Callahan, Daniel Eugene

CONFORMATION OF DNA IN SOLUTION: EXPERIMENTAL AND THEORETICAL CIRCULAR DICHROISM STUDIES

University of California, Santa Barbara PH.D. 1986

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Santa Barbara

Conformation of DNA in Solution: Experimental and Theoretical Circular Dichroism Studies

A Dissertation submitted in partial satisfaction
of the requirements for the degree of

Doctor of Philosophy

in

Chemistry

by

Daniel Eugene Callahan

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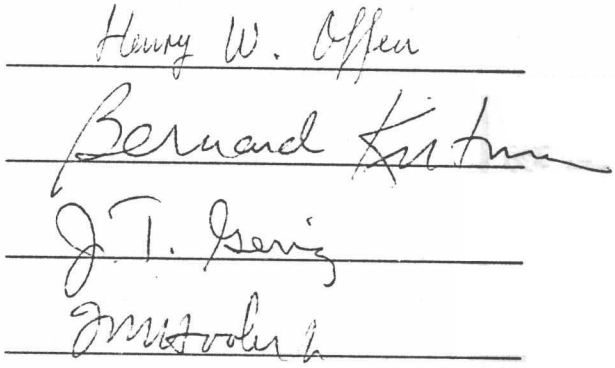
October 1986

The dissertation of Daniel Eugene Callahan is approved:



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October 1986

October 10, 1986

I dedicate this work to my wife, Franci

and to my parents, Dr. and Mrs. Donald Edward Callahan

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PUBLICATIONS

"Conformation of DNA in Solution: Circular Dichroism Calculations Based on Crystal Structures of B- and Z-DNA Fragments". D.E. Callahan and T.M. Hooker, Jr., Biopolymers, in the press.

"Circular Dichroism and Conformation of DNA Oligomers in Solution". D E. Callahan and T.M. Hooker, Jr., presented at the University of California-Santa Barbara Department of Biological Sciences Research Seminar, May 14, 1985.

"Calculated CD Spectra for B- and Z-DNA". D.E. Callahan and T.M. Hooker, Jr., presented at the 1985 Pacific Conference on Chemistry and Spectroscopy, San Francisco, Abstract #D6.

"Conformation of DNA in Solution: Comparison of Calculated and Experimental CD of A-, B- and Z- DNA". D.E. Callahan, A. Sanchez and T.M. Hooker, Jr., presented at the 1986 ASBC/Biological Chemistry Division, ACS Meeting, Washington D.C., Abstract #844.

ABSTRACT

Conformation of DNA in Solution: Experimental and Theoretical Circular Dichroism Studies

Calculations of the circular dichroism (CD) spectrum of DNA have, in some instances, been unable to reproduce observed spectra. In work presented here, an attempt was made to relate structural details of DNA conformation to observable features in the CD spectrum. CD calculations were performed using an origin-independent matrix formalism which employed a variable dielectric function for the evaluation of intertransition interactions. Calculations were based on available x-ray crystal structures of d(CGCGAATTCGCG), d(CGCGAATTbr^{5}CGCG), d(CGCG), d(CGCGCGCGCGCG) and d(i^{5}CCGG). These structures represent DNA in the B, Z and A conformations. Calculations were also performed on idealized A or B DNA models of these sequences and some related sequences. Several different optical parameter sets were used, and an attempt was made to explain the CD spectra of B, Z and A DNA using only the near-ultraviolet transitions of the nucleic acid bases. Alternate polarization directions for these near-ultraviolet transitions were examined. Experimental CD spectra were obtained for d(CGCGAATT-CGCG), d(CGCGAATTm^{5}CGCG), d(CGCGCGCGCGCG), d(CCGGCCGGCCGG) and

d(m^5CCGGm^5CCGGm^5CCGG) in various solvent systems. Good agreement was observed between experimental and theoretical CD spectra of d(CGCGAATTCGCG) when a variable dielectric constant was used. This indicated that the x-ray crystal structure of d(CGCGAATTCGCG) is a possible solution conformation for this molecule in solution. However, a more idealized B conformation was also seen to be a possible solution conformation. CD calculations were very sensitive to the variations of local helix parameters parameters in the structures of d(CGCGAATTCGCG) and d(CGCGAATTbr^{5}CGCG) and the bromine substituent was predicted to perturb the bending of the helix in a way which altered theoretical CD spectra. However, no difference was observed in the experimental CD spectra of d(CGCGAATTCGCG) and d(CGCGAATTm^{5}CGCG). If a variable dielectric constant was used, it was possible to calculate an inverted CD spectrum for Z form d(CGCGCGCGCGCG), but overall agreement between theory and experiment was poor. A series of conformational transitions were observed for d(CCGGCCGGCCGG) and d(m^5CCGGm^5CCGGm^5CCGG) as solvent conditions were varied, and the methyl substituent was seen to perturb the conformation. CD calculations indicated that both fragments may adopt an A conformation in 80% 2,2,2-trifluoroethanol. In addition, the oligomer d(CCGGCCGGCCGG) may adopt an A conformation in 4 M NaCl, while the oligomer d(m^5CCGGm^5CCGGm^5CCGG) adopts a B conformation in both 0.1 M and 4 M NaCl.

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CHAPTER I

DNA CONFORMATION