 Nucleic Acids Research, 2001, Vol. 29, No. 3 **1–4**

SUPPLEMENTARY MATERIAL

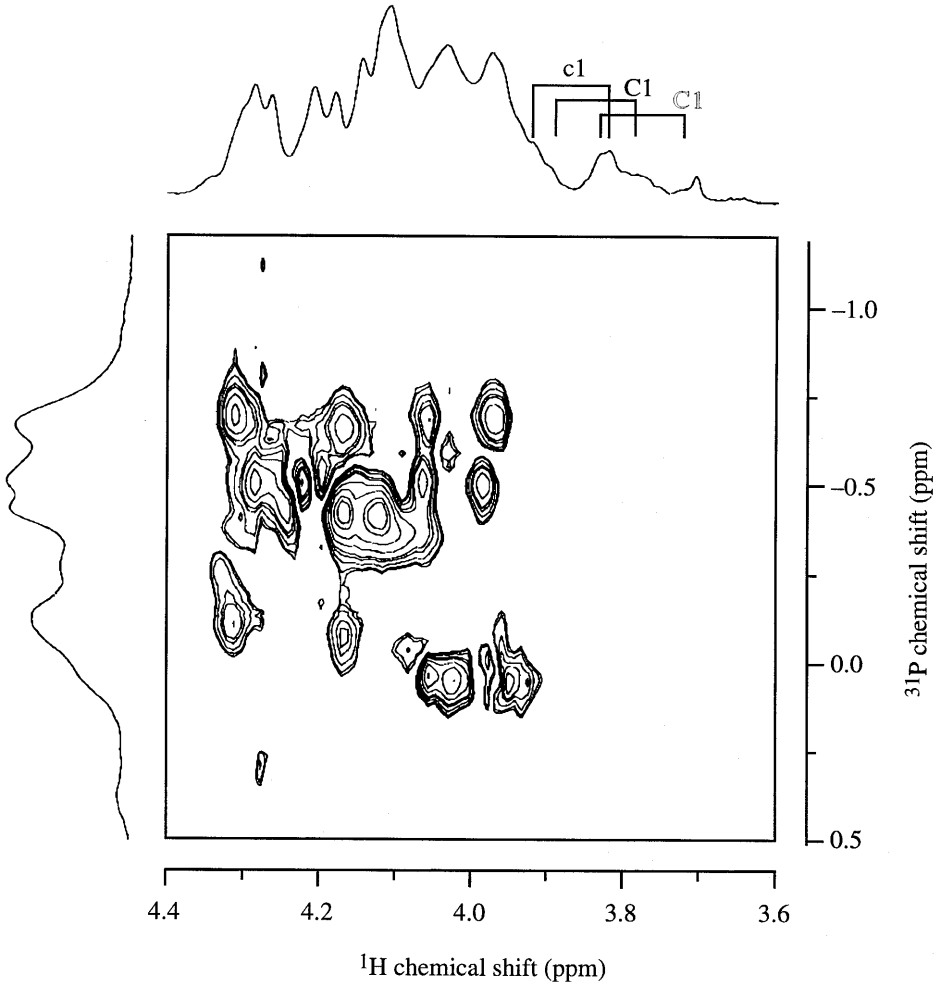

Figure S1. ^{1}H-^{31}P HMQC spectrum for d(CCCTAA) measured at 20°C, pH 4.5. The region of H5′/5″-^{31}P is displayed with 1D spectra. Lowercase, capital and outlined letters represent H5/5 peaks of *R*-, *S*- and *T*-form, respectively.

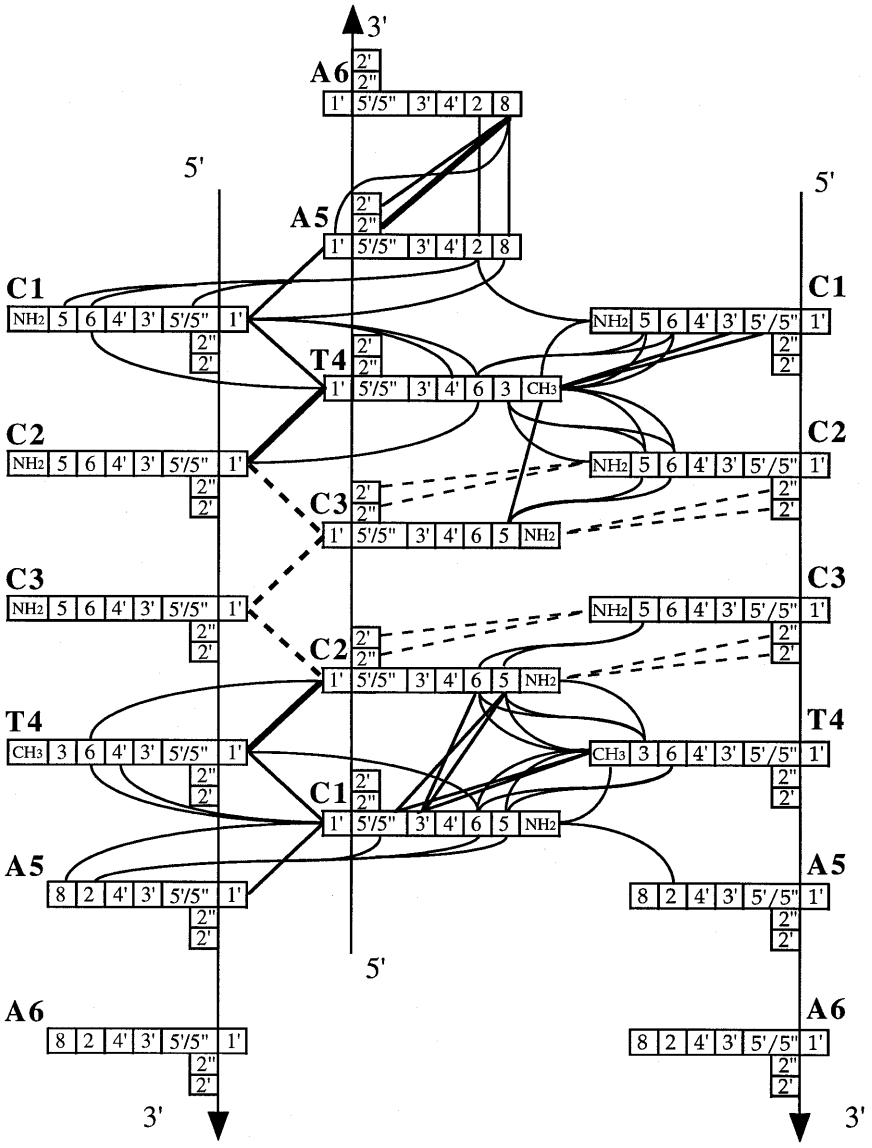

Figure S2. Schematic representation of inter-residue inter-proton distances derived from NOESY cross-peaks. NOE connectivities only from the central strand are shown. Strong and medium NOEs are indicated by thick lines, and weak and very weak NOEs by thin lines. Broken lines indicate distance restraints to maintain the i-motif structure in the region of C2 and C3 residues, which could not be observed separately on account of signal overlapping.

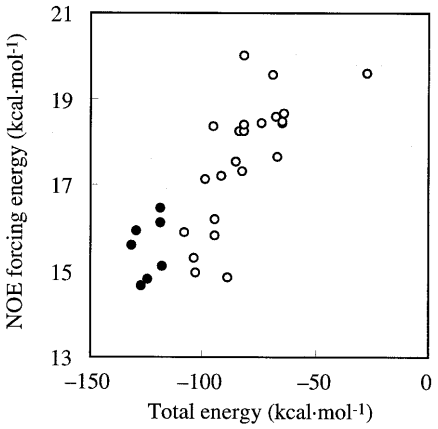

Figure S3. Correlation between total and NOE forcing energy terms of the restrained dynamics calculations for *T*-form. Circles correspond to the energy terms of all 30 calculated structures. Closed circles indicate the structures that are employed in the structure analysis. The total energy consists of seven energy terms, given in Table S2.

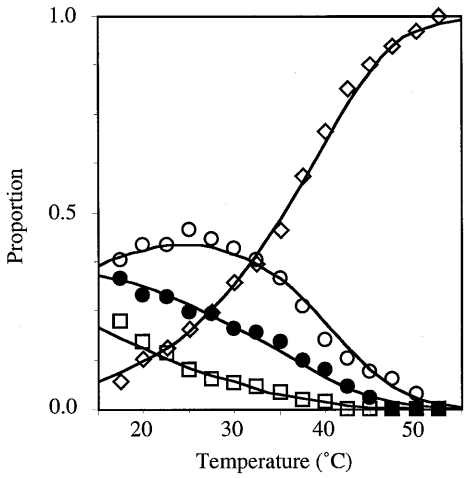

Figure S4. Denaturation profiles of the three d(CCCTAA) tetramers. Open and closed circles and open squares indicate the *R*-, *S*- and *T*-form tetramer fractions, respectively. Diamonds indicate the single-stranded fractions. Lines through the points represent theoretical population curves for each tetramer obtained by using ΔH and ΔS values in Table 2.

Table S1. ^{1}H chemical shifts (p.p.m.) of the three i-motif structures of d(CCCTAA) at pH 4.5 and 20°C

		H8/H6	H2/H5/CH_3	H1′	H2′	H2″	H3′	H4′	H5′, H5″	Others
C1	*R*	7.82	5.94	6.38	2.01	2.52	4.79	4.11	3.83, 3.93	
	S	7.76	5.89	6.00	2.01	2.41	4.71	3.98	3.80, 3.90	
	T	7.35	5.55	5.29	2.13	2.13	4.46	4.03	3.72, 3.83	H4 7.95, 8.84
	s	7.91	6.02	6.14	2.31	2.55	4.73	4.21	3.74, 3.79	
C2	*R*	7.75	5.88	6.39	2.14	2.58	4.73	4.10	3.89, 4.30	
	S	7.73	5.88	6.33	2.11	2.55	4.70	4.11	4.06, 4.28	
	T	7.68	5.64	6.32	2.12	2.45	4.82	4.11	3.98, 4.29	H4 7.97, 9.20
	s	7.92*	6.08	6.22	2.26	2.54	4.84	4.38*		
C3	*R*	7.66	5.97	6.13	1.90	2.46	4.60	4.11	3.97, 4.29	H4 8.38, 8.96
	S	7.64	5.87	6.33	2.09	2.42	4.69	4.06	4.00, 4.29	
	T	7.55	5.82	6.32	2.14	2.55	4.65	4.06	3.99, 4.25	
	s	7.94*	6.08	6.22	2.26	2.54	4.84	4.34*		
T4	*R*	7.42	1.76	5.97	2.02	2.28	4.59	4.12	4.02, 4.15	H3 11.35
	S	7.29	1.82	5.98	1.63	2.23	4.63	4.08	3.97, 4.15	H3 11.35
	T	7.20	1.71	5.93	2.10	2.20	4.55	4.00	4.00*, 4.21	H3 11.02
	s	7.39	1.84	5.97	1.80	2.23	4.76	4.15		
A5	*R*	8.11	7.78	6.02	2.45	2.57	4.92	4.27	3.96, 4.05	
	S	8.15	7.93	6.02	2.57	2.59	4.95	4.28	3.96, 4.05	
	T	7.94	7.90	5.94	2.39	2.55	4.88	4.13	4.00, 4.03	
	s	8.20	7.96	6.03	2.59	2.59	4.95	4.30		
A6	*R*	8.26	7.83	6.21	2.48	2.70	4.71	4.19	4.10, 4.15	
	S	8.27	7.87	6.25	2.48	2.68	4.72	4.21	4.12, 4.21*	
	T	8.23	7.73	6.15	2.47	2.68	4.76	4.21	4.14, 4.21*	
	s	8.32	7.99	6.27	2.48	2.72	4.77	4.25		

R, *S*, *T* and *s* indicate *R*-form, *S*-form, *T*-form and single-strand, respectively. Asterisks indicate ambiguous values due to signal overlapping.

Table S2. Final energy parameters[a] and characterization of the seven lowest energy structures

Total energy	–124.1 ± 5.6
Bond energy	8.4 ± 0.3
Angle energy	82.9 ± 4.4
Dihedral energy	255.7 ± 5.6
H-bond energy	0.3 ± 0.1
Non-bond energy	–248.5 ± 4.2
Coulomb energy	–219.4 ± 3.6
NOE forcing potential	15.5 ± 0.7
Largest NOE violations[b]	0.18 ± 0.02
Number of violations[c]	7.6 ± 0.8
Residual r.m.s.d. values[d]	
C1	0.64 ± 0.17
C2	0.81 ± 0.26
C3	0.60 ± 0.18
T4	0.84 ± 0.28
A5	1.24 ± 0.55
A6	2.37 ± 0.83

[a]The units of energy are in kcal/mol.
[b]The unit is Å.
[c]The number of the NOE violations, which is >0.1 Å.
[d]The unit is Å.
The r.m.s.d. values are calculated for each residue from the coordinates of the seven lowest-energy conformers superimposed on the averaged coordinate.