

## NMR study of the interaction of T4 Endonuclease V with DNA

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T4 Endonuclease V (Mw 16,000) acts as a repair enzyme for UV induced pyrimidine dimers in DNA. Many researchers have studied the biochemical characteristics of the enzyme. However the precise action mechanism of T4 endo V has not fully elucidated yet. In our laboratory NMR spectroscopy technique is being used for the structural study of T4 endo V. Because of its low temperature stability and high content of  $\alpha$ -helix, the conventional  $^1\text{H}$  NMR technique was inapplicable. Therefore we utilized stable isotope labeling technique and so far prepared about 10 amino acid specific labeled proteins. The HSQC spectra of amino acid specific labeled proteins will help us to interpret the triple resonance 3D, 4D data which are under processing. We also studied the behaviors of specific amino acid residues whose roles might be critical. When the enzyme labeled by  $^{15}\text{N}$ -Thr was mixed with the substrate oligonucleotide (semispecific -TT- sequence), one crosspeak in its HSQC spectrum was completely disappeared, which means that one of seven Thr residues is in the binding site of the enzyme with DNA. This result is well consistent with previous report that implicated the Thr 2 residue in the activity of the enzyme. Similar studies were carried on the behaviors of Arg and Tyr residues.