($=1/T_1$) value of Ala^2-NH pointing to its accessibility to the solvent (Figure 1). In Table I the amide relaxation rates as $R_{1p} = (1/T_{1\,exp}) - (1/T_1)$ are given. T_2 relaxation times from line widths were also investigated. As expected addition of TEMPO caused a different broadening of each of the three amide resonances—the R_2^* (24.4 s^{-1}) for Ala^2-NH is faster than R_2^* (3 s^{-1}) for residues 1 and 3. The results clearly support the proposal that Ala^1 and AEO^3 amide protons are shielded from the solvent.[12]

All the NMR data therefore unambiguously indicate the differences in the environment of Ala^2-NH proton compared with that of Ala^1-NH and AEO^3-NH. The latter two protons are solvent shielded; Ala^2-NH is instead accessible to the solvent. IR data indicated that even at low concentration two protons are hydrogen bonded. We propose therefore that Ala^1-NH and AEO^3-NH are involved in intramolecular hydrogen bonds. A similar hydrogen-bonding pattern leading to a bis-γ-turn configuration was found for other cyclic tetrapeptides.[14]

Although more detailed studies will be necessary, a conformation containing two γ turns can be also proposed for **2** (Figure 2). The derivatization of **1** to **2** yielded no significant changes in chemical shift or $^3J_{NH-\alpha}$ of α and amide protons, indicating that no conformation modification had occurred in the peptide ring (see Table I). Furthermore, the ^{13}C chemical shift for the Pro^4 C_β of **1** in chloroform solution is consistent with a γ turn involving the Pro^4 residue.[15] We therefore suggest that HC toxin also assumes the conformation of Figure 2.

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Registry No. 1, 83209-65-8; **2**, 89890-87-9.

Far-Ultraviolet Electric Linear Dichroism of Poly(α-L-glutamic acid) in the Helical and Coiled Conformations[1]

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In order to verify Moffitt's prediction that the absorption spectrum of the peptide chromophore in helical polypeptides should show a band split,[2] we initiated an electric linear dichroism (ELD) study in the far-ultraviolet region.[3] We can now report some preliminary results on reduced dichroism ($\Delta A/A$) of electric-field-oriented poly(α-L-glutamic acid), (Glu)$_n$, in helical and coiled conformations in the 230–187-nm wavelength region. On the basis of the dependence of ($\Delta A/A$) on wavelength and applied field strength, we conclude that at least three apparent component bands are involved in the isotropic spectra of the peptide chromophore of (Glu)$_n$ in the two extreme conformations and that the transition moment direction of each band makes a different angle with respect to the orientation axis of the polymer chain.

The ELD apparatus consists of an electric pulsing system and an optical system, which contains a 200-W deuterium lamp, a double-grating monochromator, and a Rochon-type magnesium fluoride polarizer. The details will be given elsewhere,[3] together with data acquisition and processing.[3-6] The sodium salt of purified (Glu)$_n$ with a degree of polymerization of 708 was dissolved in distilled water (pH 6.89). By dialysis of this stock solution against dilute HCl, a salt-free acid (Glu)$_n$ solution was prepared (pH 4.13). The reduced dichroism is given as $\Delta A/A = (A_\parallel^E - A_\perp^E)/A = (^3/_2)(3\cos^2\theta - 1)\Phi$, where $A_\parallel^E$ is the absorbance of a sample solution for incident light linearly polarized in parallel to an applied electric field, $A_\perp^E$ is the same in perpendicular to the field, A is the field-free isotropic absorbance, θ is the angle between the orientation axis of (Glu)$_n$ and the direction of a transition moment, and Φ is the orientation factor at a given field strength E.[7,8]

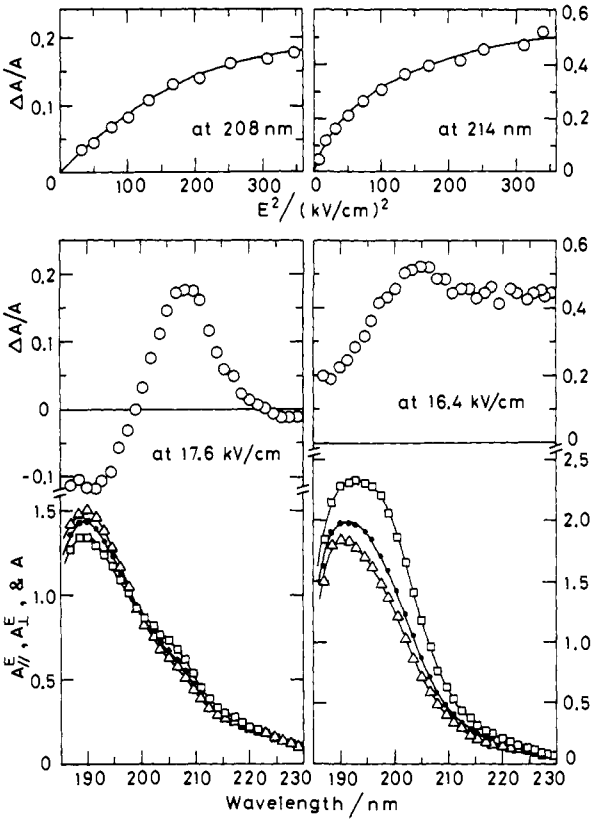

Figure 1. Dependence of reduced dichroism $\Delta A/A$ on the second power of electric field strength E^2 (top) and on wavelength (middle), and the absorption spectra (bottom) in the presence ($A_\parallel^E$ and $A_\perp^E$) and in the absence (A) of external electric field for helical (Glu)$_n$ (left halves) and coiled (Glu)$_n$ (right halves). Symbols denote experimental points (□ for $A_\parallel^E$, Δ for $A_\perp^E$, and ○ for $\Delta A/A$). The ELD signals were measured at 20 °C with "Kerr" cells, whose path lengths are 0.60 and 2.00 cm,[4,5] while the isotropic spectra (—·—) were measured on a Hitachi EPS-3T recording spectrophotometer with a pair of 0.5-cm quartz cells under nitrogen gas purge. Absorbances on the ordinate were all normalized to a path length of 0.6 cm. The residue concentrations of (Glu)$_n$ are 0.58 mM at pH 4.13 and 0.43 mM at pH 6.89. The relation $3A = A_\parallel^E + 2A_\perp^E$ was found to hold over an entire field-strength region (0–20 kV/cm) in all cases.

The dependence of $\Delta A/A$ on the square of electric field for helical and coiled (Glu)$_n$ is shown at the top of Figure 1. $\Delta A/A$ values obey the Kerr law in the low field region, but tend to saturate at high field strengths. By extrapolating $\Delta A/A$ values to infinitely high field, the saturated reduced dichroism, $(\Delta A/A)_s$,[6,8] could be estimated to be 0.280 at 208 nm for helical (Glu)$_n$ and 0.719 at 214 nm for coiled (Glu)$_n$. Hence, the degree of orientation in terms of Φ is ca. 0.61 at a field strength of 17.6 kV/cm for helical (Glu)$_n$ and ca. 0.65 at 16.4 kV/cm for coiled (Glu)$_n$. The anisotropic absorption spectra, $A_\parallel^E$ and $A_\perp^E$, were

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measured at those constant field strengths. They are shown at the bottom of Figure 1, together with field-free isotropic spectrum A. The $A_{\parallel}^{E}$ and $A_{\perp}^{E}$ spectra of helical $(Glu)_n$ cross each other at 199 nm and near 223-222 nm. For coiled $(Glu)_n$, the $A_{\parallel}^{E}$ spectrum is larger than the $A_{\perp}^{E}$ spectrum throughout the 230-187-nm region, while the peak of $A_{\parallel}^{E}$ is shifted toward the red side by 2 nm and that of $A_{\perp}^{E}$ slightly toward the blue relative to the peak of the isotropic spectrum. Since the relation $3A = A_{\parallel}^{E} + 2A_{\perp}^{E}$ holds for the helical and coiled states within experimental errors, the electrochromism does not seem to occur.[8]

The dependence of $\Delta A/A$ on wavelength was determined, for the first time, for the peptide chromophore in the 230-187-nm region (middle figures). It is the dependence of reduced dichroism $\Delta A/A$,[6] but not dichroism ΔA,[9] on wavelength and on field strength that quantitatively reveals spectral features, such as angles, θ, numbers of transition moments, and weak absorption bands hidden in a solution[10] or film spectrum.[11] Changes in $\Delta A/A$ with wavelength indicate that at least three overlapping absorption bands, each with a different angle, exist in the peptide spectra of $(Glu)_n$ above 187 nm regardless of its overall conformations. For helical $(Glu)_n$, those bands may be separated into three regions: 230-215, 215-200, and 200-187 nm. For coiled $(Glu)_n$, $\Delta A/A$ values unexpectedly show a three-step change: a constant 230-210-nm region, a maximum in 205-203 nm, and a minimum near 190 nm. Therefore, the broad 190-nm peptide band of coiled $(Glu)_n$ must be a composite of at least two intense component bands and a weak one. The nonsplit broad peak in the isotropic spectrum has been considered to be characteristic of the random-coiled conformation,[12,13] but we now propose that the spectral feature of the coiled form is closely related to that of the helical form in the 210-187-nm region.

The 230-210-nm band has been assigned to an n-π^* transition for helical and coiled $(Glu)_n$ because of its weak intensity.[13] The value of $(\Delta A/A)_s$ should be +3.0 or -1.5, if an isolated transition moment is either parallel ($\theta = 0°$) or perpendicular ($\theta = 90°$) to the orientation axis ($\Phi = 1$).[4,6,8] By using observed values of $(\Delta A/A)_s$, the angles were calculated to be ±51.0° at the 208-nm shoulder and ±57.3° at the 190-nm peak for completely oriented $(Glu)_n$ helices. This unexpected result is partly due to a strong overlap of the two closely located bands. It is, however, difficult to simulate the observed wavelength dependence of $\Delta A/A$ on the basis of a Moffitt-type band split,[2,9,13] unless the intensity ratio of the 208-nm band (the parallel band[2]) to the 190-nm band (the perpendicular band[2]) is assumed to be ca. 1.0:1.5 even at the center of the 208-nm band and also the 190-nm band is overlapped by another strongly positive ($\Delta A/A > 0$) band below 190 nm. It is difficult at present to evaluate the exact angle of the two transition moments inside the broad 190-nm peptide band of coiled $(Glu)_n$, because (i) the axis of orientation may not coincide with the axis of molecular symmetry[14] and (ii) the 205-203-nm and 190-nm transitions overlap each other. A rough estimate, however, indicates that these transitions appear to be inclined neither at 0° nor at 90° relative to the $(Glu)_n$ chain.

This ELD study revealed that, if the predicted band split of a peptide chromophore occurs,[2] it should be seen in the isotropic spectra of $(Glu)_n$ not only in helical but also in coiled conformations. Careful reinvestigations are needed on the polarization spectra of low-molecular-weight amide compounds for better understanding the optical transition and the solution conformation of polypeptides.

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Rhodium(I)-Catalyzed Hydrogenation of Olefins. The Documentation of Hydroxyl-Directed Stereochemical Control in Cyclic and Acyclic Systems

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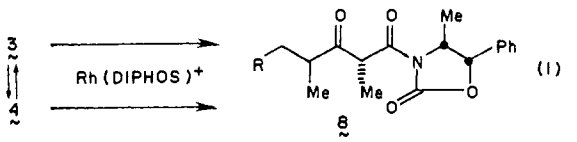
Several studies have recently appeared that document the important observation that proximal hydroxyl groups can be employed to "direct" the stereochemical course of selected transition-metal-catalyzed olefin hydrogenations. Stork[1] and Crabtree[2] have convincingly demonstrated that the cationic iridium catalyst $Ir(COD)(py)(PCy_3)PF_6$ (**1**)[3] is effective in the directed reduction of *cyclic* allylic and homoallylic alcohols. In a complimentary study, Brown and Naik[4] have provided two cases where the cationic rhodium catalyst $[Rh(NBD)(DIPHOS\text{-}4)]BF_4$ (**2**)[5] is also responsive to the same stereochemical control elements as demonstrated in the reduction of an *acyclic* allylic and homoallylic alcohol. In ongoing studies in our laboratory we have had the opportunity to critically evaluate each of these catalysts in both cyclic and acyclic systems, and our *unanticipated* observations should prove to significantly extend the utility of these hydrogenation catalysts.

On the basis of the principles of acyclic conformational analysis in allylic systems,[6] the hydroxyl-directed reduction of the allylic alcohols **3** and **4**,[7] via their preferred C_3-C_4 conformers might be expected to lead stereoselectively to the complementary diastereomeric aldol adducts **5** and **6**, respectively (Scheme I).[8] Surprisingly, the iridium catalyst **1**, which exhibits excellent levels of directed hydrogenation when the hydroxyl function is rigidly disposed on one olefin diastereoface,[1] proved ineffective in the stereoselective reductions of acyclic allylic alcohols **3** and **4** either at atmospheric (Table I) or elevated hydrogen pressures.[9] Even more surprising was the observation that the cationic rhodium complex **2** appeared to afford low levels of reaction diastereoselection under the reported hydrogenation conditions (CH_2Cl_2, 25 °C, 15 psi H_2).[4] In addition, the presence of ketonic byproducts such as **8** (eq 1) suggested that competitive olefin isomerization



(1)

might be a major problem. Fortunately, these reductions were found to respond dramatically to an increase in hydrogen pressure. For example, when the analogous reactions were carried out at elevated hydrogen pressures (640 psi), this catalyst exhibited

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