Biochemistry

 Volume 28, Number 12 June 13, 1989

Accelerated Publications

Effects of the O2′ Hydroxyl Group on Z-DNA Conformation: Structure of Z-RNA and (araC)-[Z-DNA]†

Mai-kun Teng,‡ Yen-Chywan Liaw,‡ Gijs A. van der Marel,§ Jacques H. van Boom,§ and Andrew H.-J. Wang*,‡

Department of Physiology and Biophysics, University of Illinois at Urbana-Champaign, Urbana, Illinois 61801, and Gorlaeus Laboratories, Leiden State University, 2300RA, Leiden, The Netherlands

Received March 23, 1989; Revised Manuscript Received April 19, 1989

ABSTRACT: The left-handed Z structures of two hexamers [d(CG)r(CG)d(CG) and d(CG)(araC)d(GCG)] containing ribose and arabinose residues have been solved by X-ray diffraction analysis at 1.5-Å resolution. Their conformations closely resemble that of the canonical Z-DNA. The O2′ hydroxyl groups of both rC and araC residues form intramolecular hydrogen bonds with N2 of the 5′ guanine residue and replace the bridging water molecules in the deep groove of Z-DNA, which stabilize the guanine in the syn conformation. The araC residue can be incorporated into the Z structure readily and facilitates B to Z transition, as supported by UV absorption spectroscopic studies. In contrast, in Z-RNA the ribose of the cytidine residue is twisted in order to form the respective hydrogen bond. The potential biological roles of the modified Z-DNA containing anticancer nucleoside araC and of Z-RNA are discussed.

The polymorphism of nucleic acid structures is now a well-established observation. DNA, in particular, is known to adopt various conformations such as the right-handed B- and A-DNA and the left-handed Z-DNA depending on the nucleotide sequence as well as several environmental factors. In addition, many chemical modifications on the DNA molecules can also have varying degrees of effects on their structures and properties, which may result in profound biological consequences. For example, the restriction methylation modification on the N6 of adenine or C5 of cytosine prevents the host DNA from being cleaved by restriction enzymes. In those examples, it has been shown that the methylation on the base does not cause any substantial conformational changes on DNA (Frederick et al., 1988). On the other hand, the same methylation on cytosine C5 in poly(dC-dG) dramatically shifts the B–Z equilibrium toward the formation of Z-DNA (Rich et al., 1984).

There are other types of modifications involving the nucleic acid backbone that have profound effects on the stability and reactivity of the nucleic acids. For example, the replacement of the phosphodiester linkage with a thiophosphate or methyl phosphate group alters the nuclease susceptibility of the nucleic acids (Smith et al., 1986; Matsukura et al., 1987). Another integral part of the backbone is the sugar moiety. It has been known for some time that the substitution of the deoxyribose ring with the ribose or 2′-fluoro-2′-deoxyribose ring will drive the nucleic acid molecule into the RNA-11 conformation from the B-DNA structure (Saenger, 1984; Gushlbauer & Jankowski, 1980). The introduction of the 2′-hydroxyl group in the ribose significantly increases the rigidity of the sugar conformation by holding it in the C3′-endo conformation. This may reflect the fact that the RNA double helix is less flexible than the B-DNA structure due to the rigidity of the ribose ring. Furthermore, RNA has a more dominant effect over the DNA counterpart on the helical conformation of RNA–DNA hybrid molecules by keeping them in the A-DNA conformation (Wang et al., 1982).

However, the effect of the 2′-hydroxyl on the Z-DNA structure is less clear. While it was pointed out previously that there is no steric reason to prevent the poly[r(G-C)] RNA polymer from forming the left-handed Z-RNA structure (Wang et al., 1979), nonetheless poly[r(G-C)] has proven to be more difficult to convert from the A-RNA conformation to Z-RNA, as this transition requires a high concentration (4 M) of $NaClO_4$ and high temperature (60 °C) (Hall et al., 1984; Klump & Jovin, 1987). This hindrance in the A to Z

† This work was supported by a grant from the NSF (DMB 8612286) to A.H.-J.W. Y.-C.L. acknowledges the support from the Institute of Molecular Biology (Taiwan, ROC).

* To whom correspondence should be addressed.
‡ University of Illinois at Urbana–Champaign.
§ Leiden State University.

0006-2960/89/0428-4923$01.50/0

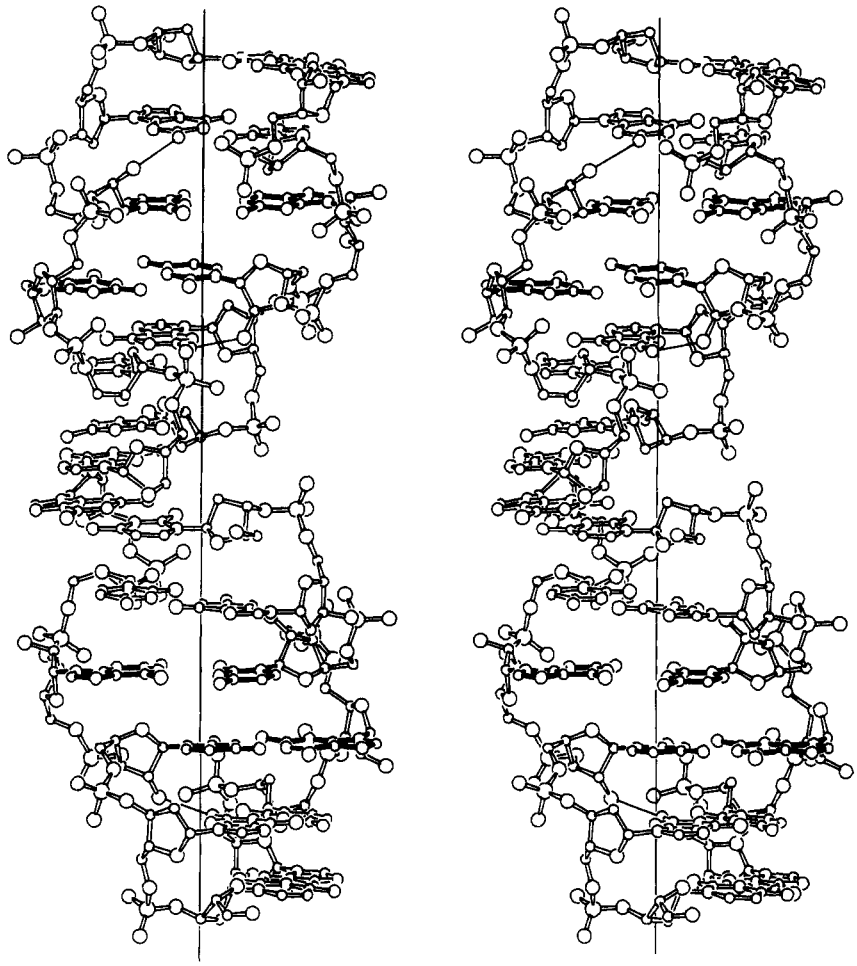

FIGURE 1: Stereoscopic skeletal drawing of the d(CG)r(CG)d(CG) Z-DNA structure. Two hexamers are stacked end over end along the helix axis (also the *c* axis) and tipped slightly toward the reader to illustrate the hydrogen bond (thin line) between the O2′ hydroxyl group and the N2 amino group of guanine in the GpC steps. The upper hexamer has the deep minor groove facing the reader, while the lower hexamer has the prominent surface facing toward the reader. The two ribonucleotide base pairs are shaded with dark bonds.

transition of the RNA molecule may again be related to the tendency of the ribose ring to stay in the C3′-endo conformation. In the Z structure, the pyrimidine residue has a C2′-endo conformation. It would require additional energy to maintain the ribose ring in the latter conformation. We also extend the same question to ask what would be the effect of the Z-DNA with modifications on the deoxyribose by replacing it with an arabinose derivative, an anticancer drug arabinosylcytosine. In this paper, we describe the molecular structure in the Z conformation of two DNA hexamer nucleotides containing either the ribose or arabinose sugar. The O2′ hydroxyl group of cytosine nucleotide units is found to form a hydrogen bond to the NH_2 amino group of guanine in the GpC step, which stabilizes the guanine in the syn glycosyl conformation.

Materials and Methods

Several hexamer nucleotides containing ribose nucleotides and arabinosylcytosines including d(CG)r(CG)d(CG), r(CGC)d(GCG), d(CG)(araC)d(GCG), and (araC-dG)$_3$ were synthesized by the hydroxybenzotriazole phosphotriester method described previously (de Vroom et al., 1988).

The hexamer d(CG)r(CG)d(CG) was crystallized from a solution containing 1 mM hybrid hexamer (single-strand concentration), 50 mM sodium cacodylate buffer at pH 7.0, 2.5 mM magnesium chloride, 2.0 mM spermine tetrachloride, 60 mM sodium chloride, and 1.5% 2-methyl-2,4-pentanediol (2-MPD), equilibrated against 10% 2-MPD by the vapor diffusion technique. Under these conditions, crystals formed as clusters in 7–10 days, and some fragments were large enough for data collection. These crystals had the morphology of small parallelepipeds. A crystal having a size of 0.1 × 0.2 × 0.3 mm was mounted in the thin-walled capillary and sealed with a droplet of the crystallization mother liquor for data collection. The crystal is in the orthorhombic space group $P2_12_12_1$ with unit cell dimensions of a = 18.25 Å, b = 30.93 Å, and c = 43.05 Å. The diffraction data were collected on a Rigaku AFC5R rotating-anode diffractometer with an ω-scan mode at 20 °C to 1.5-Å resolution with Cu Kα radiation (1.5401 Å with graphite monochromater) at the power of 50 kV and 180 mA. A total of 1796 unique reflections were considered to be observable at a 2.0 $\sigma(F)$ level above background, and they were used in the refinement. The crystals of the araC-containing hexamer were obtained under similar conditions, except no sodium chloride was added. In this case, well-formed pseudohexagonal plates appeared in a few days. A crystal of 0.2 × 0.3 × 0.4 mm in size was used for data collection on the same diffractometer to 1.5-Å resolution at 20 °C. A total of 2720 reflections were measured above the 2.0 $\sigma(F)$ level and used in the refinement. The crystal belongs to the same space group as the ribose-containing hexamer crystal with essentially the same unit cell dimensions: a = 18.57 Å, b = 30.68 Å, and c = 42.85 Å. Lp, empirical absorption, and decay corrections were applied for both data sets.

While both crystal forms have the same space group and similar unit cell dimensions as the original d(CG)$_3$ Z-DNA

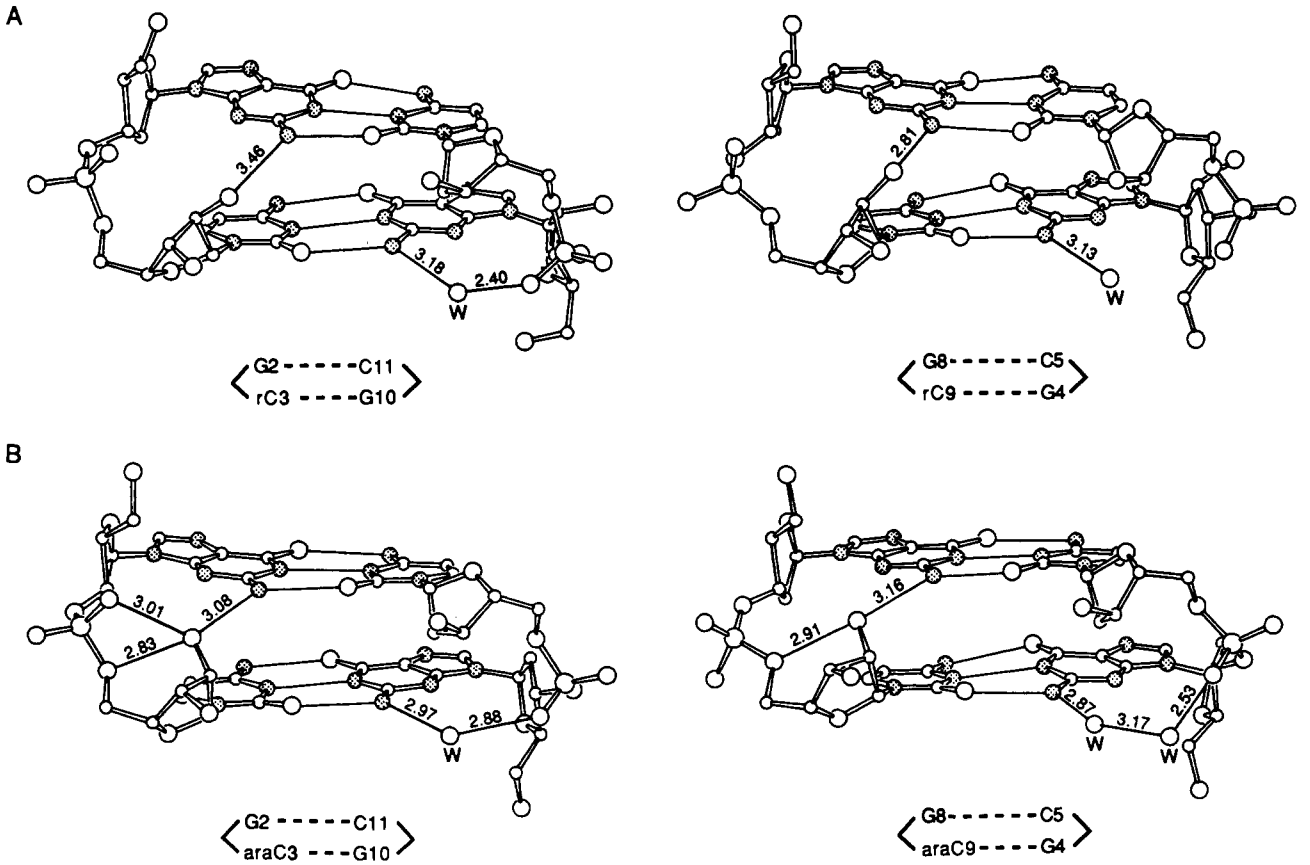

FIGURE 2: Detailed skeletal diagrams of the four GpC steps in the two hexamer crystals. The top panel (A) shows the two dGprC steps of d(CG)r(CG)d(CG) in which the hydroxyls of rC3 and rC9 form hydrogen bonds to the N2 of G2 and G8, respectively. The bottom panel (B) shows the corresponding steps in the d(CG)(araC)d(GCG) hexamer with similar but more extensive hydrogen bonds. Those O2′ hydroxyls fulfill the role of the bridging water molecules, some of which are shown in the diagrams. The hydrogen-bond distances are in angstroms.

crystals (Wang et al., 1979), there are significant differences in their diffraction intensity patterns. Furthermore, the new crystal form has a significantly shorter c axis (~43 Å vs 44.8 Å). Upon close examination, this new crystal form appears to be isomorphous to the $d(CG)_3$ crystals crystallized in the presence of strontium ion or a high concentration of urea (unpublished results). All three crystal structures have been solved by the molecular replacement method using the program ULTIMA (Rabinovich & Shakked, 1984), and they are shown to be of Z-DNA structure. The structures of both d(CG)r(CG)d(CG) and d(CG)(araC)d(GCG) crystal forms were independently refined by using the Konnert–Hendrickson constrained refinement procedure (Hendrickson & Konnert, 1979; Westhof et al., 1985). Solvent molecules located from difference Fourier maps were gradually included in the refinements. For the ribo-Z hexamer structure, the final R factor was 20.4% with one duplex molecule, one spermine, and 68 water molecules per asymmetric unit. For the araC-Z hexamer structure, the final R factor was 16.7% with one duplex molecule, one spermine, and 77 water molecules per asymmetric unit. The two structures have final root mean square deviations of bond lengths from the ideal value of 0.037 and 0.033 Å, respectively. The full results of the analysis will be published elsewhere, and the final atomic coordinates of the structures will be deposited in the Brookhaven Protein Data Bank.

In addition to those two hexamers, several other ribose- and araC-containing hexamers with alternating pyrimidine–purine sequences that have the potential of forming Z structures were also tested for crystallization. Only two of them, r(CGC)d(GCG) and $(araC\text{-}dG)_3$, formed suitable crystals for diffraction work. However, both have crystallized in the hexagonal lattice with the space group $P6_5$ and have unit cell dimensions of $a = b = 17.9$ Å and $c = 43.3$ Å. This crystal form is closely related to other Z-DNA lattices, but it has a disordered arrangement of the hexamer duplexes stacked along the 6_5 screw axis, which makes it impossible to have an accurate structure determination. Consequently, these crystals were not investigated further.

Results and Discussion

Molecular Conformation. The overall conformations of these two modified hexamers are very similar to the canonical left-handed Z-DNA structure observed in the d(CGCGCG) crystal, as can be seen in Figure 1 where only the RNA-containing hexamer is shown. The structure of the araC-containing hexamer is very similar (data not shown). In Figure 1, the alternation of anti–syn conformation in the glycosyl torsion angles is associated with the alternating pyrimidine–purine sequence of the molecule. The resulting zigzag pattern of the sugar–phosphate backbone is evident. The introduction of either ribose or arabinose residues into the molecule does not grossly change any of the torsion angles or the sugar puckers. The sugar puckers of the two rG residues have pseudorotation angles of 46° and 48°, values typically associated with C3′-endo conformation, in comparison to the average value of 30° found in dG residues of the spermine form of d(CGCGCG) (Wang et al., 1979). Similarly, the two rC residues adopt the C2′-endo conformation with pseudorotation angles of 141° and 166°, in comparison to the average 153° of $d(CG)_3$.

One of the surprising observations in both structures is the hydrogen bonds between the O2′ hydroxyl group of ribose (and arabinose) and the NH_2 of guanine in the GpC steps, which

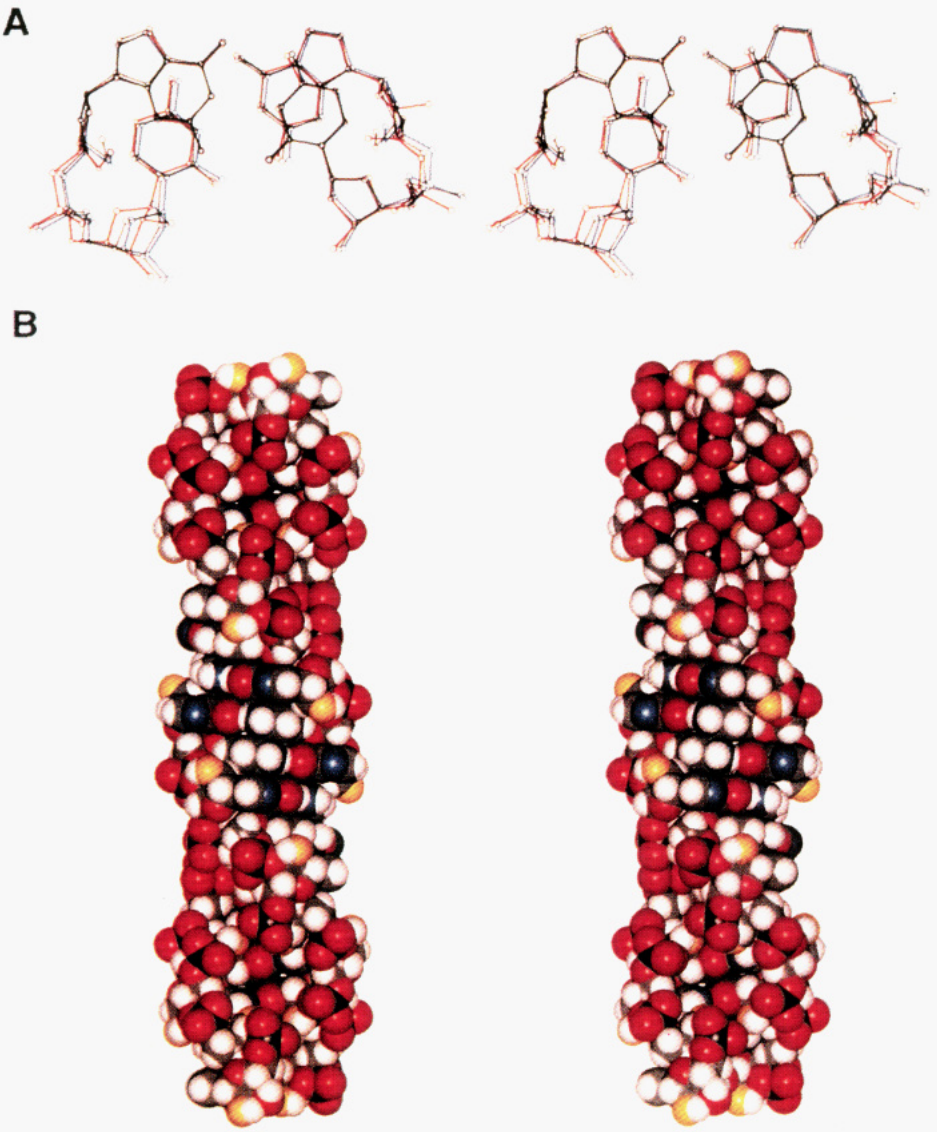

FIGURE 3: (A) Composite diagram showing the stacking interaction and the intramolecular hydrogen bond of the same G2pC3 step (left strand) in three different Z-DNA crystals: d(CGCGCG) form II in $P2_12_12_1$ space group (blue), d(CG)(araC)d(GCG) (brown), and d(CG)r(CG)d(CG) (red). Notice there is little conformational change in going from blue to brown, but substantial twisting rotational movement of the ribose ring (red) is needed to form the hydrogen bond. The uneven stacking pattern of the two complementary strands (G2pC3 vs G10pC11) also occurs in the other GpC step (i.e., G8pC9–G4pC5), both of which are due to crystal packing interactions and not to the intramolecular hydrogen bonds. This is evident from the form II d(CGCGCG) structure, which has an almost identical stacking pattern (blue molecule in this figure) without such hydrogen bonds. (B) Stereoscopic van der Waals diagram of the idealized Z-RNA polymer. Notice that the hydroxyl groups (in yellow) from the guanosine residues are exposed prominently on the surface of the helix, while those from the cytidine residues are well buried in the deep groove of the helix.

are shown as thin lines in Figure 1. This is better illustrated in Figure 2. In Figure 2A (top panel), the two independent dGprC steps are viewed from a similar orientation. It can be seen that the O2′ of ribose is in close proximity to the N2 of guanine, with distances of 3.46 and 2.81 Å, respectively. Although the former distance is slightly longer than a normal hydrogen bond, no other atom or solvent molecule is found between the two groups (O2′ and N2). Therefore, it may be considered a weak hydrogen bond. The latter distance is quite normal for the NH–O hydrogen bond. It is likely that in solution there is a dynamic fluctuation such that those intramolecular hydrogen bonds can vary between 2.8 and 3.5 Å. Note also that the O2′ atom occupies almost the same position as the bridging water moleucle (between N2 and O1P) that stabilizes the G10 in the syn conformation. It seems that the O2′ group fulfills the role of the bridging water in Z structure. Notice that there are significant propeller twists (~6°) in the base pairs that might be due to these hydrogen bonds.

In the (araC)-[Z-DNA] hexamer structure, the two O2′ hydroxyl groups form similar hydrogen bonds, as can be seen in Figure 2B. In fact, the position of those O2′ atoms fits perfectly to form an array of intramolecular hydrogen bonds. For the araC3 residue, its O2′ receives a hydrogen bond (3.08 Å) from N2 of G2 and it forms bifurcated hydrogen bonds to its own O5′ and O1P. The single bridging water associated with G10 has very similar H-bond distances to N2 and O1P (2.97 and 2.88 Å). For the araC9 residue, the N2–O2′ and O5′–O2′ hydrogen bonds are 3.16 and 2.91 Å, respectively. In this case, O2′ does not form a direct H-bond to O1P of the phosphate as it is in the Z_{II} conformation pointing away from the groove (Wang et al., 1981). The bridging water structure of the complementary G4 residue is of the second type using two water molecules, which are also frequently observed in other Z-DNA structures.

O2′ of rC/araC Stabilizes syn-Guanine in the Z Helix. The present structures address one of the interesting questions

Table I: Conformational Parameters[a] of Left-Handed Z Structures

		α	β	γ	δ	ϵ	ζ	χ	ν_{max}	P	sugar pucker
Z-DNA[b]	G	64	−176	178	97	−117	−69	60	30	32	C3′-endo
	C	−145	−124	51	145	−96	76	−152	36	152	C2′-endo
Z-RNA[c]	G	49	−178	−168	102	−108	−74	67	18	41	C4′-exo
	C	−148	−138	56	148	−92	72	−157	55	156	C2′-endo
(araC)-[Z-DNA][c]	G	36	−150	−165	90	−100	−87	62	28	36	C3′-endo
	C	−159	−136	75	135	−88	91	−158	41	151	C2′-endo

[a] Torsion angles are defined as P—O5′—C5′—C4′—C3′—O3′—P (α β γ δ ϵ ζ) and χ for the glycosyl angle. ν_{max} and P are the pseudorotation amplitude and angle, respectively (Saenger, 1984). [b] Average values based on the 1-Å atomic resolution structure of the spermine and magnesium forms of d-(CGCGCG) (Gessner et al., 1989). [c] Idealized polymer structure.

relating to the stability of the Z-DNA structure when the deoxyribose ring is replaced with other types of sugars. In the case of Z-RNA, the O2′ hydroxyl groups of guanosine residues are located on the concave surface of the helix, readily accessible to the solvent molecules. On the other hand, the O2′ of the cytidine residues forms a hydrogen bond to the NH_2 of guanine in the GpC step and is buried in the deep groove of the helix. The formation of this hydrogen bond requires a twisting rotation of the ribose ring so that the O2′ can be positioned to receive the hydrogen bond from the guanine amino group (see Figure 3A). If the O2′ oxygen atom is placed on a regular Z-DNA structure, its position is midway between two adjacent base pairs (with the vertical distance of O2′ to the plane of guanine of 1.9 Å and the N2–H···O2′ angle of 94°), not favorable to receive a hydrogen bond despite the fact that its distance to N2 of G is only 3.4 Å. Therefore, even though the twisting produced by changes in the torsion angles is distributed evenly over several bonds, some energy cost is incurred. Some of this energy cost is recovered by forming the hydrogen bond and removing the small empty space between O2′ and N2. Further, the guanine bases are stabilized in the syn conformation by those O2′ hydroxyl groups that replace the role of the bridging water spine running along the helix axis in the deep groove. The entropy gain due to the liberation of those bridging water molecules should also contribute to the stability of Z-RNA.

In contrast, araC can be incorporated into a Z-DNA structure with minimum conformational distortion. The O2′ of arabinose occupies a position 2.95 Å from N2 of G with good geometry for a hydrogen bond. This is clearly shown in Figure 3A where the five-membered ring of the arabinose (brown) can be superimposed onto the same five-membered ring of deoxyribose (blue) almost perfectly, without any significant conformational adjustment. This ease of the incorporation of araC into a Z-DNA structure may be associated with the fact that araC-containing hexamers appear to crystallize in Z structure more readily than does the $d(CG)_3$ hexamer. We have addressed this question of the ease with which regular alternating (dC-dG) and (araC-dG) oligomers can form Z structure by measuring their A_{290}/A_{258} absorbance ratio, a quantity for determining the extent of Z content. For poly(dC-dG), this ratio is 0.277 in low salt solution (associated with B-DNA), while it is 0.454 in 4 M NaCl solution (associated with Z-DNA) (Pohl & Jovin, 1972). For $d(CG)_3$, the respective ratios are 0.277 and 0.376. The lower ratio with high salt reflects the incomplete conversion to Z-DNA of the $d(CG)_3$ at 4 M NaCl due to the small size of the hexamer. However, the corresponding values are 0.320 (in low salt) and 0.393 (in high salt) for $(araC\text{-}dG)_3$, indicative of a higher proportion of Z content as compared to that of $d(CG)_3$. The crystal structure of the araC-containing hexamer therefore provides us a satisfactory explanation for this observation.

Z-RNA and (araC)-[Z-DNA] Polymers. We have used the structural information obtained from the present crystallographic analysis of the two hexamers, d(CG)r(CG)d(CG) and d(CG)(araC)d(GCG), to build extended polymers for Z-RNA and (araC)-[Z-DNA] as described below. In both polymer structures, we assume they have similar helical parameters as those of the canonical Z-DNA; i.e., they have a 12-fold left-handed double helix conformation with the alternating anti(C)–syn(G) dinucleotides as repeating units. In each dinucleotide step, GpC or CpG, there is a molecular diad axis between two adjacent Watson–Crick base pairs perpendicular to the helix axis. However, an additional important geometric constraint, the hydrogen bond between the N2 of G and the O2′ of the cytosine sugar in the GpC step, was put in to account for the observations found in the crystals. In the case of Z-RNA, the hydrogen-bond distance was set at 2.98 Å, while for (araC)-[Z-DNA], the distance was 3.00 Å. Attention was paid to the direction of the NH–O2′ hydrogen bond so that angle N2–H–O2′ is within the acceptable range (>120°). After the initial polymer model was constructed by a helix-building program, it was refined with the NUCLSQ program (Hendrickson & Konnert, 1979; Westhof et al., 1985) including those constraints described above. The (araC)-[Z-DNA] model converged smoothly in a few cycles. However, the Z-RNA model took many cycles before it converged. Presumably this is due to the fact that in Z-RNA the ribose ring is required to twist around in order to move the O2′ to a proper position to accept the hydrogen bond from N2 of the 5′-side guanine. The comparison of the conformational parameters of the three Z structures (Z-DNA, Z-RNA, and (araC)-[Z-DNA]) is summarized in Table I.

Figure 3B shows a van der Waals diagram of the idealized model of the Z-RNA molecule, which is similar to the canonical Z-DNA. However, there are some differences between them. It can be seen that the O2′ hydroxyl groups of the guanosine residues are prominently exposed along the two edges of the concave surface ("major groove") of the helix. The closest distance between two O2′ groups along the same chain is 11.0 Å, while that from across the groove is 12.1 Å. It is also interesting to note that the G O2′ is very close to both the C8 atom of the same guanine and the C5 atom of the next cytosine in the 3′ side. Bromination of these two positions on poly[r(G-C)] has been shown to greatly facilitate the A- to Z-RNA transition (Hardin et al., 1987). The different organization of solvent structure near the O2′ atom in both A-RNA and Z-RNA may contribute to the position of the A–Z equilibrium of the bromine-modified polymer.

The O2′ hydroxyl groups of the cytidine residues, on the other hand, have a very different environment. They are tucked inside the deep groove. As mentioned earlier, they serve to replace the role of the bridging waters that are believed to stabilize the syn conformation of guanine residue. One may ask why Z-RNA is difficult to form in comparison to its DNA counterpart, if the O2′ provides the additional intramolecular hydrogen bond. This may be explained by three factors. First, riboC nucleotides in the Z structure are kept in the C2′-endo

(instead of the energetically favored C3′-endo) conformation with high pseudorotation amplitude (ν_m = 54°). Second, small but significant changes in the torsion angles of the backbone are needed to twist the ribose ring such that the O2′–N2(G) hydrogen bond may be formed. Third, some loss of base pair stacking interaction is observed in the Z-RNA polymer model (data not shown). Therefore, the energy gain from the hydrogen-bond formation is not enough to compensate for the total energy losses from those three components.

Biological Implications. Our high-resolution structures of the left-handed Z helix incorporating ribose and arabinose residues provide important information regarding the morphology of the helix, the energetics of the B–Z equilibrium, and its possible biological implications (Rich et al., 1984; Jaworski et al., 1987). The conformations of both the Z-RNA and the (araC)-[Z-DNA] structures are similar to that of the canonical Z-DNA. Therefore, proteins that bind Z-DNA are expected to bind both Z-RNA and (araC)-[Z-DNA], although their affinity may be altered. This is borne out from the results of the binding studies of anti-Z-DNA antibody to the Z-form of Br-poly[r(G-C)], which showed that they have reduced affinity (Hardin et al., 1987). This can be rationalized by the observation that the O2′ hydroxyl group of the rG residues in Z-RNA is prominently exposed on the surface of the helix near the phosphate group. It is likely that there will be steric hindrance between the G-O2′ hydroxyl group and the antibody molecule. Conversely, there may exist a group of proteins that can bind to Z-RNA more specifically, due to the interactions between the G-O2′ and the protein.

Arabinosylcytosine is an effective agent in the treatment of human acute myelogenous leukemia and has been shown to incorporate into DNA, but not RNA (Kufe & Spriggs, 1985). This may be explained by examining the steric consequences of the insertion of araC into B-DNA and RNA-11 double helices. In the former case, no serious hindrance occurs between the O2′ hydroxyl group and other atoms in DNA. In contrast, when an araC is put into the RNA double helix, many severe close contacts between O2′ and other atoms (e.g., C6 of pyrimidine or C8 of purine, O4′ and O5′ of the 3′ residue) arise. Therefore, it is possible that araC cannot be inserted into the resultant DNA–RNA hybrid (which has an RNA-11 helix conformation) during transcription due to these steric clashes. An opposite effect is seen in (araC)-[Z-DNA]. The insertion of araC into the alternating dC-dG sequence actually facilitates the B to Z conversion by providing a strong intramolecular O2′–N2 hydrogen bond, thereby stabilizing the guanine in the syn conformation. Furthermore, the O2′ hydroxyl groups of the araC residues are completely buried in the deep groove of the Z-DNA helix, not easily accessible to the outer solvent region. Insofar as the Z-DNA binding proteins are concerned, (araC)-[Z-DNA] has the same binding surface as the native, unmodified Z-DNA molecule. All other known chemical modifications that can facilitate B to Z transition, such as bromination or methylation at the C5 position of cytosine, introduce perturbations on the surface of the Z helix. Therefore, it may be worthwhile to explore the potential of (araC)-[Z-DNA] polymer as a Z-helix probe for a variety of biochemical experiments, since it may be stabilized in the Z form under milder conditions and yet has an identical binding surface for protein recognition.

The structure of the (araC)-[Z-DNA] hexamer increases our repertoire of information concerning DNA oligomers incorporated with anticancer nucleosides (Coll et al., 1989; Wang, 1987). We have recently determined the structure of Z-DNA containing another anticancer drug, 5-fluorodeoxyuridine, in which the function of this drug may be attributed to the unique properties associated with the small size, hydrophobicity, and high electronegativity of the fluorine atom (Coll et al., 1989). Whether the anticancer activity of araC is related to its ability to facilitate the Z-structure formation remains to be answered.

ACKNOWLEDGMENTS

We thank Dr. S. Kahn for providing the facility to produce Figure 3B using his drawing program MacModel. We also thank Dr. H. Robinson for helpful comments.

REFERENCES

Coll, M., Saal, D., Frederick, C. A., Aymami, J., Rich, A., & Wang, A. H.-J. (1989) *Nucleic Acids Res. 17*, 911–923.

de Vroom, E., Roelen, H. C. P. F., Saris, C. P., Budding, T. N. W., van der Marel, G. A., & van Boom, J. H. (1988) *Nucleic Acids Res. 16*, 2987–3003.

Frederick, C. A., Quigley, G. J., van der Marel, G. A., van Boom, J. H., Wang, A. H.-J., & Rich, A. (1988) *J. Biol. Chem. 263*, 17872–17879.

Gessner, R. G., Frederick, C. A., Quigley, G. J., Rich, A., & Wang, A. H.-J. (1989) *J. Biol. Chem.* (in press).

Gushlbauer, W., & Jankowski, K. (1980) *Nucleic Acids Res. 8*, 1421–1425.

Hall, K., Cruz, P., Tinoco, I., Jr., Jovin, T. M., & van de Sande, J. H. (1984) *Nature 31*, 584–586.

Hardin, C. C., Zarling, D. A., Puglisi, J. D., Trulson, M. O., Davis, P. W., & Tinoco, I., Jr. (1987) *Biochemistry 26*, 5191–5199.

Hendrickson, W. A., & Konnert, J. (1979) in *Biomolecular Structure, Conformation, Function and Evolution* (Srinvasan, R., Ed.) pp 43–57, Pergamon, Oxford.

Jaworski, A., Hsieh, W.-T., Blaho, J. A., Larson, J. E., & Wells, R. D. (1987) *Science 238*, 773–777.

Klump, H. H., & Jovin, T. M. (1987) *Biochemistry 26*, 5186–5190.

Kufe, D. W., & Spriggs, D. R. (1985) *Semin. Oncol. 12*, 34–48.

Matsukura, M. K., Shinozuka, K., Zon, G., Mitsuya, H., Reitz, M., Cohen, J., & Broder, S. (1987) *Proc. Natl. Acad. Sci. U.S.A. 84*, 7706–7710.

Pohl, F., & Jovin, T. M. (1972) *J. Mol. Biol. 67*, 375–396.

Rabinovich, D., & Shakked, Z. (1984) *Acta Crystallogr. A40*, 195–200.

Rich, A., Nordheim, A., & Wang, A. H.-J. (1984) *Annu. Rev. Biochem. 53*, 791–846.

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

Smith, C. C., Aurelian, L., Reddy, M., Miller, P. S., & Ts'o, P. O. P. (1986) *Proc. Natl. Acad. Sci. U.S.A. 83*, 2787–2791.

Wang, A. H.-J. (1987) *Mol. Biol. Nucleic Acids 1*, 53–69.

Wang, A. H.-J., Quigley, G. J., Kolpak, F. J., Crawford, J. L., van Boom, J. H., van der Marel, G. A., & Rich, A. (1979) *Nature 282*, 680–686.

Wang, A. H.-J., Quigley, G. J., Kolpak, F. J., van der Marel, G. A., van Boom, J. H., & Rich, A. (1981) *Science 211*, 171–176.

Wang, A. H.-J., Fujii, S., van Boom, J. H., van der Marel, G. A., van Boeckel, S. A. A., & Rich, A. (1982) *Nature 299*, 601–604.

Westhof, E., Dumas, P., & Moras, D. (1985) *J. Mol. Biol. 184*, 119–145.