Nuclear Magnetic Resonance Study on the CRP and CRP*RNA polymerase complex

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Cyclic AMP receptor protein(CRP) from E. Coli plays a key role in regulation of the expression of more than 20 genes of the bacterium. CRP binds in the presence of cAMP to a specific target site near the promoter of each gene under its regulation. CRP is a dimer (Mr~47.000) of two identical subunits. There are two binding domains in the CRP monomer, one for the binding of the cAMP and the other for the binding of specific DNA sequences. In order to elucidate the structure-function relationship of CRP, we have taken NMR study of this protein and its complex with cAMP, oligonucleotides, and RNA polymerase α subdomain. To assign the resonances of protein, we used stable isotope labeling (15N, ¹³C, Deuterium). Several proton signals monitored in this study indicate that the contration of the CRP molcule only occurs in the cAMP-binding domain and not in the DNA-binding domain. The conformational transition mostly occurs when one cAMP molecule binds to one of the dimer subunits, but is completed only when both cAMP binding sites are saturated. The protein-protein interaction between CRP and RNA polymerase α subunit was also investigated with carbonvl ¹³C NMR.