Volume 17 Number 1 1989 Nucleic Acids Research

Sequence criteria for Z-DNA formation: studies on poly d(ACGT)

Kumud Majumder, Rakesh K.Mishra, Manju Bansal and Samir K.Brahmachari*

Molecular Biophysics Unit, Indian Institute of Science, Bangalore 560 012, India
Submitted November 24, 1988

A novel polymer poly d(ACGT) has been synthesised and used for structural studies in order to understand the critical requirement of specific base sequences for stabilisation of Z-conformation in natural sequences. The oligonucleotide d(ACGTACGT) was synthesised and polymerised through ligation. The polymer (ACGT)n (n=8-12) was purified by gel filtration HPLC. The base sequence was characterised by RsaI (GTAC) digestion of the polymer.

Poly d(ACGT) exists in typical B-DNA conformation in 100 mM NaCl and does not undergo B to Z transition even in the presence of 5 M NaCl and 40 mM $NiCl_2$ (Fig 1) unlike poly d(AC).poly d(GT) which has been shown to give inverted CD spectrum characteristic of the left handed Z-conformation under this condition (1). No binding to poly d(ACGT) was observed with Z-DNA antibody and the polymer did not show any resistance to cleavage by RsaI under Z-favouring condition. These results show that poly d(ACGT) does not adopt left handed Z-conformation under conditions which are sufficient to induce B--->Z transition in poly d(AC).poly d(GT). Poly d(ACGT) in the B-form has few degrees higher Tm than poly d(AC).poly d(GT) under similar condition. Interestingly, under the influence of supercoiling force the ACGT track has been found to adopt cruciform structure whereas d(AC)n d(GT)n sequences adopt Z-conformation (2).

To understand this difference between these two polymers regarding their potential to adopt left handed Z-conformation model building studies have been carried out. In the Z-conformation, the GTA sequence is less favourable as compared to GTG by about 2.2 Kcal/mole due to unfavourable electrostatic repulsion between the three carbonyl group oxygens 06G, 04T and 04T (cross strand)in the GpTpA triplets. TGC and GCA sequences which have been shown to adopt Z-conformation in oligonucleotides in solution (3) do not pose such unfavourable electrostatic interactions. Thus, exact base-base interaction decides the relative potential for a given sequence to adopt Z-conformation. This makes (AC)n.(GT)n sequence to have higher Z-potential than (ACGT)n. In genomic DNA long stretches of most Z-potential (CG)n sequences and least Z-potential (ACGT)n are rare while sequences like (TG)n which have moderate potential to adopt Z-conformation occur widely.

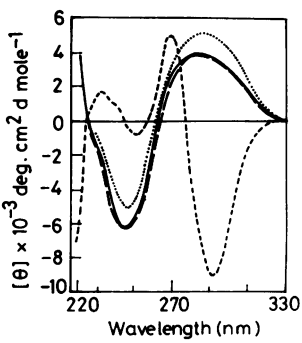

Fig 1. CD spectrum of poly d(ACGT) in 1 mM sodium cacodylate, 1 mM EDTA(pH 7.4) with 100 mM NaCl(·····) 5 M NaCl(— — —), 5 M NaCl + 40 mM $NiCl_2$(———). A similar spectrum was obtained in the presence of 2 M $MgCl_2$ or 2 mM hexamine cobalt chloride. Poly d(AC).Poly d(GT) in 5 M NaCl + 40 mM $NiCl_2$(------).

ACKNOWLEDGEMENT: Authors thank CSIR (India) for financial support. RKM is a NBTB PDF.

*To whom correspondence should be addressed

REFERENCES: 1.Taboury, J.A. and Taillandier, E. (1985) Nucl.Acids.Res. 13,4469. 2. Nayloc, L.H., Yee, H.A. and van de Sande, J.H. (1988) J. Biomol. Str. Dyn. 5,895. 3. Mishra, R.K., Latha, P.K. and Brahmachari, S.K. (1988) Nucl.Acids Res. 16, 4651.