontaneously formed from guanidinium-functionalized monolayers and their combination with nucleobase-containing monolayers.

The recognition process of the artificial receptors for nucleotides and oligonucleotides is facilitated through multifunctional interactions which combine ionic pairing, aromatic stacking, and complementary hydrogen bonding.[1-4] Such multifunctional receptors may be realized more readily by utilizing self-assembly of amphiphilic molecules at the air–water interface. We established that Langmuir monolayers at the air–water interface served as powerful, selective hosts for a variety of biorelated water-soluble compounds via hydrogen bonding[5,6] or ionic pairing.[7] In particular, a guanidinium-functionalized monolayer of **1** specifically recognized phosphate units of AMP and ATP, with formation of the guanidinium/phosphate pair through ionic and hydrogen-bonding interactions.[7] We intended to achieve discrimination of different nucleotides by the addition of nucleobase monolayer components **2** and **3**.[8]

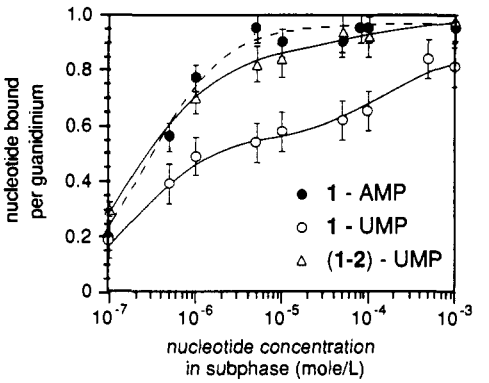


**Figure 1.** Nucleotide binding to monolayers of **1** and **1-2**. Solid lines are theoretical adsorption curves obtained by using $K$ and $\alpha$ values from Table I.

**Table I.** Binding of Mononucleotides to Guanidinium-Functionalized Monolayers

| amphiphile | substrate[a] | $10^6 \times K$,[b] $M^{-1}$ | $\alpha$[b] |
|---|---|---|---|
| **1** | AMP | 3 ± 0.5 | 1.0 |
| **1-2** | AMP | nonspecific[c] | |
| **1-3** | AMP | 8 ± 2 | 0.7 |
| **4** | AMP | 3 ± 1 | 1.0 |
| **1** | UMP | 5 ± 1 | 0.6 |
| **1-2** | UMP | 5 ± 2 | 0.9 |
| **1-3** | UMP | 6 ± 2 | 0.6 |
| **4** | UMP | 2 ± 1 | 1.0 |

[a] AMP (Oriental yeast, 99%) and UMP (Sigma, 98%) were used as received. [b] The adsorption equation is applied to the concentration range where simple substrate saturation is observed. [c] Binding saturation is not observed within the concentration range of this study.

Monolayer characteristics of **1** have been described briefly.[7] Monolayers consisting of equimolar mixtures of **1** and **2** and of **1** and **3** showed good mixing behavior as inferred from nonlinear changes in surface pressure with component ratios.

Monolayer-bound nucleotides were determined by XPS analyses of LB films transferred from the aqueous nucleotide-laden subphase ($10^{-7}$–$10^{-3}$ $M^{-1}$).[5-7] A saturation phenomenon in the binding curve (Figure 1) indicates the presence of a specific binding site. Binding constants, $K$, and the fraction of the occupied guanidinium sites at saturation, $\alpha$, were determined by fitting these binding data to a general adsorption isotherm via iteration[10] and are given in Table I.

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(9) Details to be reported in an upcoming full paper. Gaines, G. L., Jr. *Insoluble Monolayers at Liquid-Gas Interface*; John Wiley & Sons, Inc.: New York, 1966.
(10) A general adsorption equation is given by

$$n = \alpha[S]/(1/K + [S])$$

where $n$ is the number of substrate bound per guanidinium group, $\alpha$ is the number of substrate bound per guanidinium group at saturation binding, [S] is the substrate concentration in the subphase, and $K$ is the binding constant. Marshall, A. G. *Biophysical Chemistry: Principles, Techniques, and Applications*; John Wiley & Sons, Inc.: New York, 1978.

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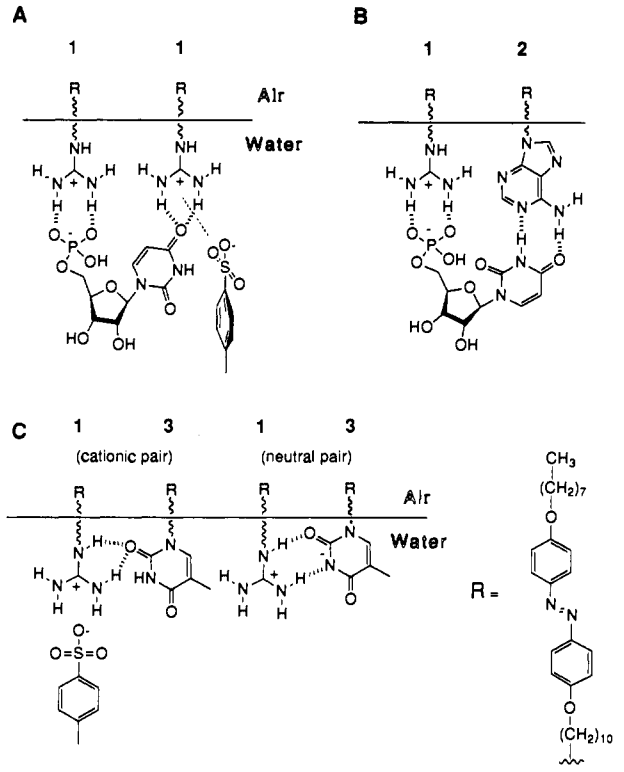


Figure 2. Monolayer receptors from amphiphiles **1**, **2**, and **3** and plausible schemes of nucleotide binding.

AMP is bound specifically to the guanidinium monolayer by the formation of the guanidinium/phosphate pair (Figure 1).[7a] A single set of parameters describes the binding behavior: $K = 3 \times 10^6$ M$^{-1}$, $\alpha = 1.0$ AMP/guan. The $\alpha$ value of 1.0 reveals that AMP binds to the guanidinium monolayer in a 1:1 correspondence. In contrast, UMP displays a binding saturation of 0.5–0.6 at $10^{-7}$–$10^{-5}$ M, and secondary binding occurs at higher UMP concentrations. The electrostatic interaction alone cannot explain this unique behavior, since UMP shows a simple equimolar saturation toward trimethylammonium monolayer **4**. The guanidinium unit is known to interact with the uracil carbonyl groups in protein–DNA/RNA complexes.[11] Thus, UMP can bind to monolayer **1** via both of the guanidinium–phosphate and guanidinium–uracil pairs (see Figure 2A).[12] An enhanced binding constant for UMP relative to that for AMP supports this interpretation.

The newly found role of the guanidinium monolayer is endorsed by bicomponent receptor **1–2** which combines guanidinium and adenine units. An equimolar saturation behavior ($\alpha = 0.9$ UMP/guan) is observed for UMP. This can be explained by assuming the formation of complementary adenine/uracil pairs as the secondary interaction (Figure 2B). As expected, AMP substrate does not display specific binding toward this bifunctional monolayer. Secondary interactions of the adenine component with AMP appear to interfere with the formation of specific complexes.

A third multifunctional receptor was prepared by a 1:1 mixed monolayer of **1** and **3**. Although this mixed monolayer exhibits saturation toward AMP and UMP, all of the guanidinium sites are not occupied at saturation ($\alpha < 1$). An IR spectrum of the transferred monolayer **1–3** exhibits shifts of the $\nu_{C=N}$ (**1**) and $\nu_{C2=O}$ (**3**)[13] peaks around 1700 cm$^{-1}$ by >20 cm$^{-1}$ relative to those of the single-component monolayer of **1** and **3** together with the appearance of a new peak at 1522 cm$^{-1}$, indicating hydrogen-bond formation between guanidinium and thymine head groups. XPS measurements showed a 25–30% reduction of bound anionic species (i.e., *p*-toluenesulfonate and nucleotides) at all nucleotide concentrations. The forced proximity of head groups may promote deprotonation of the thymine unit to form guanidinium/thymine ion pairs[14] and cause IR spectral changes and release of *p*-toluenesulfonate ion. The neutral ion pairs thus formed cannot bind nucleotides, thereby yielding [illegible]alues. The **1–3** pair in which the thymine unit is *not* d[illegible]ted acts as a specific receptor toward AMP and UMP.[15] [illegible]ding constant of AMP toward receptor **1–3** is enhanced (2.7 times) relative to that toward receptor **1**. In contrast, UMP shows virtually the same binding constants. The enhanced AMP binding appears to be induced by cooperative interaction of the guanidinium and thymine units.

The present findings amply demonstrate the versatility of guanidinium-based monolayer receptors. Spontaneous assembly of secondary recognition units gives rise to varied modes of nucleotide binding.

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**Supplementary Material Available:** IR spectra of **1**, **3**, and **1–3** deposited from water and of **1–3** deposited from UMP and AMP and a plot of the binding curve of AMP and UMP to the guanidinium monolayer (5 pages). Ordering information is given on any current masthead page.

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(12) The average guanidinium–guanidinium distance in monolayer **1** can be calculated from the surface pressure–molecular area ($\pi$–$A$) isotherm to be 7.4 Å on 0.1 mM aqueous UMP. This distance is consistent with the binding mode described in Figure 2A, because the distance between the phosphate group and the carbonyl group of UMP in this binding mode is 7.5 ± 0.5 Å.

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(15) The IR peak at 1522 cm$^{-1}$ is ascribable to the neutral ionic pair of the guanidinium and thymine groups. This peak disappears completely at $10^{-3}$ M aqueous AMP where all of the guanidinium groups are expected to interact with the phosphate group of AMP (AMP/guanidinium = 1.2) but not with the thymine group of **3**. This observation supports formation of complexes as illustrated in Figure 2C.

## Larger and More Weakly Coordinating Anions: $Nb(OTeF_5)_6^-$ and $Ti(OTeF_5)_6^{2-}$

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The ability to generate coordinative unsaturation for a wide variety of cationic species (e.g., $[SiR_3]^+$,[1] $[Fe(Por)]^+$,[2,3] $[Re(Cp)(NO)(PPh_3)]^+$,[4] $[ZrCp'_2R]^+$[5]) in solution remains an elusive goal for synthetic and catalytic chemists because no solvent or anion is truly noncoordinating. Recent examples of larger and more weakly coordinating anions include fluorinated derivatives of $BPh_4^-$ such as $B(C_6F_5)_4^-$ and $B(3,5\text{-}C_6H_3(CF_3)_2)_4^-$,[6] $CB_{11}H_{12}^-$

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(3) Abbreviations: Por = any porphyrinate dianion; TBA = tetra-*n*-butylammonium cation; teflate = pentafluorooxotellurate ($OTeF_5$ or $OTeF_5^-$).

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