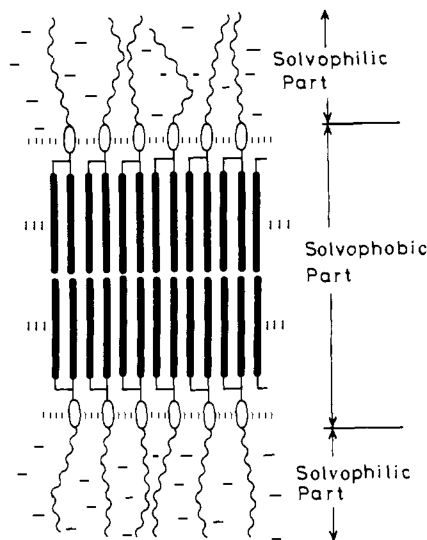

Figure 3. Schematic illustration of the bilayer structure of **2** and **3** in organic media.

of the solution were applied to carbon-coated Cu grids and dried. A few drops of lead(II) bis(acetylacetonate) in CH_3OH were then added as the staining agent, and the samples were observed by a transmission electron microscope (Hitachi, H600). As shown by Figure 1, the stained samples reproducibly contained twisted tapes (width, 1600 Å) and smaller vesicles (diameter, 200–1000 Å). The inner core of the vesicles cannot be seen in Figure 1b, because of the poststaining method we employed. In a direct observation of the cyclohexane dispersion by a dark-field optical microscope (Olympus BH-2), numerous fiber-like aggregates (width less than 1 μm) were found. These microscopic observations of varied morphologies reflect different degrees of development of the molecular aggregate, since vesicles were dominant in samples prepared at 25 °C and the tape and lamella morphologies were abundant at 15 °C.

Compound **3** gave lamellar and rod-like morphologies in benzene (layer thickness, ca. 100 Å), as will be discussed in detail elsewhere.[12]

Figure 2 compares circular dichroism (CD) spectra of **1** in water and **2** in $CHCl_3$ and cyclohexane. CD peaks due to the phenylene unit are observed at 250–300 nm. The fluorocarbon bilayer of **1** in water displays a very small peak ($[\theta]_{266}$(15 °C) 7000), in contrast with a related hydrocarbon bilayer which gives a $[\theta]_{260}$ value of 4×10^5 at temperatures below the phase transition.[13] On the other hand, fluorocarbon compound **2** at low temperatures gives strong CD peaks in cyclohexane: $[\theta]_{266} = 1.3 \times 10^5$ at 6 °C. The intensity is lessened with rising temperature. The solvent used is crucial, and the signal is much smaller in $CHCl_3$. It has been shown that much-enhanced CD spectra of *aqueous* chiral bilayers are derived from exciton coupling among the organized chromophore.[13] Thus, the observed CD data indicate that the component molecules in the aggregate are highly organized in *cyclohexane*.

The data shown in Figures 1 and 2, together with the structural similarity of component molecules with common, bilayer-forming compounds, strongly suggest that the molecular bilayer is the fundamental building unit of the aggregate, as schematically illustrated in Figure 3. Macroscopic and microscopic assemblage of amphiphilic molecules in organic media has been reported in the past. For example, a phospholipid (dipalmitoylphosphatidylcholine) formed reversed micelles in aromatic solvents[14] and macroscopic gels in aliphatic solvents.[15] *N*-Octylaldonamides similarly formed gels in xylene.[16] We demonstrated recently that the Ca^{2+} complex of synthetic phosphate bilayers retained their molecular organization in organic solvents.[17] The polar interaction is the dominant driving force of molecular assembly in these instances. In contrast, the organized molecular assembly in the present case is formed by the use of the solvophobic property of the fluorocarbon chain.

The present study together with our previous finding implies that compartmentalization of the molecular space can be realized even in organic media.

Registry No. 2, 122948-36-1; **3**, 122948-37-2.

Supplementary Material Available: Electron micrographs of bilayer aggregates of related fluorocarbon amphiphiles (1 page). Ordering information is given on any current masthead page.

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Convenient Procedure for the Preparation of Specific Mixed DNA–RNA Polymers

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The chemical synthesis of specific DNA oligomers[1] and, more recently, RNA oligomers[2] has become routine in many laboratories. With the development of the alkylsilyl groups as the 2′-hydroxyl protecting groups, in combination with the phosphoramidite coupling procedure for assembling the oligoribonucleotide chain, the synthesis of RNA oligomers can be achieved as efficiently as is the case for DNA oligomers.[3,4] In this communication, we describe a convenient procedure for the solid-phase synthesis of specific DNA–RNA mixed polymers using deoxyribonucleoside phosphoramidite and 2′-silylated ribonucleoside phosphoramidite intermediates. While van Boom[5] has recently reported the solution synthesis of a mixed polymer where a deoxyribonucleotide fragment is linked to a ribonucleotide fragment, the method we describe allows for the synthesis of specific nucleotide sequences with deoxyribonucleotides and ribonucleotides being interspersed. Such mixed DNA and RNA nucleotide polymers will be useful in the study of biological processes and

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Table I. Synthetic Mixed DNA-RNA Oligomers

no.	sequence	Rm[a]
1	ACGGUCU*dC*ACGAGC	0.78
2	GCUCGUC*dTdGdA*U*dGdA*GUCCGUGAGGAC*dGdAdA*AGACCGU	0.36
3	GCUCGUCUGAU*dGdA*GUCCGUGAGGAC*dGdAdA*AGACCGU	0.35
4	GCUCGUCUGAU*dGdA*GUCCGUGAGGACGAAAGACCGU	0.34

[a] Rm is the distance relative to Bromophenol Blue on 20% polyacrylamide/7 M urea gel electrophoresis.

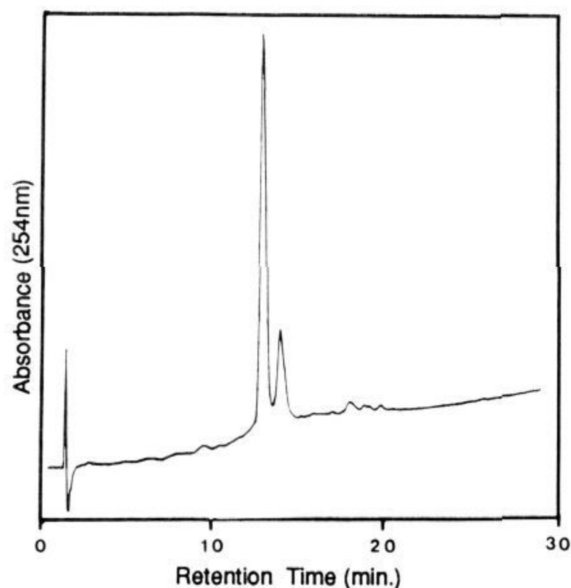

Figure 1. HPLC profile of the crude product from the synthesis of the polynucleotide ACGGUCU*dC*ACGAGC. HPLC conditions: column, AQUAPORE RP-300 (4.6 × 100, ABI); solvent, linear gradient of CH_3CN from 7 to 20% over 30 min in triethylammonium acetate (0.1 M, pH 7); flow, 1 mL/min.

in determining structure/function relationships involving nucleic acids.

The automated solid-phase synthesis of RNA polymers using 2′-silylated nucleoside phosphoramidites is fully compatible, in terms of equipment and reagents, with DNA synthesis using deoxyribonucleoside phosporamidites. The difference between the two syntheses is the use of ribonucleoside instead of deoxyribonucleoside phosphoramidites in RNA synthesis. The reaction time for coupling and detritylation steps in RNA synthesis is slightly longer.[4a] Deprotection of assembled DNA and RNA nucleotide chains is similar, but for RNA synthesis, a desilylation step is needed to remove the 2′-silyl protecting groups. This overall similarity allows for the synthesis of a specific sequence of mixed oligonucleotides with interspersed deoxyribonucleotides and ribonucleotides.

To illustrate the procedure, a 14-mer ribonucleotide with a deoxycytidine inserted at the eighth position (**1**, Table I) was prepared. The base sequence is derived from the substrate of the previously described ribozyme reaction.[6] The assembly of the oligonucleotide was performed on an automated nucleic acid synthesizer (Applied Biosystem 380B). 5′-Monomethoxytritylated, 2′-*tert*-butyldimethylsilylated (triisopropylsilyl for guanosine), N-protected (benzoyl for cytidine, either benzoyl or phenoxyacetyl for adenosine, phenoxyacetyl for guanosine) ribonucleoside methylphosphoramidites and the derivatized CPG (controlled pore glass) were prepared as previously described.[4,7] The deoxyribonucleoside cyanoethylphosphoramidites were obtained from Pharmacia and were added through the extra base ports on the synthesizer. The synthesizer was set up such that the ribonucleotide units were assembled using the standard RNA synthesis

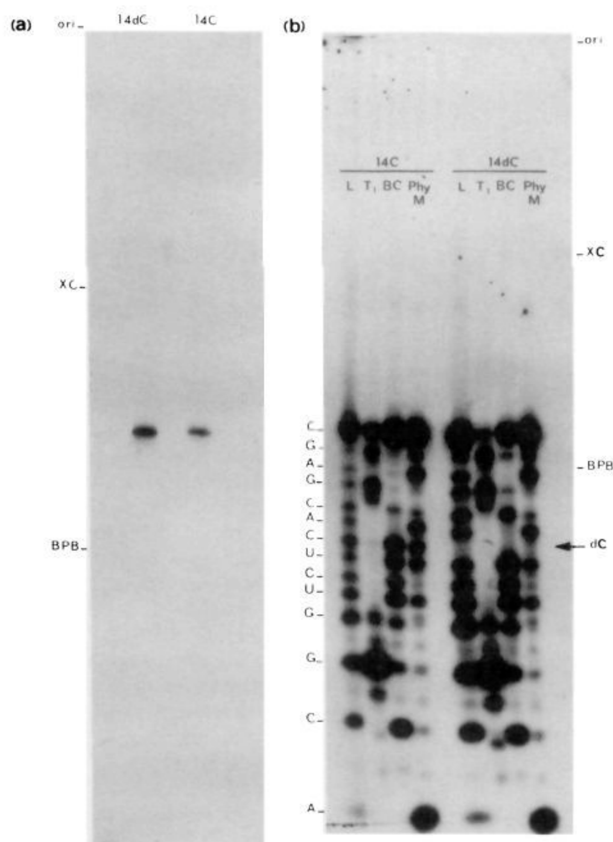

Figure 2. (a) A 20% polyacrylamide/7 M urea gel electrophoresis of the gel-purified 5′-end-labeled polynucleotide ACGGUCU*dC*ACGAGC (14dC) and the polyribonucleotide ACGGUCUCACGAGC (14C). (b) A 15% polyacrylamide sequencing gel of 5′-end-labeled ACGGUCU*d*-*C*ACGAGC (14dC) and ACGGUCUCACGAGC (14C). Letters represent the following: L, formamide ladder; T_1, ribonuclease T_1 (specific for G); BC, ribonuclease from *Bacillus cereus* (specific for C and D); Phy M, ribonuclease Phy M (specific for A and U). XC = Xylene Cyanol; BPB = Bromphenol Blue.

cycle and the deoxyribonucleotide units were assembled by using the DNA synthesis cycle. This is readily achieved by the full programming flexibility on the automated synthesizer. The average coupling yield for the assembly was 97% as determined by UV quantitation of the released trityl cation (478 nm for monomethoxytrityl and 504 nm for dimethoxytrityl). The assembled chain was deprotected by following the same steps as those for ribonucleotides. The methyl and cyanoethyl phosphate protecting groups were removed by thiophenoxide (thiophenol/dioxane/triethylamine, 1/2/2). We have recently shown that deacylation with anhydrous methanolic ammonia instead of aqueous ammonium hydroxide avoids the cleavage of the assembled ribonucleotide chain,[7a] and these conditions were used during the deprotection of the 14-mer. Treatment of the nucleotide chain by saturated methanolic ammonia for 16 h resulted in cleavage from the solid support and removal of the phenoxyacetyl protecting group on guanosine and adenosine residues as well as the benzoyl group on cytidine and deoxycytidine. The product was then treated with tetrabutylammonium fluoride (1 M in THF) for 12 h to remove silyl groups from the ribonucleotide units in the chain. The reaction mixture was desalted on the Sephadex G25 instru-

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ment to give the crude synthetic product. The HPLC profile for the crude products is shown in Figure 1. The mixed oligonucleotide was found to have a longer retention time (t_R 13.5 min) than the corresponding all-ribonucleotide sequence (t_R 11.5 min) prepared by following standard RNA synthesis procedures.[4a]

The mixed oligomer was fully characterized by gel electrophoresis[8] (Figure 2a) and the RNA gel-sequencing technique[9] (Figure 2b) and compared to the corresponding RNA sequence. Comparison of the sequences in Figure 2b shows the absence of ribo C in the mixed 14-mer. Several 35-unit hybrid DNA-RNA oligomers related to the sequence of the self-cleaving ribozyme[6] were prepared (**2**, **3**, **4**, Table I) and their sequences established.

In conclusion, the procedure described herein allows the facile preparation of heretofore unavailable (deoxy)ribonucleotide polymers. Several of these synthetic polymers related to the sequence of the ribozyme and its substrate have been made and are currently being used to probe the mechanism of RNA catalysis.[10]

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New Approach to Mesophase Stabilization through Hydrogen-Bonding Molecular Interactions in Binary Mixtures

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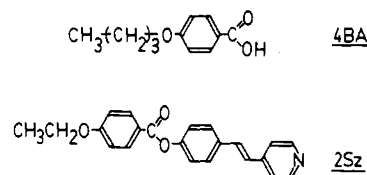
We report a new approach to novel liquid crystalline moieties having a greatly enhanced mesomorphic range through the formation of intermolecular hydrogen bonds between two dissimilar mesogens.

Intermolecular interactions contribute greatly to the formation of molecular aggregates. In liquid crystals, mesomorphicity[1] results from a proper combination of the shape of a molecule and the magnitude and direction of molecular interactions between molecules. While the importance of dipole-dipole interactions in the formation of mesophases has long been established, we have hypothesized that the occurrence of intermolecular hydrogen bonding should have great potential for ordering thermotropic liquid crystals because H-bonding is much stronger than dipole-dipole interactions. However, with some notable exceptions described below, this approach to mesophase formation has remained unsuccessful as, in most cases, the magnitude and direction of H-bonded interactions are not appropriate to keep the fluid state ordered.

A survey of the literature shows that a few monosaccharides[2,3] and aromatic acid derivatives[4] show mesophase formation because of the existence of a stable dimer formed through hydrogen bonding. In all of these cases, the compounds always exist as symmetrical dimers in both their crystalline and mesophase states and the dimer itself may be considered as a single component.

Our approach has been to build a new liquid crystalline system through intermolecular hydrogen bonding between two *different* and *independent* components. The mixing of H-bond donor and acceptor moieties can be expected to form a liquid crystalline hydrogen-bonded complex if the structure is designed properly.

In the present study, 4-butoxybenzoic acid (**4BA**) (nematic from



147 to 160 °C) and *trans*-4-[(4-ethoxybenzoyl)oxy]-4′-stilbazole (**2Sz**), containing a 4-pyridyl group[5,6] (nematic from 165 to 213 °C) at the extremity of its mesogen, have been selected as H-bond donor and acceptor moieties, respectively. These two molecules have the appropriate structure to form a hydrogen-bonded complex with an overall molecular geometry directed along the long axis of the individual rodlike molecules. In particular, the lone pair of the 4′-pyridyl group of **2Sz** is directed along the molecular axis of the mesogen.

In order to break the self hydrogen bonding of the 4-butoxybenzoic acid dimer units, the complex resulting from an equimolar binary mixture of **4BA** and **2Sz** was prepared by slow evaporation of the mixture from pyridine solution.[7] The binary complex that was obtained showed an exceedingly stable mesophase as confirmed by differential scanning calorimetry (DSC) (Mettler DSC 30) and by hot-stage polarizing microscopy (Mettler FP 82).

The DSC data shown in Figure 1 (curve B) features endothermic peaks at 136 and 238 °C corresponding respectively to melting (T_m) and isotropization (T_i) transitions for the 1:1 mixture. The isotropization temperature deviated noticeably in the positive direction, and the mesomorphic range ($T_i - T_m$) of the mixture was extended to 102 °C as compared to 13 °C for **4BA** (curve C) and 48 °C for **2Sz** (curve A). Moreover, for the 1:1 mixture, a smectic phase which did not appear for either single component existed between 136 and 160 °C as confirmed by our observation of a focal conic texture under a polarizing microscope. However, this transition was not apparent in our DSC thermogram, and this smectic phase has not yet been classified. The subsequent nematic phase was stable up to 238 °C.

We attribute this unusually strong stabilization of the mesophase of the 1:1 mixture to the *formation of a new and extended mesogen obtained through intermolecular hydrogen bonding* which allows the 1:1 complex to behave as a *single* liquid crystalline component as shown in Figure 2. This is supported by FT-IR measurements which strongly suggest that the acid dimer of **4BA** is replaced by the 1:1 complex. If a simple 1:1 *mechanical* mixture of **4BA** and **2Sz** is prepared, a band at 1681 cm^{-1} due to the carbonyl group of the carboxylic acid dimer (C=O···HO—) of **4BA** is observed. In contrast, measurements on the 1:1 molecular mixture prepared from pyridine solution show a new band at 1704 cm^{-1} replacing the band at 1681 cm^{-1} and indicating the existence of the free carbonyl group of the carboxylic acid complexed with the pyridine

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