Topography of Nucleic Acid Helices in Solutions. IX. Models for the Interactions of Optically Active Diamines, Amino Acid Amides, Diamino Acids, and Lysyl Dipeptides with Nucleic Acid Systems[1]

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Abstract: Molecular framework models of the complexes formed between the salts I, $N^+H_3CHRCONHCH_2CH_2N^+Me_2H \cdot 2Br^-$; II, $N^+H_3C(CO_2^-)H(CH_2)_nN^+H_3 \cdot Cl^-$ (n = 2, 3, and 4); III, $N^+H_3C((CH_2)_4N^+H_3)HCONHCHRCO_2^- \cdot Cl^-$; and IV, $N^+H_3CHRCH_2N^+H_3 \cdot 2Cl^-$ (R = Me and CO_2H) and adjacent phosphate anions of a helical polynucleotide chain have been examined in detail; 15-, 16-, 17-, and 18-membered rings are formed as the result of complexing the salts I–IV to the nucleic acid helix. It is shown that certain conformations are not as favorable as others. Moreover, it is possible to predict which of the two optical isomers of the salts I–IV should interact to a greater extent with a nucleic acid helix. The results using 11 different sets of optical isomers of the salts I–IV on three different helical structures, *i.e.*, poly I–poly C, poly A–poly U, and calf thymus DNA, have been predicted correctly and consistently.

It has been shown recently that several L-amino acid derivatives, *i.e.*, the amides[2] I, $N^+H_3CHRCONHCH_2CH_2N^+Me_2H \cdot 2Br^-$; the diamino acids[3] II, $N^+H_3C(CO_2^-)H(CH_2)_nN^+H_3 \cdot Cl^-$ where n = 2, 3, and 4; and lysyl dipeptides[3] III, $N^+H_3C((CH_2)_4N^+H_3)HCONHCHRCO_2^- \cdot Cl^-$, interact more strongly than the corresponding D enantiomers with nucleic acid helices. In order to understand the nature of these highly selective interactions, molecular framework models of the complexes formed between the nucleic acids and the salts I–III were examined in detail. This paper reports the results of these studies and the extension of the study to other optically active systems, *i.e.*, the salts IV, $N^+H_3CHRCH_2N^+H_3 \cdot 2Cl^-$, where R = methyl and carboxyl group.

Results and Discussion

In order to formulate models for the interaction of the salts I–IV with nucleic acid helices a number of highly reasonable assumptions must be made.

Assumption I. The dipositively charged salts I–IV stabilize the nucleic acid helix by binding to adjacent phosphate anions on the same chain. The arguments in favor of this interpretation are the following. (1) The distance between adjacent phosphate anions on the same chain of a Watson–Crick double-stranded helix is approximately 7 Å, while the smallest distance between phosphate anions on different chains is greater than 11 Å.[4] It is observed that maximum stabilization of the helices by the salts V, $R_1R_2R_3N^+(CH_2)_nN^+R_1R_2R_3 \cdot 2Br^-$, where $R_1 = R_2 = R_3 = H$, occurs at n = 3, in accord with the structural model of helix–diamine complex formed by binding to two adjacent negative charges on the same chain.[5] Moreover, it is impossible to bind the salts of type V for n = 2–4 to two phosphate anions on two separate chains, simply because the distance between the two anions is too large. (2) Maximum stabilization of the helices by the salts V shifts from n = 3 to n = 2 as the cation size of V is made larger in accord with what might be expected by our postulated model. The significance of the change has been discussed at length.[5] (3) The salts V and the amides I have been shown by Gabbay and Shimshak,[6,7] and by Gabbay, Kleinman, and Shimshak[8] to selectively inhibit the RNase-catalyzed hydrolysis of the single-stranded polyadenylic and polyuridylic acids. The inhibition by the diammonium salts V was shown to be due to specific interactions with the polynucleotide chain rather than with the enzyme since it was found that the rate of the RNase-catalyzed hydrolysis of substrates that are not expected to bind the salts V, *e.g.*, cytidine 2′,3′-cyclic phosphate and cytidylyl-3′,5′-adenosine phosphate (CpA), is slightly accelerated in the presence of the diammonium salts V. Morever, maximum inhibition of the RNase-catalyzed hydrolysis of polyadenylic acid at 50.0° and pH 6.20 and of polyuridylic acid at 37.0° and pH 6.20 by the salts V, $R_1R_2R_3N^+(CH_2)_nN^+R_1R_2R_3 \cdot 2Br^-$, where $R_1 = R_2 = R_3 = H$, occurs at n = 3.[6,7] These findings can only be interpreted in terms of binding of the diammonium salts V to adjacent phosphate anions on a single polynucleotide strand. Furthermore, the fact that maximum inhibition occurs at n = 3 is entirely consistent with our previous postulated structural model for the diamine–polynucleotide complex[5] and with the results of other investigators concerning the structure of the single-chain polynucleotide, *e.g.*, polyadenylic acid has been shown to exist primarily as a stacked single chain at neutral pH and ambient temperatures.[9–12] (4)

(1) For part VIII in this series see E J. Gabbay, R. Kleinman, and R. R. Shimshak, *J. Am. Chem. Soc.*, **90**, 1927 (1968).
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Finally, simple electrolytes such as LiCl, NaCl, etc., are well known to stabilize nucleic acid helices with the exception of acid poly A.[4,13] These ions presumably stabilize the helix by lowering the effective charge on the phosphate anions, thus lowering the electrostatic repulsion between strands.[14-16] This effect is also noted with the diammonium salts I–V. For example, binding of these salts to adjacent phosphate groups on the same strand will lower the effective charge of the phosphate anions, and since the opposite strand is also expected to bind to other molecules of I, the net effect will be a lowered electrostatic repulsion between the two strands. The net result will be reflected by an increase in the T_m of the helix–coil transition. Therefore, the relative stabilization of the helical structure with respect to the coil in the presence of the diammonium salts is explained by the lowering of electrostatic repulsion between strands, even though the binding is to adjacent anions on the same strand.

Additional Assumptions. In order to limit the various possible structures for the complex formed between the polynucleotide and the diammonium salts I–V, the following reasonable assumptions have also been made.

Assumption II. If one examines a molecular framework model of a helical stacked polynucleotide chain it is clear that one may distinguish between the two free oxygen atoms of each phosphate group. One of the atoms is pointing into the solvating medium, and the other is relatively inaccessible since it points approximately parallel to the helical axis. The distinction between the two oxygen atoms has been reported for the Watson–Crick model of DNA.[13,17-19] Even more dramatically it has been shown through the X-ray work of Fresco[20] and Rich, *et al.*,[21] that in the double-stranded acid poly A helix the two oxygen atoms are vastly different, *i.e.*, one points away from the solvating medium and into the helical axis while the other is at an angle with respect to the helical axis and points into the medium. It will be assumed that the salts I–V bind to two adjacent phosphate oxygen atoms which are readily accessible, *i.e.*, those which point into the medium. The assumption is indeed reasonable since it is sterically difficult (if not impossible) to bind the diamines in any other manner.

Assumption III. In order to construct the best possible molecular framework models for the polynucleotide–diamine complex, the backbone chain of the diammonium salts I–V is assumed to be fully stretched to allow for maximum electrostatic interaction with the helix and to maintain the best possible staggered conformation. Moreover, the distance between the negatively charged oxygen atom of the phosphate group to the positively charged nitrogen atom of the diammonium salts is assumed to be approximately 2.8–3.0 Å, which is what might be expected for normal H bonding and electrostatic interaction of this type.[22]

Construction of Models for the Nucleic Acid–Diammonium Salt Complex

Examination of the double-stranded Watson–Crick helical structure reveals that the positions of adjacent oxygen atoms are relatively fixed and rigid with respect to one another. Similarly, the positions of adjacent oxygen atoms with respect to each other in a single-stranded 100% stacked polynucleotide are also fixed and rigid. However, at the melting temperature, T_m, of the helix–coil transition of a double-stranded helical structure (in the cases to be reported T_m is greater than 50°), the relative positions of the adjacent oxygen atoms in the helical structure remain relatively fixed whereas they may change drastically for the random coils. Since it is well known that the single-chain polynucleotides at ambient temperatures and neutral pH exist in an equilibrium between the stacked single-stranded helical conformation and the unstacked random coil,[23-34] it is reasonable to expect that the relative distance between adjacent anions will vary significantly. For example, in the unstacked random coil a number of rotations in the molecular structure between adjacent phosphate anions are possible—two carbon–oxygen bond rotations, two oxygen–phosphorus bond rotations, one carbon–carbon bond, and one carbon–nitrogen bond rotation.

Since the binding of the diammonium salts I–V to the random coils would necessitate the freezing out of some of these rotations it is reasonable to expect the binding of the salts I–V to the random coils to be substantially lower than to the helical structure. Evidence to this effect has already been obtained. For example, it has been found by direct binding studies that the affinity constant of the salt I, $N^+H_3CHRCO\text{-}NHCH_2CH_2N^+Me_2H\cdot 2Br^-$, where R = H, for the double-stranded poly I–poly C helix is five times that for the single-stranded poly C at 30.0° and pH 6.30.[35] The difference increases to approximately 20-fold at 50.0° and pH 6.30. Similar results have also been obtained for the diamino acid II, $N^+H_3C(CO_2^-)H\text{-}(CH_2)_nN^+H_3\cdot Cl^-$, where $n = 2$.[36] It is therefore very important to recognize that the stabilization of the helical structure by the diammonium salts I–V as measured by T_m of the helix–coil transition depends to a large extent on the interaction of the salts with the helical structure rather than with the random coils. Therefore, only the following equilibria at the

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melting temperature of a helical structure need be considered

$$D + \text{helix} \rightleftharpoons D\cdot\text{helix} \rightleftharpoons D + \text{random coils}$$

where D = diammonium salts I–V. It is now necessary to examine in detail the nature of the complex, D·helix, for a number of optically active diammonium salts I–IV, by building molecular framework models. It is the purpose of this paper to show that by examining these models it is possible to come to a generalization which not only fits all the data at hand but also has many exceedingly interesting and predictive implications. Models for the polynucleotide–diammonium salt complexes will be considered separately below in the order of increasing number of carbon atoms between the two positively charged nitrogen atoms.

Models for the Best Possible Conformations between the Salts IV, $N^+H_3CHRCH_2N^+H_3\cdot 2Cl^-$, and Adjacent Negative Charges of a Polynucleotide Helical Structure

The adjacent outer (see above) negatively charged oxygen atoms on a rigid helical structure are approximately 7 Å apart and are separated by seven atoms. Due to the very nature of the substitution on the D-ribose ring of adjacent phosphate groups with respect to each other, *i.e.*, *trans* substitution, the structure of the complex formed between the salts IV and the polynucleotide is observed to be a highly strained 15-membered ring (Figure 1). The ring structures shown in Figure 1 have been constructed where the relative positions of adjacent negative charges are relatively fixed and in accordance with the Watson–Crick helical structure. Moreover, only the best possible complexes are shown in Figure 1 and these were constructed according to the assumptions that have been stated earlier, *i.e.*, maximum distance between adjacent positive charges of the salts IV, a long bond (approximately 2.8–3.0 Å) between the negatively charged oxygen of the phosphate group and the positively charged nitrogen atom of the salts IV. There are two possible conformations for the 15-membered ring, shown in Figures 1A and 1B. On examining the molecular structure of the 15-membered ring shown in Figure 1A it is immediately possible to rule out the substitution of a large R group in positions 2, 3, 4 since it would result in large steric repulsions, *i.e.*, transannular interaction. Similarly, one may rule out the substitution of the R group at positions 1, 2, and 4 for the conformation shown in Figure 1B. Transannular interactions are at a maximum in ten-membered rings[37] and begin to disappear as the size of the ring gets larger. However, part of the 15-membered ring shown in Figure 1 contains a rigid *trans* substitution, *i.e.*, the positions of adjacent phosphate groups with respect to each other on the D-ribose ring. For this reason, transannular steric interaction in these ring systems become important. Therefore, according to these models, the substitution of the R group in positions 1 for the conformer shown in Figure 1A and 3 for the conformer shown in Figure 1B would lead to the two best possible complexes between the helical structure and the salts IV. If these two complexes so

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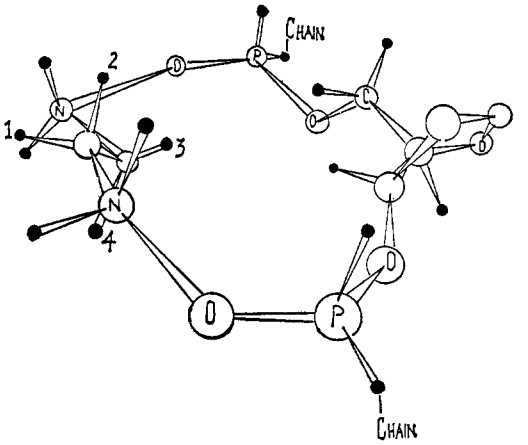


Figure 1A. The complex formed between $N^+H_3C(R)HCH_2N^+H_3\cdot 2Cl^-$ and adjacent negative charges of a rigid helical structure. Conformations 1A-2, IA-3, and 1A-4 are unfavorable. Conformation 1A-1 corresponds to D-$N^+H_3C(CH_3)HCH_2N^+H_3\cdot 2Cl^-$ and L-$N^+H_3C(CO_2^-)HCH_2N^+H_3\cdot Cl^-$ (see text).

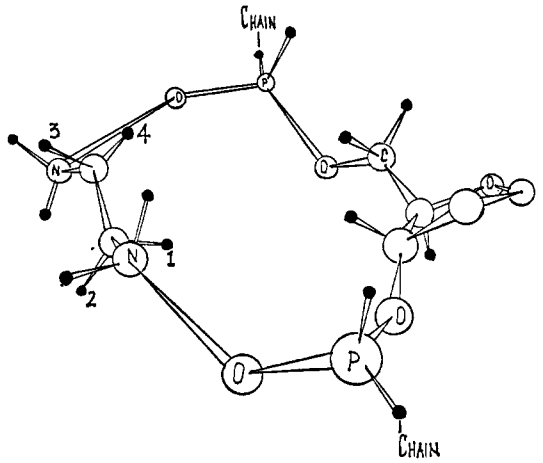


Figure 1B. The complex formed between $N^+H_3C(R)HCH_2N^+H_3\cdot 2Cl^-$ and adjacent negative charges of a rigid helical structure. Conformations 1B-1, 1B-2, and 1B-4 are unfavorable. Conformation 1B-3 corresponds to L-$N^+H_3C(CH_3)HCH_2N^+H_3\cdot 2Cl^-$ and D-$N^+H_3C(CH_3)HCH_2N^+H_3\cdot Cl^-$ (see text).

generated are of equal stability it would be predicted that there should be no difference in the degree of stabilization of the helical structure by the L or the D enantiomers of the salts IV. However, experiments have shown (see below) that there is a difference between the degree of stabilization by the two enantiomers. The results indicate that the enantiomer in which the R substituent is located in the 3 position of Figure 1B is the one which shows the greater degree of stabilization of the helical structure (see below).

The interaction of the salts IV, $N^+H_3CHRCH_2N^+H_3\cdot 2Cl^-$, where R = CH_3 and CO_2^-, with the poly I–poly C helix have been examined, and the results are reported in Table I. It is interesting to note that the data indicate that the L-diaminopropane dihydrochloride interacts more strongly with the helical structure than the corresponding D enantiomer. The situation, on the other hand, is reversed for the 2,3-diaminopropionic acid, *i.e.*, the D enantiomer interacts more strongly with the helix than the corresponding L enantiomer. The relationship of the absolute configuration of these two systems is shown in Chart I. It is obviously clear that the relative position of the CH_3 group in L-diamino-

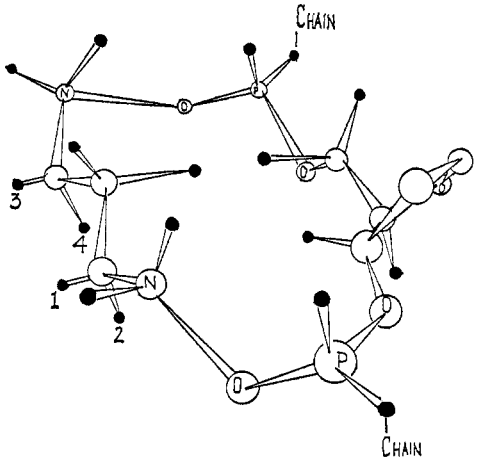


Figure 2. The complex formed between $N^+H_3C(CO_2^-)HCH_2$-$N^+H_3 \cdot Cl^-$ and adjacent negative charges of a rigid helical structure. Conformations 2-2 and 2-4 are unfavorable. Conformations 2-1 and 2-3 correspond to the D- and L-$N^+H_3C(CO_2^-)$-$HCH_2CH_2N^+H_3 \cdot Cl^-$, respectively (see text).

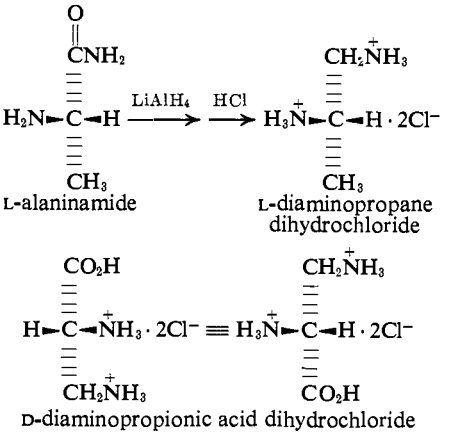
Table I. The Effect of Various Concentrations of IV, $H_3N^+CHRCH_2N^+H_3 \cdot 2Cl^-$, on the T_m of the Helix–Coil Transition of rI–rC in 0.025 *M* Sodium Phosphate Buffer (0.025 *M* in Na^+), pH 6.25[a,b]

Compd	R	Absolute conf of enantiomer (see text)	T_m,[a-c] deg at 0.01 × 10^{-2} *M*	0.05 × 10^{-2} *M*	0.10 × 10^{-2} *M*	2.0 × 10^{-2} *M*
1[d]	CH_3	L	55.8	62.6	65.6	80.4
2[d]	CH_3	D	54.2	61.0	64.4	79.0
3[e]	CO_2H	L	...	...	...	63.0[f]
4[e]	CO_2H	D,L	...	...	...	64.1[f]

[a] Melting curves were measured in 1-ml cuvettes thermostated with a Haake constant-temperature water circulator equipped with a Neslab temperature programmer. A Gilford Model 240 spectrophotometer equipped with automated recording accessories was used, and the temperature of the cell compartment was measured directly by using an iron–constantan thermocouple connected to a Leeds-Northrup Model 8290 potentiometer. [b] The T_m of the rI–rC (Miles Labs) in the absence of the diamino acids II is found to be 50.3 ± 0.4°. [c] T_m curves of the nucleic acid helices in the presence of the salts II were run concurrently for each pair, *i.e.*, L and D or DL mixture. Under these conditions ΔT_m [T_m(L) − T_m(D or DL)] is found to be reproducible to better than ±0.1°. [d] Titration curves of the 1,2-diaminopropane dihydrochloride in 0.025 *M* NaCl showed that at pH 6.20 the molecule contains 1.64 and 1.48 positive charges at 30.0 and 50.0°, respectively. These experiments were performed using the Radiometer automatic titrator. [e] The diaminopropionic acids **3** and **4** at pH 6.20 contain 1.75, 1.47, and 1.30 positive charges per molecule at 30.0, 50.0, and 75.0°, respectively. [f] In the presence of 0.02 *M* sodium bromide.

propane dihydrochloride (**1**) is the same as that of the CO_2H group in D-diaminopropionic dihydrochloride. The conformation shown in Figure 1B where the R group occupies position 3 corresponds to the L-diaminopropane dihydrochloride if R = CH_3 and to the D-diaminopropionic if R = CO_2^-. On the other hand, if the R group occupies position 1 of the conformation shown in Figure 1A, then the diamine would correspond to the D-diaminopropane dihydrochloride if R = CH_3, and to the L-diaminopropionic acid if R = CO_2^-. The results shown in Table I indicate that in two separate systems where R = CH_3 and R = CO_2^-, conformer 1B with the R group occupying the equatorial position is the preferred one since it leads to a greater stabilization

Chart I



(1)

of the rI–rC helical structure. The question remains as to the nature of the difference between conformer 1A-1 (R group in position 1) and conformer 1B-3 (R group in position 3). Examination of the molecular framework models of the 15-membered ring structure (Figures 1A and 1B) shows that both conformers 1A-1 and 1B-3 are sterically strain free and appear to be energetically equivalent.[38] The observable difference between the two conformers 1A-1 and 1B-3 resides in the distance between the R group and the rigid cyclic structure of the D-ribofuranose ring. In conformer 1A-1 the R group is approximately 1.0–1.5 Å closer to the D-ribofuranose ring than in conformer 1B-3. One explanation of the results is that the motion of the R group in conformer 1A-1 is relatively more restricted than in conformer 1B-3 since in the latter conformation the R group is farther removed from the rigid cyclic ring structure. If this simple explanation is accepted then it becomes possible to explain the data obtained not only with this system, but also with all the other optically active systems studied (see below).

Models for the Best Possible Conformations between the Salts II, $N^+H_3CHR(CH)_nN^+H_3 \cdot 2Cl^-$ (where R = CO_2H, and *n* = 2, 3, and 4), and Adjacent Negative Charges of a Polynucleotide Helical Structure

Molecular framework models of the complex formed between the salts II, $N^+H_3CHR(CH_2)_nN^+H_3 \cdot 2Cl^-$ (where R = CO_2H, and *n* = 2, 3, and 4), and adjacent negative charges of a polynucleotide helical structure were constructed. The structures for *n* = 2, 3, and 4 are shown in Figures 2, 3, and 4, respectively, and the results are discussed separately below.

The 2,4-Diaminobutyric Acid Complex. As shown in Figure 2 the complex formed between 2,4-diaminobutyric acid and adjacent negative charges is in fact a rigid 16-membered ring. Again, it is immediately possible to rule out two positions that the carboxyl group may occupy—*i.e.*, positions 2 and 4 of the conformer shown in Figure 2. These positions are unfavorable for the following reasons. (1) Steric transannular interactions caused by an essentially "axial" substitution at positions 2 and 4. (2) Electrostatic repulsion between the carboxylate anion and the phosphate group is expected to be quite large since the two

(38) It is exceedingly difficult to categorically state that both conformers 1A-1 and 1B-3 are energetically equivalent. Examination of molecular framework models indicate that the R group in conformer 1A-1 and 1B-3 is in a chemically similar environment.

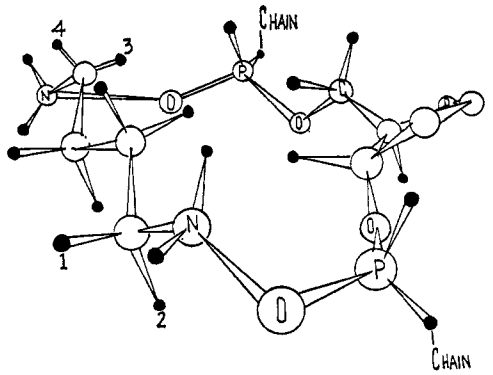


Figure 3A. The complex formed between $N^+H_3C(CO_2^-)HCH_2$-$CH_2CH_2N^+H_3 \cdot Cl^-$ and adjacent negative charges of a rigid helical structure. Conformations 3A-2, 3A-3, and 3A-4 are unfavorable. Conformation 3A-1 corresponds to D-$N^+H_3C(CO_2^-)HCH_2CH_2$-$CH_2N^+H_3 \cdot Cl^-$ (see text).

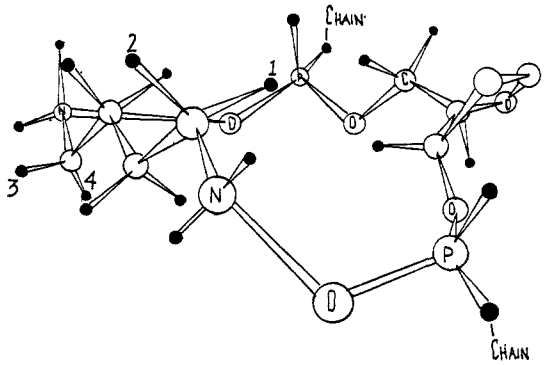


Figure 3B. The complex formed between $N^+H_3C(CO_2^-)HCH_2$-$CH_2CH_2N^+H_3 \cdot Cl^-$ and adjacent negative charges of a rigid helical structure. Conformations 3B-1, 3B-2, and 3B-4 are unfavorable. Conformation 3B-3 corresponds to L-$N^+H_3C(CO_2^-)HCH_2CH_2$-$CH_2N^+H_3 \cdot Cl^-$ (see text).

groups would be closer by approximately 1.60 Å than they would be if the CO_2^- group occupies positions 1 or 3 (Figure 2). (3) The carboxylate anion would obviously prefer the equatorial positions 1 and 3 rather than 2 and 4 since in the former conformer the anion is more readily solvated by the medium. If the carboxyl group occupies positions 1 or 3 of the conformer shown in Figure 2 it would correspond to the D or L enantiomer of the diaminobutyric acid, respectively. The only difference between the two conformers 2-1 and 2-3 appears to be the distance between the substituent and the rigid cyclic D-ribofuranose ring. The distance between the CO_2^- group and the ring is greater for conformer 2-3 than for conformer 2-1. Again, if one accepts the simple explanation that the motion of the R group is more restricted the closer it is to the ring structure, it is possible to conclude that conformer 2-3 is the favored one. It would predict that L-diaminobutyric acid should interact more strongly with the nucleic acid helix than the corresponding D enantiomer. This is indeed the case, and it has been shown by Gabbay, *et al.*,[1] that the L enantiomer stabilizes the poly I–poly C and poly A–poly U helices to a greater degree than the corresponding DL mixture.

The 2,5-Diaminopentanoic Acid Complex. Figure 3 shows the two possible conformations for the 17-membered ring formed between 2,5-diaminopentanoic acid (ornithine) and adjacent negative charges of a helical polynucleotide chain. On careful examination

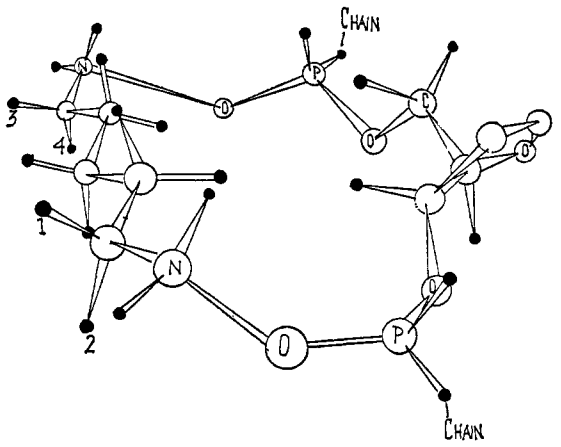


Figure 4. The complex formed between $N^+H_3C(R)HCH_2CH_2$-$CH_2CH_2N^+H_3 \cdot X^-$ (where R = CO_2^- and $CONHCHR'CO_2^-$, *i.e.*, lysylphenylalanine and lysylleucine) and adjacent negative charges of a rigid helical structure. Conformations 4-2 and 4-4 are unfavorable. Conformations 4-1 and 4-3 correspond to the D and L enantiomers, respectively (see text).

of the molecular framework of these conformers it was possible to rule out the substitution of the CO_2^- group at positions 2, 3, and 4 for conformer 3A, and positions 1, 2, and 4 for conformer 3B, *i.e.*, conformers 3A-2, 3A-3, 3A-4, 3B-1, 3B-2, and 3B-4. The two best possible conformers that still remain are those that would result from the substitution of the CO_2^- group at position 1 of conformer 3A (corresponds to the D enantiomer) and at position 3 of conformer 3B (corresponds to the L enantiomer). Again, if one accepts the simple argument that the motion of the R group is more restricted the closer it is to the ring structure it is possible to conclude that conformer 3B-3 is more favored than 3A-1. It would predict that L-ornithine should interact more strongly with the nucleic acid helix than the corresponding D enantiomer. This is indeed the case, and it has been shown by Gabbay, *et al.*,[1] that the L enantiomer stabilizes the poly I–poly C helix to a greater degree than the corresponding DL mixture.

The 2,6-Diaminohexanoic Acid Complex. Figure 4 shows the best possible complex formed between 2,6-diaminohexanoic acid (lysine) and adjacent negative charges of a helical polynucleotide chain. It is immediately possible to rule out two positions that the carboxyl group may occupy—*i.e.*, positions 2 and 4 which correspond to conformers 4-2 and 4-4. The two best possible conformers that remain, 4-1 and 4-3, correspond to the D and L enantiomers of lysine, respectively. According to previously stated argument (see above), conformer 4-3 is more favored than 4-1, and therefore the L-lysine is predicted to interact more strongly with the nucleic acid helix than the corresponding D enantiomer. Gabbay, *et al.*,[1] have in fact shown that the L enantiomer stabilizes the poly I–poly C helix to a greater degree than the corresponding D form.

Models for the Best Possible Conformations between Lysyl Dipeptides III, $N^+H_3C((CH_2)_4N^+H_3)HCONH$-$CHRCO_2^- \cdot Cl^-$, and Adjacent Negative Charges of a Polynucleotide Helical Structure

Gabbay, *et al.*,[1] have shown that L,L-lysyl dipeptides stabilize poly I–poly C, poly A–poly U, and calf thymus

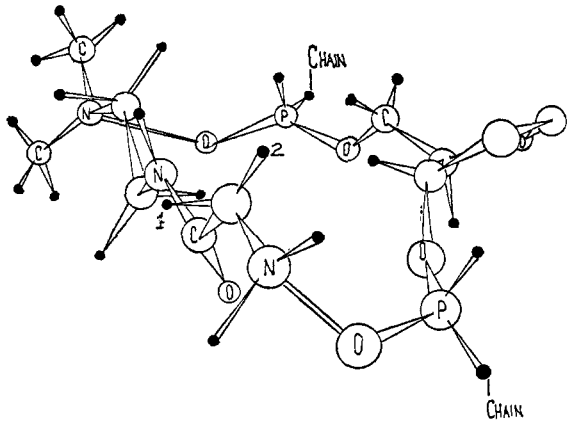


Figure 5A. The complex formed between $N^+H_3CHRCONHCH_2$-$CH_2N^+Me_2H \cdot 2Br^-$ and adjacent negative charges of a rigid helical structure. Conformation 5A-2 is unfavorable. Conformation 5A-1 corresponds to the L enantiomer (see text).

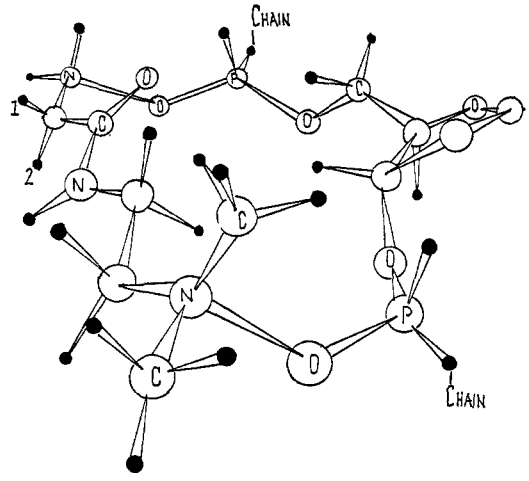


Figure 5B. The complex formed between $N^+H_3CHRCONHCH_2$-$CH_2N^+Me_2H \cdot 2Br^-$ and adjacent negative charges of a rigid helical structure. Conformation 5B-2 is unfavorable. Conformation 5B-1 corresponds to the D enantiomer (see text).

DNA to a greater degree than the corresponding D,D enantiomers. The effect was observed for the two peptides that were reported, *i.e.*, lysylleucine and lysylphenylalanine. Since the complex formed between the lysyl dipeptides and adjacent negative charges of a polynucleotide chain should be identical with that formed between the amino acid, lysine, and the polynucleotide chain (*cf.* Figure 4), it is reasonable to expect that all simple N-terminal L-lysyl peptides interact more strongly than the corresponding D enantiomers. The fact that *two* N-terminal L,L-lysyl dipeptides stabilize *three different helices* to a greater degree than the corresponding D,D,-lysyl dipeptides lends further credibility to the original premise that the side group (in this case $CONHCHRCO_2^-$) prefers position 3 of the conformer shown in Figure 4. It is quite significant to note that the conclusions drawn from the studies on the molecular models of the complexes formed between the salts I–V and the polynucleotide chain *are independent of the nature of the base and the multiplicity of the polynucleotide*, *i.e.*, single-stranded, double-stranded, and/or multistranded helical polynucleotide. In other words, if the L enantiomer of a particular diammonium salt is predicted to interact to a greater degree with a polynucleotide chain than the corresponding D enantiomer, it should do so with all nucleic acid helices irrespective of the nature of the bases and the multiplicity of the helix. The sugar–phosphate backbone chain of all nucleic acids is stereochemically identical (D-ribofuranoside for RNA, and D-deoxyribofuranoside for DNA). Since the binding of the salts I–V occurs to adjacent phosphate anions *on the same strand* the conclusions drawn from the studies of molecular models of the complexes formed with nucleic acids does not depend on the multiplicity or the nature of the bases in the polynucleotide system. The complexes formed between the L and D enantiomers of the diammonium salts I–IV and any polynucleotide chain will have the conformations shown in Figures 1–5. The relative orientation of the side chain R group of the L and D enantiomers of the salts I–IV in the polynucleotide–diamine complex does not depend on the distance between adjacent phosphate anions, *i.e.*, in the range of 6.0–8.0 Å. This conclusion may readily be demonstrated by molecular framework models of the polynucleotide–diamine complex. The validity of these conclusions are attested by the fact that all of the polynucleotide systems studied, *i.e.*, rI–rC, rA–rU, rA–rU_2, and calf thymus DNA, are stabilized to a greater degree by the same enantiomer of a D and L pair. Further testing of these conclusions are in progress.

Models for the Best Possible Conformations between the Amino Acid Amides I, $N^+H_3CHRCONHCH_2CH_2$-$N^+Me_2H \cdot 2Br^-$, and Adjacent Negative Charges of a Polynucleotide Helical Structure

There are two possible conformations for the complex formed between the amides I, $N^+H_3CHRCONHCH_2CH_2N^+Me_2H \cdot 2Br^-$, and the polynucleotide chain as shown in Figure 5, the two conformations being the ones where the dimethylammonium group of the amide is either next to the 5′ end of the ribose ring (conformation 5A) or the 3′ end of the ribose ring (conformation 5B). Again, if one accepts the ideas presented earlier (*i.e.*, the closer the large group is to the rigid cyclic ring structure, the more its motion is restricted), it is then predicted that conformer 5A is more stable than 5B. The substituent R of the amides I, $N^+H_3CHRCONHCH_2CH_2N^+Me_2H \cdot 2Br^-$, may occupy either position 1 or 2 of conformer 5A. Position 2 of conformer 5A is not favorable since the substituent in this position points into the ring causing a transannular steric repulsion. It is therefore concluded from the studies of these models that the best possible conformer is 5A-1 which corresponds to the L-amino acid amides I. Gabbay and Kleinman[2] have indeed shown that the L-amide derivatives, I, of alanine, α-aminobutyric acid, proline, and lysine[39] interact more strongly with nucleic acids than the corresponding D enantiomers.

Summary and Conclusions

It has been demonstrated that it is possible to predict through the use of molecular framework models which of two possible diammonium salt enantiomers should

(39) The amide I derivative of lysine may bind to adjacent negative charges on a polynucleotide chain in two ways, *i.e.*, either as shown for lysine in Figure 4 or as shown for the amides I in Figure 5. In either case, the L enantiomer is predicted to interact with the polynucleotide to a greater degree than the corresponding D enantiomer.

interact more strongly with nucleic acid helices. The construction of these complexes was based on some reasonable assumptions. The credibility of these assumptions is demonstrated by their ability to predict correctly all of the observations thus far recorded. Further testing of these ideas using other types of optically active diammonium salts are in progress.

Experimental Section

Materials. Calf thymus DNA and polyadenylic and polyuridylic acids were obtained from Calbiochem. Polycytidylic and polyinosinic acids were obtained from Miles Labs. L- and D-diaminopropanes were resolved using the procedure outlined by Dwyer, *et al.*[40,41] The specific rotations, $[\alpha]^{35}D$, of the L- and D-diaminopropane dihydrochloride in 1 *N* HCl were found to be −3.82 and +3.48°, respectively. The reported value of $[\alpha]^{18}D$ for the L enantiomer of −3.6° is in agreement.[42] L-Diaminopropionic acid hydrochloride was obtained from Biochemical Research, $[\alpha]D$ +25.6°. DL-Diaminopropionic acid was synthesized from 2,3-dibromopropionic acid hydrobromide according to the method of Greenstein and Winitz.[43] Solutions of the salts IV, N^+H_3-$CHRCH_2N^+H_3$, were prepared in glass-distilled water and carefully adjusted to pH 6.30 by addition of 0.1 *N* sodium hydroxide. In the case of the L-diaminopropionic acid hydrochloride and DL-diaminopropionic acid hydrobromide an equivalent amount of sodium bromide and sodium chloride was added to each, respectively. The final solution in both cases contained the same amount of chloride and bromide ions.

Acknowledgment. This work was supported by the Rutgers Research Council and by Grants GM-13597 and GM-15308 from the U. S. Public Health Service. I am also grateful to Dr. Norman Boggs, III, Mrs. Barbara Gaffney, Miss Roberta Kleinman, Mr. Robert Glaser, and Mr. Robert R. Shimshak for many valuable discussions. Thanks are also due to Miss Betty Hays who drew Figures 1–5 and to the talented and artistic Miss Kleinman for her helpful advice in the drawing of these figures.

(40) F. P. Dwyer, F. L. Garvan, and A. Shulman, *J. Am. Chem. Soc.*, **81**, 290 (1959).
(41) The resolution of the diaminopropanes was done by Mrs. Barbara Gaffney.
(42) S. Schnell and P. Karrer, *Helv. Chim. Acta*, **243**, 2037 (1955).
(43) J. P. Greenstein and M. Winitz, "Chemistry of the Amino Acids," Vol. 3, John Wiley and Sons, Inc., New York, N. Y., 1961.

Communications to the Editor

The Conformation of *cis,cis*-Cyclodeca-3,8-diene-1,6-dione

Sir:

The conformation of *cis,cis*-1,6-cyclodecadienes has heretofore been discussed in terms of conformers of types I and II, which we designate here as chair and boat, respectively. Generally, opinion has favored the chair form on the basis that nonbonded interactions in the boat form between ring substituents and between the two olefinic bonds are relieved in the chair form.[1] However, the possibility that the boat form is favored because of a sufficiently attractive transannular interaction between olefinic bonds has also been presented.[2] The answer to the question as to which form is favored is of interest not only for testing current methods of conformational analysis but also for providing a starting point for understanding the dynamic and chemical behavior of the ring system. Thus, the recently observed transannular addition of the elements of methyl hypobromite to the tetracarbethoxy derivative IIIa to give a *cis*-decalin ring system[3] suggests the presence of a boat conformer at some stage of reaction. At the present time, however, there is no direct experimental evidence on the conformational bias of *cis,cis*-1,6-cyclodecadienes on which to base a reasonable interpretation of such phenomena.[4] In this communication we present preliminary results of an X-ray crystallographic examination of a derivative of this ring system, diketone IIIb,[1a] which establish, at least for this derivative in the crystalline state, the previously anticipated preference for the chair form.

Crystals of *cis,cis*-cyclodeca-3,8-diene-1,6-dione (IIIb) from chloroform are orthorhombic, with $a = 8.36$, $b = 7.44$, $c = 13.94$ Å. The systematic absences are (hkl) when $k + l = 2n + 1$, $(0kl)$ when $k = 2n + 1$, and $(h0l)$ when $h = 2n + 1$. The space group is therefore either $C_{2v}{}^{17}$-Aba or $D_{2h}{}^{18}$-Abam, and the number of molecules in the unit cell must be a multiple of four. With four molecules per cell, the calculated density is 1.26 g cm^{-3}; larger multiples of this are highly unlikely. Four molecules per cell in space group Aba require a minimum molecular symmetry of C_2-2, and in Abam of C_{2h}-2/m. If the conformation is IIb the molecular twofold axis must be perpendicular to the long axis of the molecule; this twofold axis, in either space group, must be parallel to *c*. Examination of molecular models quickly shows that it is impossible to accommodate the molecules in the unit cell in this way, regardless of how they are rotated about their twofold axes relative to *a* and *b*. If, on the other hand, the conformation is Ib, the molecular twofold axis must

(1) (a) C. A. Grob and P. W. Schiess, *Helv. Chim. Acta*, **43**, 1546 (1960); (b) *ibid.*, 47, 558 (1964); (c) C. Moussebois and J. Dale, *J. Chem. Soc., C*, 264 (1966).
(2) R. M. Gipson, Ph.D. Thesis, University of Texas, 1965.
(3) R. M. Gipson, H. W. Guin, S. H. Simonsen, C. G. Skinner, and W. Shive, *J. Am. Chem. Soc.*, **88**, 5366 (1966).
(4) Although equilibration studies have shown that *cis,cis*-1,6-cyclodecadiene is markedly more thermodynamically stable than other cyclodecadiene isomers,[1c,5] the results do not permit an experimental distinction between conformational isomers.
(5) (a) A. J. Hubert, *J. Chem. Soc., C*, 2149 (1967); (b) A. J. Hubert and J. Dale, *ibid.*, 188 (1968); (c) for a review, see J. Dale, *Angew. Chem. Intern. Ed. Engl.*, **5**, 1000 (1966).