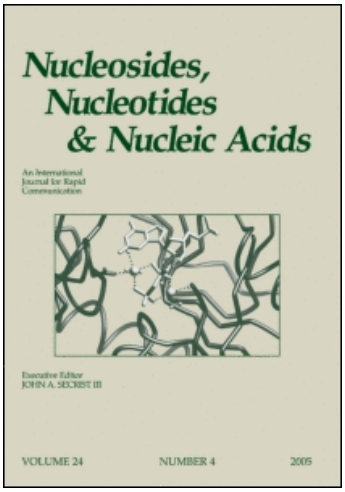

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A Raman Spectroscopic Study on Conformations of DNA Oligomers: A Dominant Effect of an AA:TT Sequence Over Those of AT:AT and TA:TA Sequences on Determining Conformations of DNA Duplexes

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A RAMAN SPECTROSCOPIC STUDY ON CONFORMATIONS OF DNA OLIGOMERS: A DOMINANT EFFECT OF AN AA:TT SEQUENCE OVER THOSE OF AT:AT AND TA:TA SEQUENCES ON DETERMINING CONFORMATIONS OF DNA DUPLEXES#

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ABSTRACT: The conformations of the following ten DNA oligomers in solution have been studied by a Raman spectroscopy; $d(ATAT)_2$, $d(TATA)_2$, $d(AATT)_2$, $d(TTAA)_2$, $d(TTATATAA)_2$, $d(AATTAATTAATT)_2$ (dodecamer **1**), $d(CGCGATATCGCG)_2$, $d(CGCGTATACGCG)_2$, $d(CGCGAATTCGCG)_2$, and $d(CGCGTTAACGCG)_2$ (dodecamer **2-5**). Each oligomer takes on the B form under low salt conditions, although some differences within a B form family are detected. Among four tetramers, only $d(ATAT)_2$ gives an indication of a salt-induced conformational change under high salt conditions. $D(TTATATAA)_2$ which contains the $d(ATAT)_2$ sequence at the center, however, does not give an indication of a salt-induced conformational change, revealing that the inherent character of the $d(ATAT)_2$ sequence is suppressed by the addition of d(AA):d(TT) sequences at both ends. It is in the same context that the conformation of dodecamer **1** is identical to that of poly(dA):poly(dT) but that it is different from that of poly(dA-dT):poly(dA-dT), indicating that the conformation of $d(AATTAATTAATT)_2$ is determined by the overwhelmingly dominant effect of the d(AA):d(TT) sequence over those of d(AT):d(AT) and d(TA):d(TA) sequences. Among dodecamers **2-5**, only $d(CGCGTATACGCG)_2$ gives an indication of the salt-induced B-Z transition. For the other dodecamers, the central tetramers which are composed of A:T base pairs hinder the transition which the $d(CGCG)_2$ sequences at both ends tend to undergo. The exclusive observation of the indication of the transition in d(CGCGTA-

This paper is dedicated to Dr. Morio Ikehara on the occasion of his 70th birthday.

$TACGCG)_2$ seems to be related to the fact that the $d(TATA)_2$ is most unstable among the four tetramers.

INTRODUCTION

Sequence dependent unique conformations of DNA, such as DNA bending, triple and quadruple helixes, and non-standard base pairing, could play a crucial role in expressing the gene information effectively. X-ray crystallography has been a powerful method to study the structure of DNA. It turns out, however, that the structure in the crystalline state is not always the same as that in solution in the case of DNA. Raman spectroscopy is a powerful method to examine the DNA structure in solution.[1]

We have already established by the Raman spectroscopy that poly(dG-dC):poly(dG-dC) undergoes a salt-induced B-Z transition,[2,3] while poly(dG):poly(dC) does a B-A transition.[3,4] DNA polymers only with A:T base pairs, on the other hand, remain within a B form family under different ionic conditions.[5] Particularly poly(dA):poly(dT) undergoes no conformational change on changing the salt conditions. This conformational stiffness is of great interest in relation to DNA bending observed for a $d(A)_n:d(T)_n$ tract. We have also shown that poly(rA):poly(dT) takes on the very peculiar conformation, where a rA strand is A form while a dT strand is B form.[5] This unique conformation is confirmed by the NMR study combined with the Raman spectroscopic study.[6]

We have expanded these Raman spectroscopic studies into DNA oligomers with various base sequences in order to get more detailed information on sequence dependent DNA conformations. We have reported the study of DNA oligomers which are composed of G:C base pairs.[7] Here we report the study on four tetramers, one octamer, and five dodecamers: $d(ATAT)_2$, $d(TATA)_2$, $d(AATT)_2$, $d(TTAA)_2$, $d(TTATATAA)_2$, $d(AATTAATTAATT)_2$ (dodecamer **1**), d(CGCGATATCGCG)2, d(CGCGTATACGCG)2, d(CGCGAATTCGCG)2, and $d(CGCGTTAACGCG)_2$ (dodecamers **2-5**). It turns out that a d(AA):d(TT) sequence has a dominant effect on determining the conformation of the DNA duplex over d(AT):d(AT) and d(TA):d(TA) sequences. In the same context of this finding, the conformation formed by the effect of the d(AA):d(TT) sequence shows resistance against the conformational disturbance by neighbouring sequences in dodecamer **4**.

MATERIALS AND METHODS

The octamer and dodecamer **1** were synthesized with a DNA synthesizer, and purified as described previously.[8] The other oligomers were synthesized and purified as described elsewhere.[7] The DNA oligomers were dissolved in either 0.15 M NaCl or 5.0 M NaCl (pH 7.0). The oligomer concentration was 10-20 % w/w. The sample was sealed in a glass capillary.

Raman spectra were recorded on a Jasco R800 Raman spectrophotometer connected with a MINC DECLAB-11/23 minicomputer at either 25 ^{o}C or -2 ^{o}C. For exciting, the 514.5 nm beam of an NEC GLG 3300 Ar^{+} laser was used. The power was controlled at about 200 mW.

RESULTS AND DISCUSSIONS

The strategy to examine the conformations of the DNA oligomers in solution by a Raman spectroscopy.

It is a big advantage of a Raman spectroscopy that this method can be applied to both solid (crystals and fibers) and solution samples. On the basis of a series of intensive collections of the Raman spectra of the crystals and the fibers, the conformations of which were known by X-ray studies, we have already established the Raman spectrum-conformation correlations of DNA.[1-5,7] When the Raman spectra of DNA in solution are obtained, the conformations can be identified immediately by means of the correlations.

In addition to the conventional terminology on sugar puckering,[9] the following terminology on the main chain conformation will be used in the text[5,7]: a_1 (β = 175^{o}, γ = 45^{o}, δ = 80^{o}), a_2 (β = 210^{o}, γ = 45^{o}, δ = 140^{o}), and b (β = 160^{o}, γ = 45^{o}, δ = 140^{o}). It should be noted that a_2 and b are the main chain conformations within a B form family, while a_1 is that of A form. Although both a_1 and a_2 give a Raman line in the same region, around at 810 cm^{-1}, two conformations can be discriminated definitely by analyzing sugar puckering (Note that the δ value, a measure of sugar puckering, is different very much between a_1 and a_2).

The conformations of the four tetramers under low salt conditions.

FIGs. 1-4 show the Raman spectra of the four tetramers under low salt conditions at either 25 ^{o}C (a) or -2 ^{o}C (b). It is known that the intensity of a 730 cm^{-1} line increases when a DNA duplex melts.[5] It is supposed that the intensity of the 730 cm^{-1} line is related to the

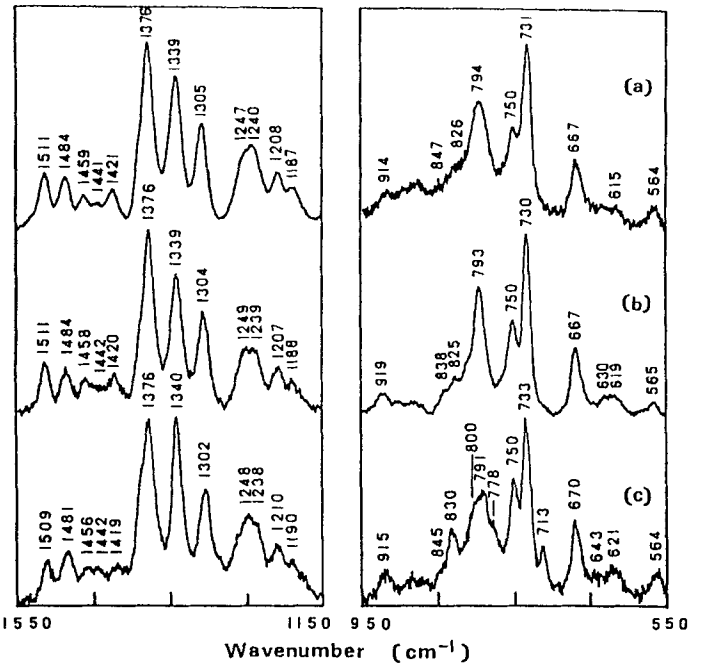

FIG. 1. Raman spectra of $d(ATAT)_2$ in 0.15 M NaCl at 25 ^{o}C (a), in 0.15 M NaCl at -2 ^{o}C (b), and in 5.0 M NaCl at -2 ^{o}C (c).

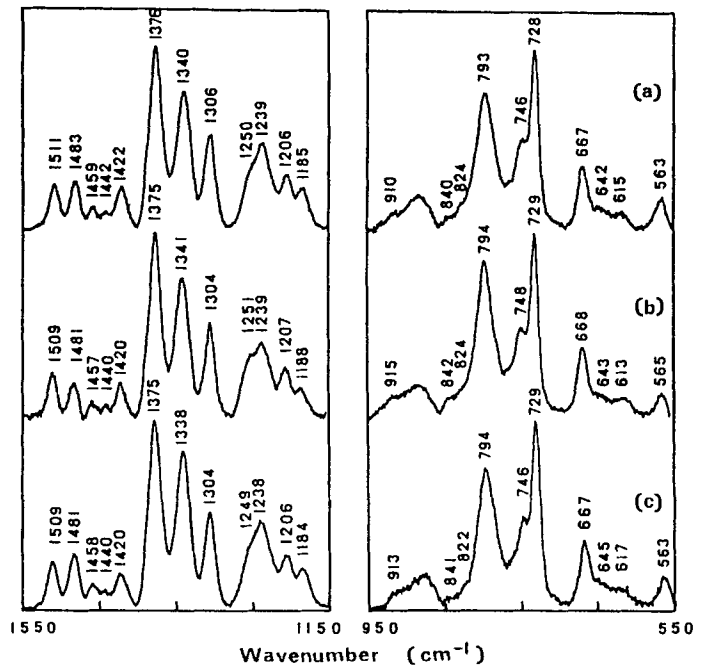

FIG. 2. Raman spectra of $d(TATA)_2$ in 0.15 M NaCl at 25 ^{o}C (a), in 0.15 M NaCl at -2 ^{o}C (b), and in 5.0 M NaCl at -2 ^{o}C (c).

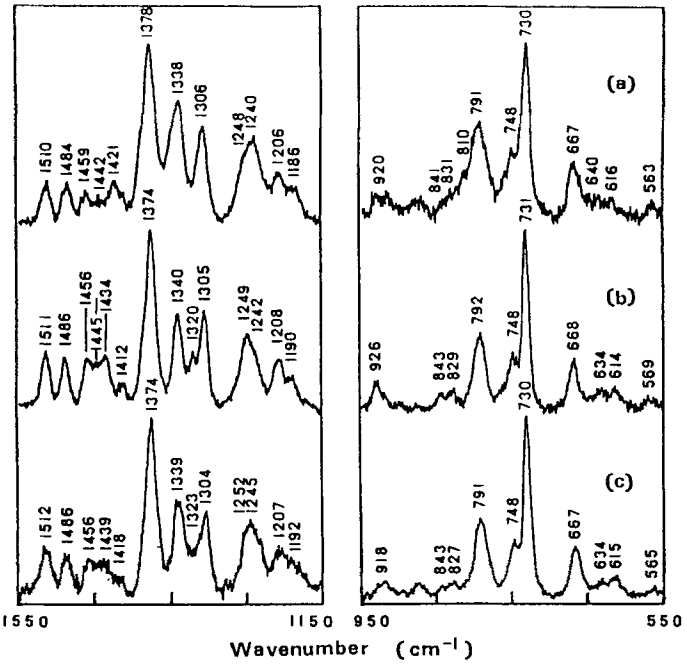

FIG. 3. Raman spectra of $d(AATT)_2$ in 0.15 M NaCl at 25 °C (a), in 0.15 M NaCl at -2 °C (b), and in 5.0 M NaCl at -2 °C (c).

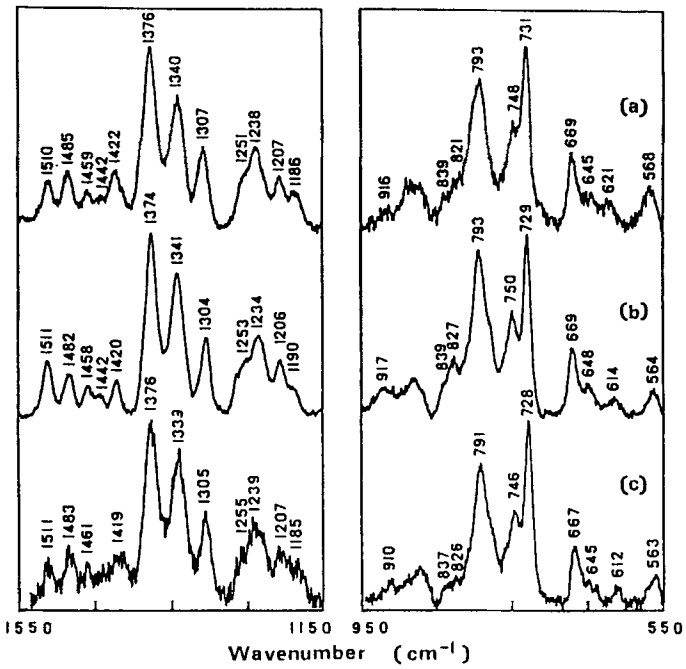

FIG. 4. Raman spectra of $d(TTAA)_2$ in 0.15 M NaCl at 25 °C (a), in 0.15 M NaCl at -2 °C (b), and in 5.0 M NaCl at -2 °C (c).

stacking interaction of bases. The four tetramers give stronger 730 cm^{-1} lines than DNA polymers do (see FIG. 6(b) and (c) for reference), indicating that the tetramers melt to some extent. However, appearance of 830-840 lines reveals that considerable amounts of duplexes are formed at -2 ^{o}C due to high DNA concentrations (20% w/w), because these lines disappear in the case of complete melting of the duplex. In fact, the increase of the intensities of these lines is observed upon lowering the temperature from 25 ^{o}C to -2 ^{o}C. The only exception is $d(TATA)_2$. This does not give evident 810-840 cm-1 lines even at -2 ^{o}C, revealing that the duplex is not formed. $D(TATA)_2$ turns out to be most unstable among the four tetramers.

The three tetramers except for $d(TATA)_2$ take on essentially the same conformations at -2 ^{o}C. The 564-569 (weak), 613-619 (w), 667-669 (strong), and 1374-1376 (s) cm^{-1} lines indicate that the thymidines take on C2'endo-anti. The relative intensities of the lines around at 1240 and 1250 cm^{-1} reveal that the conformations of the adenosines are the mixture of C2'endo-anti and C3'endo-anti. Taking into account the fact that adenosine tends to be C3'endo-anti in a single strand,[5] it is suggested that the adenosines in the duplexes take on C2'endo-anti. 830-840 cm^{-1} lines indicate that the main chain conformation is the b type. All of these results demonstrate that the tetramers in duplexes take on B form.

It should be addressed that some different kinds of features are added to the spectra of $d(AATT)_2$ upon lowering the temperature. The intensities of 731, 1445 and 1456 cm^{-1} lines increase, while that of 1340 cm^{-1} line decreases. New lines appear at 1320 and 1434 cm^{-1}. Shift of a 1412 cm^{-1} line is observed. These could suggest that a small amount of a certain conformation is co-existing at -2 ^{o}C for this short oligomer, although the main conformation is B form.

An indication of a salt-induced conformational change only for $d(ATAT)_2$ among the four tetramers.

FIGs. 1-4 (c) show the spectra of the tetramers under high salt conditions. Apparent changes of spectra upon increasing the salt concentration are observed only for $d(ATAT)_2$ (FIG. 1(c)). An 804 cm^{-1} line, which is indicative of the main chain conformation of a_1 (A form), appears under the high salt conditions. 643, 713 and 778 cm^{-1} lines, all of which are characteristic to A form, also appear under the salt condi-

tions. At the same time, the intensity of an 830 cm^{-1} line, indicative of the main chain conformation of b, increases.

594(w), 621(w), 670(s) and 1376(s) cm^{-1} lines observed for $d(ATAT)_2$ indicate that the thymidines still take on C2'endo-anti under the high salt conditions. Therefore it is concluded that the thymidines take on B form (C2'endo-anti, the main chain conformation of b), while the adenosines assume A form (C3'endo-anti, the main chain conformation of a_1) to a certain extent in the $d(ATAT)_2$ duplex. The increase of the intensity of the 830 cm^{-1} line is supposed to be the result of the suppressed flexibility of the main chain conformation within the b type.

$D(ATAT)_2$ is the only oligomer containing merely A:T base pairs for which the crystal structure is available.[10] It is interesting that the crystal structure exhibits the same alternating conformation as is concluded above. The high salt conditions might be giving a similar environment to that in the crystal.

It is considered that a d(AT):d(AT) sequence tends to undergo the salt-induced conformational change, because among four tetramers the conformational change is observed only for $d(ATAT)_2$, which has the two d(AT):d(AT) sequences in it.

The suppression of the salt-induced conformational change of the $d(ATAT)_2$ sequence by the addition of d(AA):d(TT) sequences to both ends.

FIG. 5 shows the spectra of the octamer, $d(TTATATAA)_2$. The intensity of a 727 cm^{-1} line of the octamer is weaker than those of the tetramers, and comparable to those observed for polymers. This shows that a complete duplex is formed in the case of the octamer which is longer than the tetramers.

The conformation of the octamer under low salt conditions is examined as follows (FIG. 5(b)). 666(s) and 1375(s) cm^{-1} lines indicate that the thymidines take on C2'endo-anti. A 1342 (s) cm^{-1} line and the relative intensities of 1234 and 1252 cm^{-1} lines indicate that the most (c.a. 90 %) of adenosines take on C2'endo-anti while small amounts (c.a. 10 %) of C3'endo-anti adenosines are co-existing. The same thing is observed in the B form of poly(dA):poly(dT).[5] 814(w) and 841(w) cm^{-1} lines indicate that the main chain conformation is the superposition of b and a_2. All data indicate that the $d(TTATATAA)_2$ takes on B form.

The octamer has the $d(ATAT)_2$ sequence at the center. The tetramer $d(ATAT)_2$ undergoes the salt-induced conformational change as mentioned

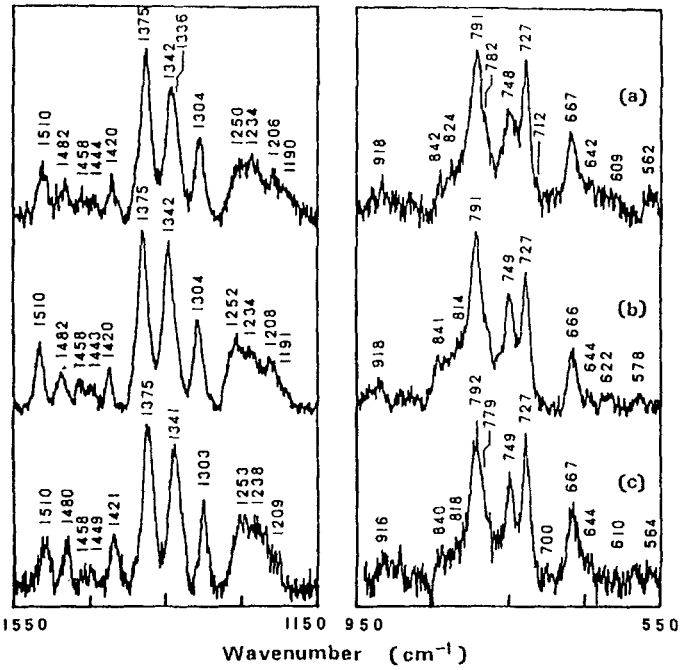

FIG. 5. Raman spectra of $d(TTATATAA)_2$ in 0.15 M NaCl at 25 °C (a), in 0.15 M NaCl at -2 °C (b), and in 5.0 M NaCl at -2 °C (c).

above. FIG. 5(c) shows the spectrum under high salt conditions. A drastic change of the spectra is not observed upon increasing the salt concentration. This means that the inherent character of the $d(ATAT)_2$ portion is suppressed by the addition of the d(AA):d(TT) sequences which do not tend to undergo the salt-induced conformational change, at both ends of the duplex. The addition of the d(AA):d(TT) sequences is having the dominant effect on determining the character of the duplex.

It is considered in the previous section that the d(AT):d(AT) sequence tends to undergo the salt-induced conformational change. Poly(dA-dT):poly(dA-dT) does not undergo the conformational change.[5] This indicates that the d(TA):d(TA) sequence is resistant to the salt-induced conformational change. The inability of the $d(AATT)_2$ and $d(TTAA)_2$ tetramers to undergo the salt-induced conformational change indicates that the d(AA):d(TT) sequence does not tend to undergo this conformational change, either. The octamer has the two d(AT):d(AT) sequences while it has the three d(TA):d(TA) and two d(AA):d(TT) se-

quences. The result on the octamer reveals that the effects of the d(AT):d(AT) sequences are overcome completely by those of d(TA):d(TA) and d(AA):d(TT) sequences in the duplex.

The dominant effect of the d(AA):d(TT) sequence over those of d(AT):d(AT) and d(TA):d(TA) sequences on determining the conformation of the $d(AATTAATTAATT)_2$.

FIG. 6 shows the spectra of the d(AATTAATTAATT)2 (dodecamer **1**) (a), together with those of poly(dA-dT):poly(dA-dT) (b) and poly(dA):poly(dT) (c). Judging from the intensity of the 729 cm^{-1} line, dodecamer **1** is all in a duplex state. The conformation of dodecamer **1** is concluded as follows. 565(w), 612(w), 669(s), 1231(w) and 1375(s) cm^{-1} lines indicate that the thymidines take on C2'endo-anti. 1252(s) and 1342(s) cm^{-1} lines indicate that the all adenosines take on C2'endo-anti. 816(w) and 843 (w) lines indicate that the main chain conformation is the superposition of b and a_2. These results show that the dodecamer **1** takes on the B form.

It should be noted that the relative intensity of 816 and 843 cm^{-1} lines of the dodecamer **1** is identical to that of poly(dA):poly(dT), but that it is different from that of poly(dA-dT):poly(dA-dT). Dodecamer **1** has the six d(AA):d(TT) sequences, which are the basic components of poly(dA):poly(dT). On the other hand, it has the two d(AT):d(AT) and three d(TA):d(TA) sequences, both of which are the basic components of poly(dA-dT):poly(dA-dT). Although the ratio of the numbers of each basic component is 6:5, dodecamer **1** takes on the same conformation as poly(dA):poly(dT) does. The result shows that the effect of the d(AA):d(TT) sequences is overwhelmingly dominant over those of the d(AT):d(AT) and d(TA):d(TA) sequences on determining the conformation of the duplex. This result is consistent with the fact that the salt-induced conformational change of the $d(TTATATAA)_2$ is suppressed by the added d(AA):d(TT) sequences at both ends.

Conformations of dodecamers 2-5 under low salt conditions.

FIGs. 7-10 show the spectra of dodecamers **2-5**. The analyses of the spectra become complex, because these dodecamers contain four different kinds of nucleosides. In spite of the complexity, the conformations of the dodecamers can be examined as follows. The conformations of sugars and bases are the same for the four dodecamers. 681-682 and 1314-1316 cm^{-1} lines together with the shoulder at 1362-1363 cm^{-1} indicate that

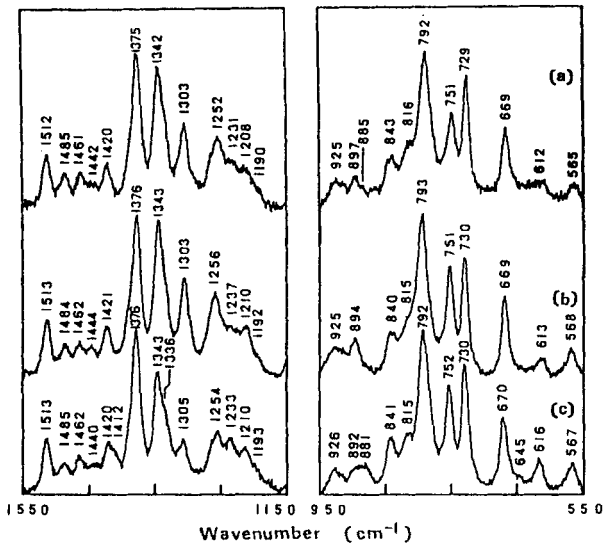

FIG. 6. Raman spectra of $d(AATTAATTAATT)_2$ in 0.15 M NaCl at -2 °C (a), poly(dA-dT):poly(dA-dT) in 0.2 M NaCl at 30 °C (b), and poly(dA):poly(dT) in 0.2 M NaCl at 30 °C (c).

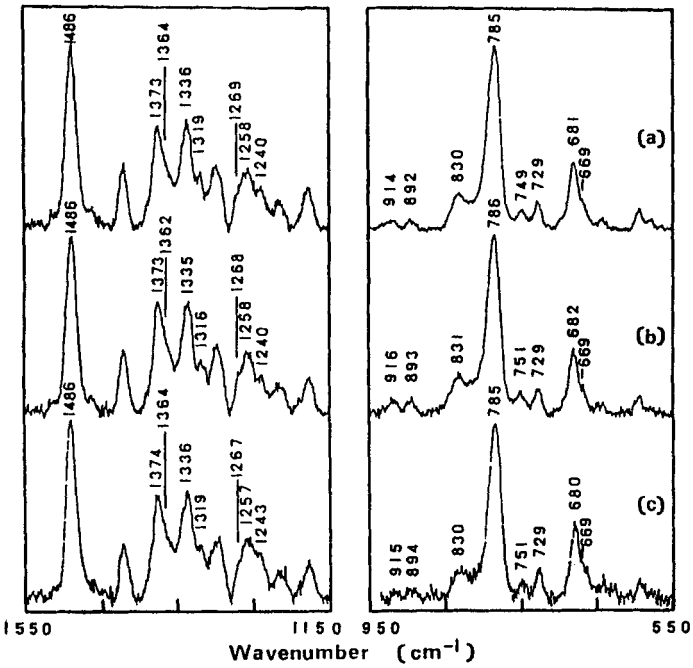

FIG. 7. Raman spectra of $d(CGCGATATCGCG)_2$ in 0.15 M NaCl at 25 °C (a), in 0.15 M NaCl at -2 °C (b), and in 5.0 M NaCl at -2 °C (c).

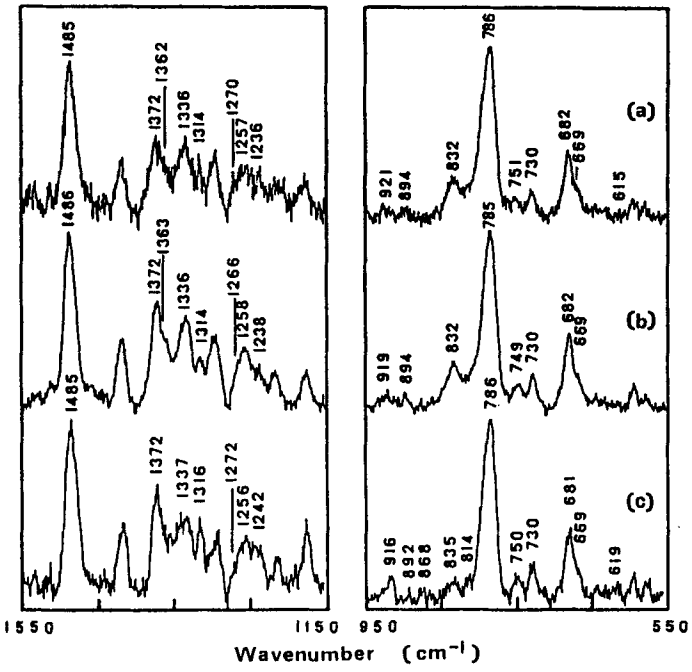

FIG. 8. Raman spectra of d(CGCGTATACGCG)$_2$ in 0.15 M NaCl at 25 °C (a), in 0.15 M NaCl at -2 °C (b), and in 5.0 M NaCl at -2 °C (c).

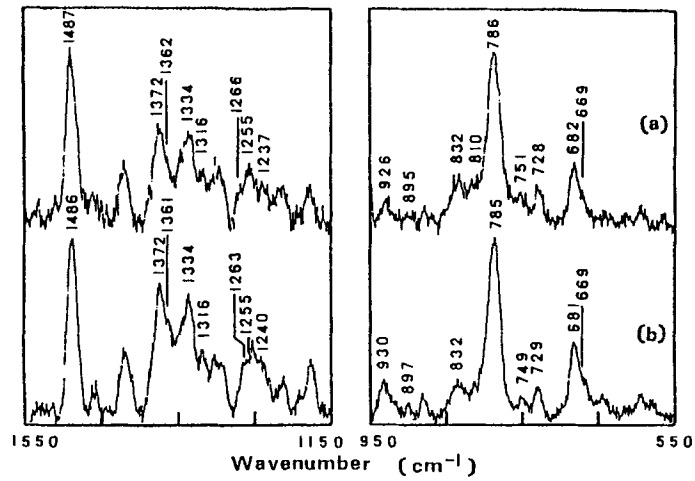

FIG. 9. Raman spectra of d(CGCGAATTCGCG)$_2$ in 0.15 M NaCl at 25 °C (a), in 0.15 M NaCl at -2 °C (b), and in 5.0 M NaCl at -2 °C (c).

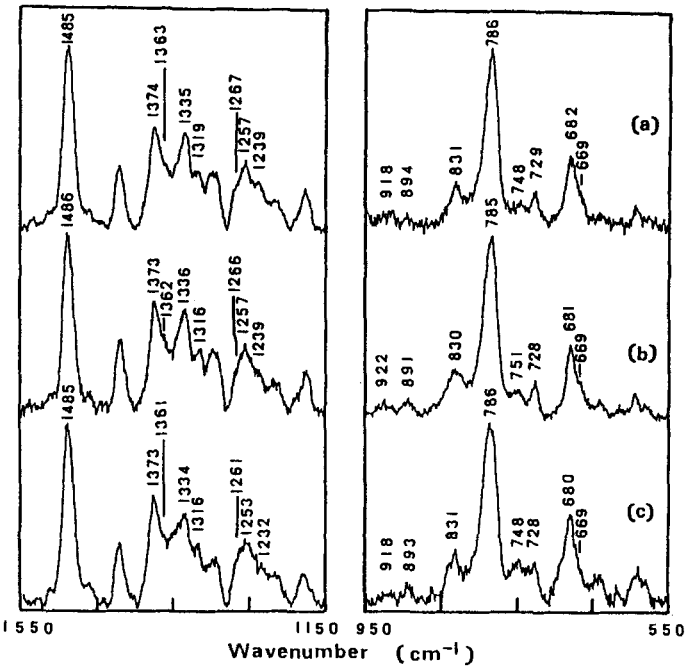

FIG. 10. Raman spectra of d(CGCGTTAACGCG)$_2$ in 0.15 M NaCl at -2 ^{o}C (a) and in 5.0 M NaCl at -2 ^{o}C (b).

the guanosines take on O4'endo-anti. The guanosine of O4'endo-anti has been observed in B form in our previous studies, too.[2-4] A 1372 (s) cm^{-1} line together with the shoulder around at 669 cm^{-1} indicate that the thymidines take on C2'endo-anti. 728-730, 1237-1240 and 1255-1258 cm^{-1} lines together with the shoulders around at 1266-1268 and 1343 cm^{-1} indicate that the both cytidines and thymidines take on C2'endo-anti.

810 and 832 cm^{-1} lines indicate that the main chain conformation of d(CGCGAATTCGCG)$_2$ is the superposition of b and a$_2$. For the other three, 830-832 cm^{-1} lines indicate that the main chain conformation is b. Thus all dodecamers take on B form.

Judging from the intensity of the 810 cm^{-1} line of the d(CGCGAATTCGCG)$_2$, it is supposed that at least the central d(AATT)$_2$ portion takes on the same conformation as poly(dA):poly(dT) does. The resistance of the inherent conformation formed by the effect of the d(AA):d(TT) sequences against the conformational disturbance by neigh-

bouring sequences seems to be correlated with the fact that the effect of the d(AA):d(TT) sequence is overwhelmingly dominant on determining the conformation of the duplex over those of the d(AT):d(AT) and d(TA):d(TA) sequences. The resistance of the conformation of the d(AA):d(TT) sequence against the disturbance might be related to the DNA bending observed for the $d(A)_n:d(T)_n$ tract, because bending is observed at the junction between the d(AA):d(TT) sequence and the other portions in the structure of $d(GGAAATTTCC)_2$ determined by NMR.[12]

An indication of the salt-induced B-Z transition detected only for the $d(CGCGTATACGCG)_2$ among dodecamers 2-5.

It is well known that the $d(CGCG)_2$ sequence undergoes the salt-induced B-Z transition.[11] Four different kinds of tetramers which are composed of A:T base pairs are sandwiched by the $d(CGCG)_2$ sequences in the dodecamers **2-5**. The bottom of FIGs. 7-10 shows the spectra of the dodecamers under high salt conditions. Very little change in the spectra is observed for the three dodecamers except for the $d(CGCGTATACGCG)_2$. Those three dodecamers take on the B form under the high salt conditions, also. It is supposed that the central tetramers are resistant to the B-Z transition, and that they restrict the conformations of the duplexes to B form, which is preferable for them, overcoming the effects of the $d(CGCG)_2$ sequences at both ends.

Only $d(CGCGTATACGCG)_2$ shows spectral changes upon increasing the salt concentration. The intensity of a 1316 cm^{-1} line increases, and a new line appears at 619 cm^{-1} although its intensity is weak. In addition, a new line appears at 814 cm^{-1}, while the intensity of a 835 cm^{-1} line decreases. The 619 and 1316 cm^{-1} lines correspond to the C3'endo-syn guanosine, which is observed in Z form. Judging from the intensities of these lines, the population of the guanosine in the C3'endo-syn is estimated to be about 5-10 %. Both the 814 and 835 cm^{-1} lines reflect the main chain conformation. The appearance of the 814 cm^{-1} line and the decrease of the 835 cm^{-1} line are explained as the co-existence of Z form with B form. Thus it is concluded that the Z form (5-10 %) becomes co-existent with the B form under the high salt conditions for $d(CGCGTATACGCG)_2$.

It is said that the preferable sequence to take on Z form is an alternating pyrimidine-purine sequence.[9] $D(CGCGTATACGCG)_2$ is the only sequence which fits the conditions among the four dodecamers. Therefore

it is reasonable that the indication of the B-Z transition is detected only in $d(CGCGTATACGCG)_2$. We have already shown that the $d(TATA)_2$ is the most unstable among the four tetramers. It can not form a duplex. In other words, the $d(TATA)_2$ sequence in the $d(CGCGTATACGCG)_2$ does not restrict strongly the conformations of the $d(CGCG)_2$ portions at both ends to B form. This might be another aspect of the exclusive observation of the B-Z transition in $d(CGCGTATACGCG)_2$.

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REFERENCES

1. Nishimura, Y. (1985) Adv. Biophys. **20**, 59-74.
2. Nishimura, Y., Tsuboi, M., and Sato, T. (1984) Nucleic Acids Res., **12**, 6901-6908.
3. Nishimura, Y., Torigoe, C., and Tsuboi, M. (1986) Nucleic Acids Res., **14**, 2737-2748.
4. Nishimura, Y., Torigoe, C., and Tsuboi, M. (1985) Biopolymers, **24**, 1841-1844.
5. Katahira, M., Nishimura, Y., Tsuboi, M., Mitsui, Y., and Iitaka, Y. (1986) Biochim. Biophys. Acta, **867**, 256-267.
6. Katahira, M., Lee, S.J., Kobayashi, Y., Sugeta, H., Kyogoku, Y., Iwai, S., Ohtsuka, E., Benevides, J.M., and Thomas, G.J. (1990) J. Am. Chem. Soc., **112**, 4508-4512.
7. Torigoe, C., Nishimura, Y., Tsuboi, M., Matsuzaki, J., Hotoda, H., Sekine, M., and Hata, T. (1986) Spectrochim. Acta, 42A, 1101-1106.
8. Katahira, M., Katahira-Sakaguchi, R., Hayashi, F., Uesugi, S., and Kyogoku, Y. (1991) J. Am. Chem. Soc., **113**, 8647-8651.
9. Saenger, W. (1983) Principles of Nucleic Acid Structure, Springer-Verlag
10. Viswamitra, M.A., Kennard, O., Jones, P.G., Sheldrick, G.M., Salisbury, S., Falvello, L., and Shakked, Z. (1978) Nature, **273**, 687-688.

11. Thomas, G.A. and Peticolas, W.L. (1984) Biochemistry, **23**, 3202-3207.
12. Katahira, M., Sugeta, H., Kyogoku, Y., and Fujii, S. (1990) Biochemistry, **29**, 7214-7222.

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