Polydepsipeptides. 10. Helix–Coil Transitions of Sequential Polydepsipeptides Having Protected Polar Side Chains

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ABSTRACT: The helix-to-coil transitions of sequential polydepsipeptides containing residues with protected polar side chains have been studied by circular dichroism. Poly[L-Ala-L-Glu(OBzl)-(*S*)-Lac] and poly[(L-Glu(OMe))$_2$-(*S*)-Lac] have been studied in tetrahydrofuran. Poly[(L-Glu(OMe))$_2$-(*S*)-Lac] and poly[L-Lys-(Z)-L-Glu(OMe)-(*S*)-Lac] have been studied in trifluoroethanol. At room temperature and below, the polydepsipeptides form stable right-handed α helices which can be denaturated thermally. The three polymers exhibit similar conformational stabilities in both solvents, suggesting that the nature of the side chain and solvent do not affect their helical stability and that the hydrogen-bonding pattern of the polydepsipeptides is the main factor affecting their conformational stability.

Introduction

Previous studies of the conformational properties of polydepsipeptides were concerned with polymers having aliphatic side chains such as methyl (L-alanine and (*S*)-lactic acid), isopropyl (L-valine and (*S*)-α-hydroxyisovaleric acid), and isobutyl (L-leucine).[2-6] The sequence of the repeating unit has been shown to determine the structure and stability of these polydepsipeptides in a consistent fashion. Polydepsipeptides having the repeating unit [AA-HA] (where AA stands for α-amino acid and HA stands for α-hydroxy acid), such as poly[L-alanyl-(*S*)-lactic acid] and poly[L-valyl-(*S*)-lactic acid], are disordered at room temperature in solvents such as chloroform, tetrahydrofuran, and trifluoroethanol. When the temperature is lowered to below −30 °C, a transition from the disordered form to a helical form is observed. The helix formed, however, is not the α helix but a left-handed helix consisting of concatenated β-I turns.[2]

Sequential polydepsipeptides having the repeating unit [AA-AA-HA], such as poly[L-alanyl-L-alanyl-(*S*)-lactic acid] (poly[(Ala)$_2$-(*S*)-Lac]) and poly[D-alanyl-D-alanyl-(*S*)-lactic acid] (poly[(D-Ala)$_2$-(*S*)-Lac]), are in an α-helical conformation at room temperature in solvents such as chloroform and tetrahydrofuran. The helix can be denaturated thermally or by means of solvents such as trifluoroethanol. Semiempirical energy calculations reported previously[2] have shown that the (*S*)-hydroxy acid units of the polydepsipeptide chains can be readily incorporated into the right-handed α helix without significant loss of van der Waals and dipolar forces, which are essential factors affecting the stability of the helix. The major perturbation of replacing several amide bonds by ester bonds is the reduction of the number of hydrogen bonds stabilizing the helix. Therefore, polydepsipeptides appear to be excellent models of polypeptides, and their conformational properties reflect the importance of hydrogen bonding in protein conformational stability.

After establishing the importance of the depsipeptide hydrogen-bonding pattern on the conformational properties of polydepsipeptides, we extended our observations to polydepsipeptides containing other amino acid residues with polar side chains to investigate the influence of the nature of these side chains on the conformational stability of the polydepsipeptides.

In this paper, we report a conformational study of sequential polydepsipeptides having the repeating unit [AA-AA-HA] and containing side-chain-protected residues of L-lysine and L-glutamic acid. The preceding paper in this series[7] was concerned with the synthesis of these polydepsipeptides containing (*S*)-lactic acid and side-chain-protected L-lysine and L-glutamic acid. All of these polymers appeared to be in a right-handed α-helical conformation at room temperature in solvents such as chloroform, tetrahydrofuran, and trifluoroethanol. In our present conformational study we show that these polydepsipeptides undergo a thermally induced conformational transition in tetrahydrofuran and in trifluoroethanol.

Experimental Section

Poly[L-alanyl-γ-benzyl-L-glutamyl-(*S*)-lactic acid] (poly[Ala-Glu(OBzl)-Lac]), poly[γ-methyl-L-glutamyl-γ-methyl-L-glutamyl-(*S*)-lactic acid] (poly[(Glu(OMe))$_2$-Lac]), and poly-[N^ϵ-carbobenzoxy-L-lysyl-γ-methyl-L-glutamyl-(*S*)-lactic acid] (poly[Lys(Z)-Glu(OMe)-Lac]) have been synthesized by polycondensation of the corresponding tripeptide pentachlorophenyl esters in solution in *N*,*N*-dimethylacetamide as described in the preceding paper.[7] The polymers were isolated by precipitation in water. Further purification was achieved by dissolution in chloroform, washing with water, drying over $MgSO_4$, precipitation in ether, and drying in vacuo. Their intrinsic viscosities determined in dichloroacetic acid at 25 °C were 0.2 dL/g for poly-[Ala-Glu(OBzl)-Lac], 0.55 dL/g for poly[(Glu(OMe))$_2$-Lac], and 0.92 dL/g for poly[Lys(Z)-Glu(OMe)-Lac], suggesting molecular masses ($\bar{M}_w$) of 30 000, 80 000, and 140 000 daltons for each of the polymers, respectively.

The solvents used were trifluoroethanol (Aldrich, Gold Seal), tetrahydrofuran (Aldrich, reagent grade), and 1,1,1,3,3,3-hexafluoro-2-propanol (Aldrich). The tetrahydrofuran was refluxed and distilled over sodium in an atmosphere of dry nitrogen and stored on 4-Å molecular sieves.

Circular dichroism spectra were obtained with a modified Cary 61 spectropolarimeter controlled by a Texas Instruments 980A minicomputer. All measurements were obtained with a Helma Co. 0.1-mm cell, which was thermostated to ±0.2 °C by a Lauda 2 k/R circulating bath. Actual temperatures within the cell were monitored with a Yellow Springs Instruments calibrated thermistor. Each spectrum is the average of multiple scans at 0.3-nm resolution. Concentrations of the polymers (1–1.2 mg/mL) were determined by weight. Results were expressed as mean residue ellipticity, $[\theta]$, defined as

$$[\theta] = \theta \times M/(10lc) \quad (\text{deg cm}^2)/\text{dmol}$$

where θ is the observed ellipticity, M is the average residue molecular weight, taken as one-third the [AA-AA-HA] repeating unit molecular weight, l is the optical path length in cm, and c is the polymer concentration in g/cm^3.

Results and Discussion

The CD spectra of poly[Ala-Glu(OBzl)-Lac] and poly-[(Glu(OMe))$_2$-Lac] in tetrahydrofuran at different temperatures are shown in Figures 1 and 2. Similar spectra for the polydepsipeptides poly[(Glu(OMe))$_2$-Lac] and poly[Lys(Z)-Glu(OMe)-Lac] in trifluoroethanol are shown in Figures 3 and 4. For all four polymers, the CD spectra indicate that right-handed α-helical conformations are assumed below room temperature. If the solutions con-

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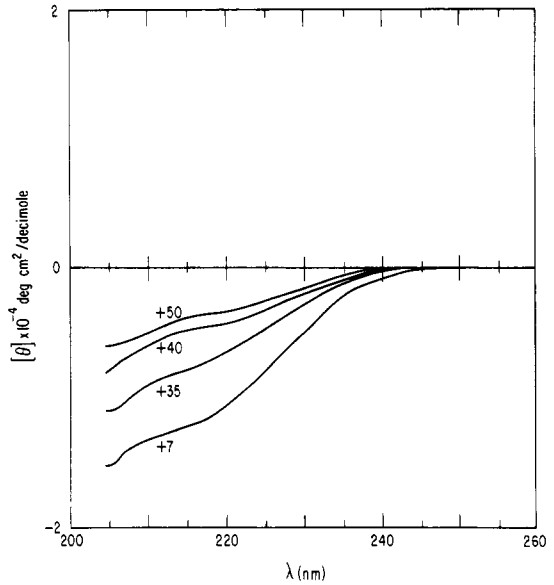


Figure 1. Temperature-dependent CD of poly[L-Ala-L-Glu(OBzl)-(*S*)-Lac] in tetrahydrofuran at the indicated temperatures.

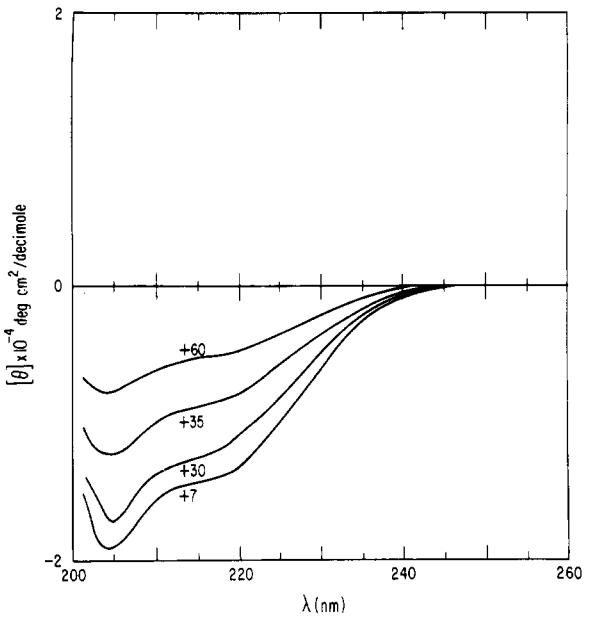


Figure 2. Temperature-dependent CD of poly[(L-Glu(OMe))$_2$-(*S*)-Lac] in tetrahydrofuran at the indicated temperatures.

taining these polydepsipeptides are warmed, the observed dichroism decreases in intensity. Such changes in the observed dichroism indicate that these polymers undergo thermally induced helix-to-coil transition in these solvents. The transitions are completely reversible.

The spectra observed for poly[Ala-Glu(OBzl)-Lac] and poly[(Glu(OMe))$_2$-Lac] at 0 °C in tetrahydrofuran are compared in Figure 5 to that observed for poly[(Ala)$_2$-Lac] under the same conditions. Both the line shapes and intensities of the three polymers are different. Poly[(Ala)$_2$-Lac] exhibits a minimum at 204 nm which arises from the parallel lobe of the $\pi\pi^*$ transition and a shoulder at 222 nm which is assigned to the $n\pi^*$ transition. The two transitions overlap strongly. A similar spectrum is observed for poly[Ala-Glu(OBzl)-Lac], but the intensity of the two transitions is greater than for poly[(Ala)$_2$-Lac]. This increase in intensity is also observed for poly[(Glu(OMe))$_2$-Lac], but the $n\pi^*$ and $\pi\pi^*$ transitions are more fully resolved.

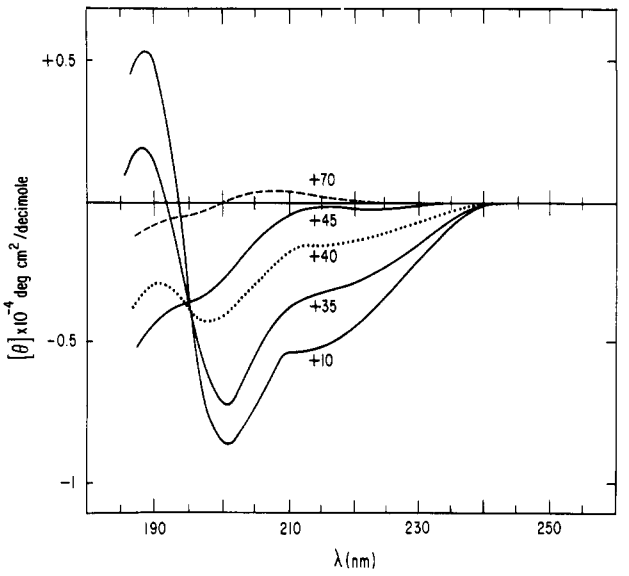


Figure 3. Temperature-dependent CD of poly[(L-Glu(OMe))$_2$-(*S*)-Lac] in trifluoroethanol at the indicated temperatures.

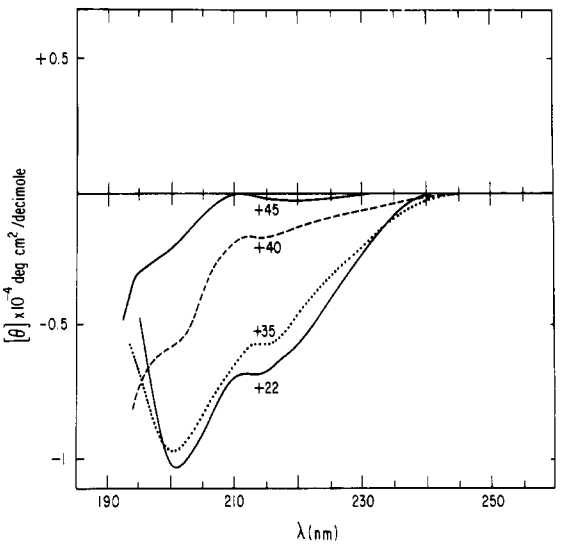


Figure 4. Temperature-dependent CD of poly[L-Lys(Z)-L-Glu(OBzl)-(*S*)-Lac] in trifluoroethanol at the indicated temperatures.

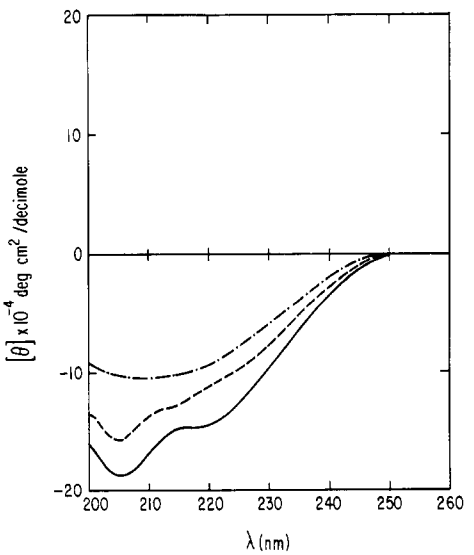


Figure 5. Comparison of the CD spectra of poly[(Ala)$_2$-Lac] (-·-), poly[Ala-Glu(OBzl)-Lac] (——), and poly[(Glu(OMe))$_2$-Lac] (---) in tetrahydrofuran at 0 °C.

The observed circular dichroism of helical polydepsipeptides in organic solvents is similar to, but not identical with, that observed for polypeptides. Aside from differences in conformation, polypeptides and polydepsipeptides would not be expected to exhibit the same spectrum for a helical structure since there are different types of chromophores in the main chains of the two types of polymers. Polypeptides contain amide chromophores while polydepsipeptides have amide as well as ester chromophores. The absorption maxima of the low-energy transitions of the amide and ester chromophores do not occur at the same wavelength and the transition dipoles, although similar, are not identical.[4] These differences most likely account for the difference in line shape between a helical polypeptide and a helical polydepsipeptide. We are currently investigating the relative contributions of conformations and main-chain chromophores to the observed circular dichroism of polydepsipeptides.

A reversible helix-to-coil transition is also observed for poly[$(Glu(OMe))_2$-Lac] and poly[Lys(Z)-Glu(OMe)-Lac] in trifluoroethanol. The limiting helical dichroism of the $n\pi^*$ transition at −40 °C is −5400 (deg cm^2)/dmol for poly[$(Glu(OMe))_2$-Lac] and −7600 (deg cm^2)/dmol for poly[Lys(Z)-Glu(OMe)-Lac]. At high temperature these values decrease to 300 and 0 (deg cm^2)/dmol, respectively. The spectra of poly[Lys(Z)-Glu(OMe)-Lac] are not observable below 200 nm because of the intense absorption by the ϵ-benzyloxycarbonyl group.

The CD spectra observed for the polydepsipeptides in trifluoroethanol are different from spectra observed for the polydepsipeptides in tetrahydrofuran or from spectra observed for helical polypeptides.[8] The spectrum of poly[Glu(OMe)] in hexafluoro-2-propanol has an intense, positive absorption near 197 nm which arises from the $\pi\pi^*$ exciton of the helix and an $n\pi^*$ intensity similar to that observed for many other polypeptides.[9] The largest $\pi\pi^*$ dichroism observed for poly[$(Glu(OMe))_2$-Lac], however, is −5500 (deg cm^2)/dmol while the $n\pi^*$ intensity is half that observed for this polydepsipeptide in tetrahydrofuran. The $n\pi^*$ intensity of poly[Lys(Z)-Glu(OMe)-Lac] in trifluoroethanol is also reduced but it is not possible to observe the $\pi\pi^*$ exciton. Similar results have been reported for poly[L-leucyl-L-leucyl-(*S*)-lactic acid] (poly[$(Leu)_2$-Lac]) and for poly[L-alanyl-L-alanyl-L-alanyl-(*S*)-lactic acid] (poly[$(Ala)_3$-Lac]).[6]

Several different effects may account for the observed spectral differences between the polydepsipeptides in the different solvents or between the polydepsipeptides and polypeptides. In part, the different line shapes arise because of the presence of ester chromophores in the main chain, as described above. In addition, the total number of helical subunits in trifluoroethanol may be less than in tetrahydrofuran. This would imply a shorter helical persistence length and larger contributions from disordered forms in trifluoroethanol. It is also known that the line shape and intensity of helical CD spectra are strongly dependent upon the exact conformation of the helix.[10,11] The observed differences in the spectra could result from different helical conformations. It has previously been shown that the conformation of poly[$(Ala)_2$-Lac], for example, differs from that observed for poly(Ala).[5] Consequently, differences in the type of helix formed, the number of residues in the helical segments, or both could account for the observed differences in the spectra of the various polypeptides.

Another possible interpretation of the weakness of the observed $\pi\pi^*$ dichroism is that the $\pi\pi^*$ transition of the amides might overlap with a negative transition at lower wavelengths. The resultant spectrum would appear as a positive band of reduced intensity. Although other amide transitions such as the $n\sigma^*$ do occur below 180 nm, they do not strongly overlap with the $\pi\pi^*$—as evidenced by the intense positive dichroism of poly(Ala) in trifluoroethanol.[12] The remaining transition near that of the amide $\pi\pi^*$ arises from the ester $\pi\pi^*$ absorption at 170 nm.[13] If this transition were either a single, negative transition or a negative lobe of an ester $\pi\pi^*$ transition polarized parallel to the helix, the resulting weak, positive amide $\pi\pi^*$ could be observed.

The extent to which each polymer is helical may be monitored by determining the intensity of the $n\pi^*$ transition as a function of temperature since the $n\pi^*$ intensity is known to vary linearly with the number of helical segments in polypeptides.[8] The limiting dichroism in tetrahydrofuran for this transition, measured at 222 nm at 0 °C, is −11 200 (deg cm^2)/dmol for poly[Ala-Glu(OBzl)-Lac] and −13 600 (deg cm^2)/dmol for poly[$(Glu(OMe))_2$-Lac]. By comparison, the limiting dichroism for poly[$(Ala)_2$-Lac] was observed to be −9500 (deg cm^2)/dmol. As the temperature is raised to nearly the boiling point of the solvent, the dichroism for the polydepsipeptides with polar side chains decreases to −3300 (deg cm^2)/dmol for poly[Ala-Glu(OBzl)-Lac] and −5000 (deg cm^2)/dmol for poly[$(Glu(OMe))_2$-Lac]. Values for the intensity of the $n\pi^*$ transition which we have observed for other disordered polydepsipeptides are between −1500 and 0 (deg cm^2)/dmol. Poly[$(Ala)_2$-Lac] in tetrahydrofuran at high temperatures, for example, exhibits a dichroism of only −1500 (deg cm^2)/dmol. Further heating of the solutions does not cause any significant change in the dichroism, so these relatively large negative dichroisms represent the end points of the transitions.

The helix-to-coil transitions of the polydepsipeptides in tetrahydrofuran and trifluoroethanol may be quantitatively assessed by means of a nearest-neighbor Ising model which takes into account the sequence of amides and esters present.[3-5] The observed fraction of helical residues in the chain (the helicity) is fit to a theoretical approximation of this fraction. Experimentally, the helicity is defined as

$$\theta_h = \frac{[\theta(T)] - [\theta_c]}{[\theta_h] - [\theta_c]} \quad (1)$$

where $[\theta(T)]$ is the molar ellipticity of the $n\pi^*$ transition as a function of temperature, $[\theta_h]$ is the limiting dichroism for the helical state, and $[\theta_c]$ is the limiting dichroism of the coil. The limiting values for the helical states of the polydepsipeptides studied are those reported in Table I.

The thermodynamics of the helix-to-coil transition are determined by fitting the experimentally observed helicities to a theoretical expression. The expression for the helicity is[3]

$$\theta_h = \frac{s(s + c + 2\sigma - 1)}{c(s + c + 1)} \quad (2)$$

$$c = [(1 - s)^2 + 4s\sigma]^{1/2} \quad (3)$$

where s and σ are the Zimm–Bragg parameters. To fit the data, σ is assumed to be temperature independent and is allowed to vary. Values for s are then determined by interval-halving. The allowed error in θ_h during interval-halfing is 10^{-5}. A similar technique has been extensively employed by Scheraga and co-workers.[14]

The accuracy with which individual choices of σ are able to reproduce the experimental data is not the only available measure of the agreement between calculated and observed values. If the error between the observed and calculated helicities does not exhibit a well-defined min-

Table I
CD Characteristics of Sequential Polydepsipeptides Having Protected Polar Side Chains

polymer	solvent	$n\pi^*$ (θ_h; θ_c)[a]	$\pi\pi^*$ (θ_h)[a]
poly[Ala-Glu(OBzl)-Lac]	tetrahydrofuran	218 (−11 200; −3300)	204 (−15 300)
poly[(Glu(OMe))$_2$-Lac]	tetrahydrofuran	218 (−13 600; −5000)	204 (−19 100)
poly[(Glu(OMe))$_2$-Lac]	trifluoroethanol	214 (−5400; +300)	201 (−8800), 189 (+5500)
poly[Lys(Z)-Glu(OMe)-Lac]	trifluoroethanol	214 (−6900; 0)	201 (−10 300), [b]

[a] λ in nm; θ_h and θ_c, in (deg cm^2)/dmol, are the mean residue ellipticities of the helix and the coil, respectively. [b] The spectrum of poly[Lys(Z)-Glu(OMe)-Lac] is inaccessible below 192 nm due to the strong absorption of the phenyl side chain chromophore.

Table II
Thermodynamics of the Helix-Coil Transition of Various Polydepsipeptides[a]

polymer	solvent	$\sigma \times 10^4$	ΔH_t°	ΔS_t°	T_d°
poly[(Glu(OMe))$_2$-Lac]	trifluoroethanol	15.9 ± 2.3	1399 ± 58	3.01 ± 0.31	35.4 ± 0.4
poly[Lys(Z)-Glu(OMe)-Lac]	trifluoroethanol	9.9 ± 2.7	1751 ± 559	3.97 ± 0.95	36.9 ± 0.4
poly[(Glu(OMe))$_2$-Lac]	tetrahydrofuran	7.0 ± 2.7	1349 ± 199	2.89 ± 0.56	37.0 ± 0.5
poly[Ala-Glu(OBzl)-Lac]	tetrahydrofuran	9.9 ± 2.5	1211 ± 106	2.64 ± 0.28	33.2 ± 0.3
poly[(Ala)$_2$-Lac][b]	tetrahydrofuran	50.0 ± 0.5	768 ± 20	1.67 ± 0.10	34.5 ± 0.3
poly[(Ala)$_2$-Lac][b]	chloroform	30.5 ± 0.5	876 ± 30	1.81 ± 0.05	47.5 ± 0.3

[a] ΔH_t° is the enthalpy melting of the parent polypeptides, in cal/mol of residue. ΔS is the average entropy per residue, in eu/residue. The enthalpy is that of a hydrogen-bonded residue in the polydepsipeptide helix, corresponding to $^2/_3$ of the slope of a van't Hoff plot. T_d° is the temperature of melting of the polydepsipeptide in °C. [b] Poly[(L-Ala)$_2$-(*S*)-Lac].[4]

imum as a function of σ, it would not be possible to determine the thermodynamics of melting. If the error depends only weakly on the choice of σ, the reported thermodynamics will contain large errors. It is also possible, however, to measure the slope of the melting curve as a function of temperature, thus adding a second, independent parameter to the determination of σ. This quantity is given by the theory as

$$\left(\frac{\partial\theta_h}{\partial T}\right)_{s=1} = \frac{\Delta H_t^\circ}{2RT_d^{\circ 2}\sigma^{1/2}} \tag{4}$$

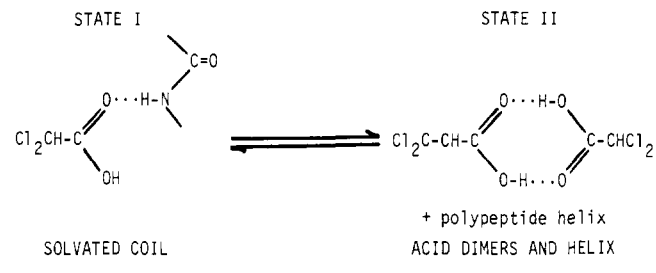
where ΔH_t° is the enthalpy of melting for a hydrogen-bonded residue, T_d° is the temperature at which the polydepsipeptide melts, and $s = 1$ implies 50% helix. The predicted and observed slopes and helicities are employed to optimize σ. Errors in the observed temperature and the $n\pi^*$ ellipticity are included in the analysis.

The results for the polydepsipeptides reported in this paper are summarized in Table II. For comparison, the results for poly[(Ala)$_2$-Lac] are also given. Apparently, all these molecules melt at approximately the same temperature (33–38 °C) in tetrahydrofuran. Introduction of polar glutamate ester or lysine side chains does not influence the melting of these polymers to any appreciable extent in this solvent. In trifluoroethanol, however, poly[(Ala)$_2$-Lac] remains disordered to the freezing point of the solvent, while the polydepsipeptides which contain polar side chains also melt near 36 °C. This difference may be partially accounted for by the nature of the side chains. It has been previously reported that poly[(Leu)$_2$-Lac] undergoes a thermal helix-to-coil transition in trifluoroethanol which is centered at −36 °C.[6] The isobutyl side chain apparently enhances the stability of the polymer to denaturation. Introduction of single glutamic acid residue, however, has a more pronounced effect. Whether this results from the increase in bulk of the side chain or from specific side-chain–solvent interactions is uncertain at this time.

As shown in Table II the enthalpy and entropy for the transition in polydepsipeptides containing polar side chains is 1.5–2 times that found for poly[(Ala)$_2$-Lac]. Since the enthalpy and entropy both increase, the melting temperature remains essentially constant. Another difference between the two materials is in the cooperativity σ. This is 3–5 times smaller in the polydepsipeptides with polar side chains than for alanine. Such differences could arise from a chain length dependence, but similar values of σ for poly[Ala-Glu(OBzl)-Lac] and poly[(Glu(OMe))$_2$-Lac] are observed, although their molecular weights differ. The σ parameter is in part a measure of the structure of the helix formed and is smaller for helices which are more highly ordered. This in turn implies that the helices of the polydepsipeptides which contain polar side chains are more ordered than those exclusively containing alanine residues. Such an observation is also in keeping with the relative magnitudes of ΔS and ΔH.

The differences between the structure and stability of alanine and glutamic acid helices have been previously noted by other workers.[15,16] Conformational transitions have been noted for poly[(Glu(OBzl)] and poly(Ala) in dichloracetic acid–dichloroethane (DCA–DCE) solutions. Under these conditions these polymers are observed to exhibit spectra typical for disordered polypeptides at low temperature and exhibit spectra of helical polypeptides as the temperature is raised. Poly[Glu(OMe)] undergoes this transition at lower temperature than poly(Ala) and has therefore been said to form a less stable helix than poly(Ala). Yet, in our observations the glutamate esters form substantially more stable helical polydepsipeptides than does alanine in analogous polydepsipeptides. These results are not inconsistent if the nature of the two transitions is properly explained.

Polydepsipeptides in organic solvents undergo a direct helix-to-coil transition with an increase of temperature. The opposite transition is observed for polypeptides in DCA–DCE solutions. A scheme for this transition has been proposed by Fujita and co-workers, based on their studies of β-benzyl aspartic acid containing polymers.[17] This scheme is shown below:



At low temperatures the disordered polypeptide chain is strongly hydrogen bonded to molecules of dichloroacetic acid (state I). Since most conformational transitions in DCA–DCE solutions are carried out with an excess of the acid (60–90% DCA), several different chemical species are present in solution. At a given temperature there exist multiple equilibria between polypeptide bound to acid, acid dimers, solvation of the polypeptide helix by solvent, etc. (state I). As the temperature is raised, this complex set of equilibria shifts and more acid dimers form, causing disassociation of the acid and peptide hydrogen bonds. If the free polypeptide is helical in DCE, a helix begins to form (state II). The transition may be reversible, as is that for the polydepsipeptides and some proteins, but involves chemically different species and several different equilibria. Consequently, the two techniques, while consistent within themselves, may not be comparable.

The observation that poly[Glu(OMe)] undergoes the transition at a lower temperature than poly(Ala) means that the tendency for the acid complex to disassociate and the helix to form is greater for poly[Glu(OMe)]. Our results for poly[$(Glu(OMe))_2$-Lac] as compared to poly-[$(Ala)_2$-Lac] indicate that the glutamate-containing polymer is more stable in a strongly hydrogen-bonding solvent. This is in qualitative agreement with the DCA–DCE studies. The difference between the two sets of results supports the notion that there are two separate effects present in the DCA–DCE solutions while the helical polydepsipeptides simply thermally melt. The two effects of acid disassociation and the tendency to form a helix in DCA–DCE are not easily separated.

Although there is qualitative agreement between the melting of poly[$(Glu(OMe))_2$-Lac] in trifluoroethanol and PMLG in DCA-DCE solutions, the quantitative agreement is poor. The enthalpy of melting has been reported to lie between 400 and 1000 cal/residue, depending on the concentration of DCA used and the exact experimental procedure.[15b,16] The current study indicates that the enthalpy for the glutamate-containing polydepsipeptides lies between 1300 and 1400 cal/residue in organic solvents.

The differences between entropies of melting determined by means of the two techniques are even greater. The acid mixtures suggest entropies which range from 1.37 to 3.41 eu/residue as compared to our results of 1.3 eu/residue. Once again these differences probably arise from the lack of compatability between the two systems used to measure the conformational transitions.

The influence of alanine residues on the helical stability of polypeptides which contain glutamate derivatives has been previously reported by Scheraga et al.[16] These authors reported that the net effect of incorporation of alanine into poly(γ-benyl glutamate) chains was the stabilization of the helix. In the current study, we observed little difference between the stabilities of poly[Ala-Glu-(OBzl)-Lac] and poly[$(Glu(OMe))_2$-Lac] in trifluoroethanol. Two differences, however, exist between the current study and the earlier report. The first, as noted above, is that the earlier research was carried out in DCA–DCE solutions. The second difference lies in the chemical makeup of the polymers. In this work strictly sequence-controlled polymers containing amino and hydroxy acids were employed, while the earlier study used random copolymers of alanine and γ-benzyl glutamate. The relative effects of the experimental approach and the chemical makeup of the polymers studied may account for the different findings. This issue will be resolved as other polydepsipeptides are synthesized and their helical stabilities determined.

Conclusions

The large difference of melting temperatures between the polydepsipeptides having protected polar side chains and the parent polypeptides, together with the apparent lack of side-chain effect on their conformational stability, suggests strongly that the main-chain hydrogen-bonding pattern is indeed among the essential factors affecting the stability of the α helix. While poly(L-Ala)[11] and poly[L-Glu(OMe)][8] are helical in hexafluoro-2-propanol and cannot be denaturated thermally up to the boiling point of the solvent, the polydepsipeptides studied here are in a fully disordered structure in this solvent. This behavior reflects the loss of stability associated with the incorporation of ester residues in the polypeptide chains and the reduction of their hydrogen-bonding ability by a factor of $^1/_3$. This reduction of the number of hydrogen bonds is sufficient to account for the large difference of melting temperatures between polydepsipeptides and polypeptides. The results presented here are then consistent with the previous observations on polydepsipeptides.[1-5] Work is in progress to extend these results to other polydepsipeptides which contain different amino and hydroxy acids or which contain different side-chain configurations in order to obtain further information on their contributions to helix stabilities.

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References and Notes

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