Infrared and Circular Dichroism Studies of the Secondary Structure of *Lac* Repressor, its Tryptic Core and its N-Terminal Headpieces

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Lac repressor is a tetrameric protein (4 x 360 residues), which controls the expression of the *lac* operon by binding to the *lac* operator. Limited trypsinolysis of *lac* repressor yields four N-terminal headpieces each containing 51 or 59 residues, and a tetrameric core with full inducer binding activity. The major part -if not all- of the binding sites for DNA and *lac* operator are located on the headpieces.

We have used circular dichroism and infrared spectroscopy to investigate the secondary structure of these various domains. The values for the content of α-helical structure for the *lac* repressor (based on CD spectroscopy) as reported by several groups are rather similar ranging from 33 to 40 %, whereas the reported values for the content of β-structure are very different, ranging from 9 to 42 %.

In order to improve the estimation of the β-content we have analyzed the infrared spectra of *lac* repressor and its tryptic core in the amide I' region. Using protein-derived reference spectra we find a β-content for *lac* repressor of 18 % and of 23 % for its tryptic core. The higher percentage of β-structure in the core is confirmed by another type of analysis (decomposition of the spectra in Gaussian curves). But as the whole repressor has 59 amino acids more than its tryptic core the absolute number of residues involved in β-structure is about the same for the core as for the whole repressor. Thus the N-terminal headpieces should contain very little or no β-structure at all. In fact the infrared spectrum of the headpieces shows no shoulder at 1630 cm^{-1} which excludes the existence of a large β-sheet in this N-terminal part of the *lac* repressor. The α-helical content of the headpiece was calculated to be 40 % using CD spectroscopy.

These results, particularly the lack of extended β-structure in the part of repressor which interacts with the operator, are discussed with respect to previous models of *lac* repressor-*lac* operator interactions.