stoichiometry and a given domain packing. Most X-ray studies have used the latter method to estimate domain size.

The only way in which a direct X-ray analysis can yield the domain size at the level of several hundred ångstroms is to assume that a domain structure exists that has a known particle scattering factor. In practice, workers assume a simple shape factor like those of spheres or rods. Only a few extrema will appear in the experimental data. Consequently the difference between sphere and rodlike behavior is not easily differentiated when only the first two extrema are observable. Only spheres have scattering patterns that are independent of the degree of orientation of the domains. Consequently, the ambiguity in the interpretation of small-angle X-ray data can only be resolved by comparison of the assumed shape and packing (guided by an analysis of the experimental extrema) with the stoichiometric volume. In this paper the sphere-like assumption gives better agreement between the domain volume calculated from scattering and the stoichiometric volume known from the synthesis.

From electron microscopy measurements, the idea has been advanced[25] that rodlike domains dominate in solvent cast films of polystyrene–polyisoprene tri- and tetra-star block copolymers. However, it should be mentioned that the synthesis procedure used for these materials may influence the morphology. The initiator used was *n*-butyllithium in benzene, an organolithium compound that reacts quite slowly with styrene in hydrocarbon solvents.[26,27] Thus, the star block copolymers synthesized *via* *n*-butyllithium possess moderately polydisperse end segments as well as chains garnished with low molecular weight (<5000) polystyrene segments; blocks too low in molecular weight to form articulate, discrete domains.[2a] The polydispersity and low molecular weight content of these polystyrene blocks has been verified *via* GPC analysis of polystyrene prepared by *n*-butyllithium. Heterogeneity indices of about 1.3 were observed. Thus, any interpretation advanced on the morphology of block copolymers where some of the end blocks can blend with the center blocks should be considered with caution.

Acknowledgment. This work was supported in part by NSF Grant GP-18587.

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The Influence of Solvent on the Secondary Structures of Poly(L-alanine) and Poly(L-proline)

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Received January 15, 1973

ABSTRACT: The conformational properties of L-alanine chains and both *cis*- and *trans*-L-proline chains were calculated as a function of interaction with solvent and chain length. The solvents considered were water, methanol, ethanol, formic acid, and acetic acid. The hydration shell model was used to compute the macromolecule–solvent interactions. The conformational properties of the molecules *in vacuo* were also computed and used as references. Both the *size* and the *polarity* of the solvent molecule are important factors in dictating chain secondary structure. The conformational properties of L-alanine chains are more dependent upon both chain length and interaction with solvent than the L-proline chains. A very delicate balance between polymer–solvent interactions and formation of intrachain hydrogen bonds dictates whether an L-alanine chain will be extended or α helical. *trans*-Poly(L-proline) has a greater stabilizing interaction with polar solvents than *cis*-poly(L-proline). Rotation about the imide bond in both *cis*- and *trans*-L-proline chains is possible, in all solvents, up to $|\omega| = 10°$.

In a previous paper[1] we reported the solvent dependent conformational analysis of some homopolypeptides in an aqueous media. The polymer–solvent interactions were taken into account through a hydration shell.[2] In this report we consider the solvent-dependent behavior of two homopolypeptides, poly(L-alanine) ($(Ala)_n$) and poly(L-proline) ($(Pro)_n$), in five different solvents, and *in vacuo*. Polymer–solvent interactions are again described by the hydration shell model. The solvents are methanol, ethanol, water, formic acid, and acetic acid. This particular set of molecules provides a considerable range of solvent polarity while minimizing the internal conformational degrees of freedom of the solvent molecules which would drastically complicate our calculations. $(Ala)_n$ has been chosen in these calculations because the conformational properties of this polymer, as it precipitates and/or crystallizes from solution, have been studied in considerable detail,[3] and because there have been a number of recent studies[4-7] of the secondary structure of this polymer in a variety of solvents.

Solvent-dependent conformational energy calculations were carried out on $(Pro)_n$ because Krimm and Venkatachalam[8] have investigated, using their polymer–solvent model, the conformational properties of $(Pro)_n$ in aqueous solution. In their model, solvent molecules are explicitly brought into the vicinity of the polymer and the resulting free energy is minimized. In this paper we discuss and compare our solvent-dependent $(Pro)_n$ conformational

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energy calculations to those of Krimm and Venkatachalam. We also report the changes in the secondary structure of $(Pro)_n$ as a function of both ordered chain length and solvent. Differences in the polymer–solvent interaction free energies for $(Ala)_n$ and $(Pro)_n$ are largely due to the presence of the N-H···solvent interaction for $(Ala)_n$ and the absence of this interaction for $(Pro)_n$. Thus we were able to estimate the significance of the N-H···solvent interaction.

Lastly, we have computed the mean-square residue length, $\langle \hat{l}_k^2 \rangle$, as a function of ordered chain length and solvent for $(Ala)_n$ and $(Pro)_n$. These values may be useful in the Flory-type[9] configurational statistics of biopolymer chains.

Theory

Polymer–Solvent Model. In a previous report[1] we described our hydration-shell model used in calculating polymer–solvent interactions in detail. Hence, we will only outline the salient features of the model here. A specific change in the free-energy interaction between a polymer atom (or group) and solvent molecules results from an overlap between the hydration shell centered on the polymer atom (or group) i and the van der Waals volume of some other nonbonded polymer atom j as depicted, in two dimensions, in Figure 1. Initially, the hydration shell contains n solvent molecules when no overlaps occur. As the number of overlaps increase, as a function of polymer conformation, solvent molecules will be ejected from the hydration shell. Each time a solvent molecule is ejected from a hydration shell there is a characteristic change in free energy Δf. The sum of these free-energy changes for all atoms and/or groups in the polymer constitutes the total polymer–solvent interaction energy.

In all, there are five parameters in the hydration-shell model. We have already mentioned two, namely, Δf and n. In addition we must know the radius of the hydration shell, R_V, and the van der Waals radius, r_j, as defined in figure one. Lastly, we need to have some measure of the tightness of packing of the solvent molecules in the hydration shell. This is characterized by a parameter V_f which is the free volume of packing of a solvent molecule in a hydration shell. Of course knowing the value of V_f presupposes we know the volume of the solvent molecule, V_s.

The van der Waals radii are well known and given elsewhere.[10] However, it is up to us to determine the values of n, R_V, V_f, and Δf for a particular atom or group of atoms in a particular solvent. This is accomplished by allowing different numbers of solvent molecules to interact with an isolated atom or group subject to the constraints that would be present when the isolated atom or group is part of a polymer chain. That particular assembly of solvent molecules in that particular configuration about the isolated solute atom or group which minimizes the total interaction free energy yields the necessary values of n, R_V, and V_f. The free energy of interaction between the solvent molecules and the solute species, f_I, is also determined. This interaction energy is computed as the sum of pairwise nonbonded, electrostatic, torsional, and hydrogen bonding terms identical with those used in previous studies.[11,12] f_I is a free energy in the sense that the configurational entropy of the various assemblies of solvent molecules about the solute species is taken into consideration in the calculations. The configurational entropy is computed using the ensemble of states used in the energy minimizations with respect to n, R_V, and V_f. Δf is obtained by subtracting f_I from the free energy of the bulk solvent at T = 298°K, which is obtained experimentally. In Figure 2 we schematically illustrate the geometry involved in the calculation of the values of the n, R_V, and Δf for trigonal nitrogen in water. The cylinders emanating from the van der Waals sphere of the nitrogen take into account steric restrictions which arise when this nitrogen is covalently bonded in a molecule. There are five degrees of spatial freedom which must be considered for the interaction of the solute species with a solvent molecule. These degrees of freedom are defined in Figure 2. The partial charges assigned to the solute species in the electrostatic energy calculations are those found for poly(L-alanine). Obviously, changes in the values of the partial charges, as usually is the case for other macromolecules, would lead to different values for the polymer–solvent parameters. Thus, the values for the polymer–solvent parameters reported here are strictly valid only for poly(L-alanine). Fortunately, the magnitude of the change in the values of the partial charges for corresponding atoms or groups in different molecules is usually sufficiently small so as to result in nearly identical polymer–solvent interactions. Hence, we consider the hydration shell parameters reported here to be universally applicable.

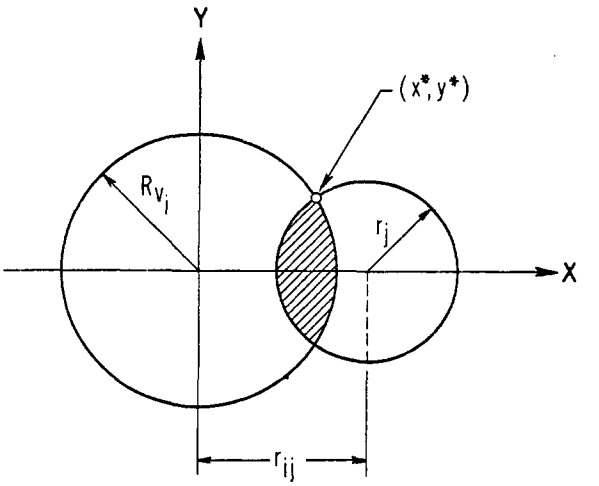

Figure 1. A two-dimensional projection of the geometry of the intersection of the solvation shell of atom i and the van der Waals sphere of atom j.

As reported in our first paper[1] on polymer–solvent interactions, the values of Δf for aqueous solution were taken from the work of Gibson and Scheraga[13] since these were experimental, rather than theoretical values. It was noted that our theoretical Δf values usually differed from the experimental Δf by about 5% and in no case by more than 12%. This reasonably good agreement between the experimental and theoretical values of the Δf encouraged us to theoretically calculate all the hydration-shell parameters for a number of solvents in addition to water. In Table I we report the hydration-shell parameters for water, methanol, ethanol, acetic acid, and formic acid. In the calculation of the hydration-shell parameters for ethanol and acetic acid, we did allow rotation about the CH_3–CH_2 and the CH_3–COO(H) bonds, respectively. The conformational energy of these two solvent molecules were calculated as a function of bond rotation by adopting the

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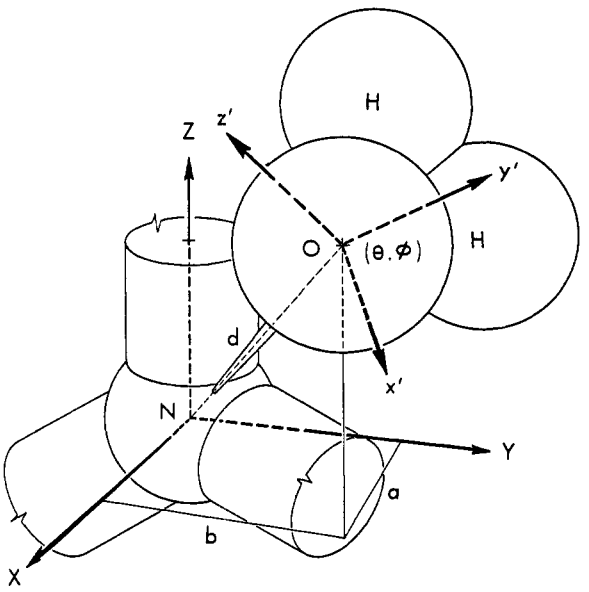
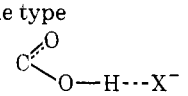


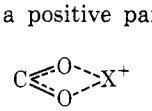
Figure 2. Geometry of an N (sp^2) atom in a molecule used to determine the characteristic polymer-solvent interaction parameters using the hydration-shell model. d is the distance of the water molecule from the N, and a and b define the position of the water molecule in the XY plane of the N cartesian frame. θ and ϕ define the relative orientation of the water molecule in the cartesian frame associated with the water molecule.

same potential functions used in the above-reported solute–solvent species interactions.

For acetic and formic acids a decision had to be made concerning the state of ionization of the solvent molecule when interacting with a solute atom or group. We adopted the convention that the neutral form of the acid interacts with those solute species having a negative partial charge; *i.e.*, interactions of the type



and the charged form of the acid interacts with those solute species possessing a positive partial charge; *i.e.*, interactions of the type



Mechanistically this scheme of interactions would correspond to a solute molecule in a solution of completely ionized acid. The ionized acid molecules would interact with the groups of the solute molecule having positive partial charge. The free protons would interact with the groups of the solute molecules having negative partial charges. In turn, the ionized acid molecules would interact with the proton-negative charge group complexes. This last interaction should be similar to the neutral form of the acid molecule interacting with solute groups having negative partial charges. In Table I we distinguish which form of the acid was used to compute the hydration-shell parameters for each solute species. The values of V_s for each of the solvent molecules were determined from Corey–Pauling–Koltrin (CPK) molecular models.

Calculation of Conformational Energies. The nonbonded, electrostatic, torsional and hydrogen bonding contributions to the conformational energy were calculated in addition to the polymer-solvent interaction free energy. The potential functions are the same as those described above for the solvent-solute interactions. However, in these calculations we insisted that the ΔH of breaking the hydrogen bonds of a poly(L-alanine) α helix in water be 1.5 kcal/mol.[14,15] One additional consideration was included in the proline calculations. Rotation about the imide bond, denoted by ω, was allowed. The torsional potential about this partial double bond is described in ref 16, where A = 20 kcal/mol and ω = 180° corresponds to the trans configuration.

Calculation of the Average Ordered Length $\langle l_k^2 \rangle$ of a k-omer. If k is the number of residues which compose the polypeptide chain, then the mean-square axial length of an ordered chain segment is

$$\langle l_k^2 \rangle = \sum_{i=1}^{N} (k\hat{l}_i')^2 P_i^{(k)} \quad (1)$$

$$\langle l_k^2 \rangle = k^2 \sum_{i=1}^{N} \langle \hat{l}_i'^2 \rangle P_i^{(k)} \quad (2)$$

$$\langle l_k^2 \rangle = k^2 \langle \hat{l}_k^2 \rangle \quad (3)$$

where N is the number of conformational states considered in the averaging process, and $\hat{l}'_i$ is the axial length, relative to the helix generated by conformation i, of a residue unit. In the calculations reported here, end effects, owing to carboxyl and amine groups, are not considered. In other words, we deal here with a chain segment located in the "middle" of a long-chain molecule. $P_i^{(k)}$ is the probability of observing the ith conformation for a k-omer

$$P_i^{(k)} = \frac{\exp(-E_i^{(k)}/RT)}{\sum_{j=1}^{N} \exp(-E_j^{(k)}/RT)} \quad (4)$$

where $E_j^{(k)}$ is the total conformational energy of state j. $E_j^{(k)}$ can be expressed in terms of the sum of the pairwise residue–residue neighbor interactions.

$$E_j^{(k)} = kE_j(0) + (k-1)E_j(1) + (k-2)E_j(2) + \cdots + E_j(k-1) \quad (5)$$

$$E_j^{(k)} = \sum_{i=0}^{k} (k-i)E_j(i) \quad (6)$$

where $E_j(i)$ is the interaction energy between ith nearest-neighbor residues each of which are in the jth conformational state. Only the first six nearest-neighbor residue–residue interactions were considered in our calculations

$$E_j^{(k)} \approx \sum_{i=0}^{6} (k-i)E_j(i) \quad \text{for } k > 6 \quad (7)$$

Method

Conformational energy maps of linked L-alanine and linked L-proline planar units of varying lengths were constructed using 20° increments in the values of the angles. The interrelationship between conformation and minimum energy is probably not meaningful to any greater degree than given here due to the many inherent assumptions in the potential functions. Thus we did not locate relative minima precisely. A set of maps was constructed for each solvent and for *in vacuo* studies. Potential energy surfaces within 5 kcal/mol per residue of the global minimum on each map were recorded. Equations 6 and 7 were used to compute the energies. The length of the chains varied from 2 residue units up to 50 residue units which was chosen to be equivalent to the polymer. The value of χ_1, rotation of the methyl side-chain group about the C^α–C^β bond, for L-alanine residues was chosen so as to minimize the total conformational energy for each choice

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Table I
Polymer–Solvent Parameters for the Hydration–Shell Model for a Variety of Solvents

Atom or Group	n	Δf (kcal/mol)	R_V (Å)	V_f (Å^3)	Mol Species
		a. Formic Acid[a]			
N (sp^2)	2	1.90	5.80	24.40	b
C (sp^2)	2	2.85	5.25	16.50	a
O (carbonyl)	2	3.40	5.40	43.80	b
H (amide)	1	5.95	4.30	22.70	a
CH_3 (aliphatic)	6	−0.09	5.95	30.75	a
CH_2 (aliphatic)	4	−0.08	5.95	28.60	a
CH (aliphatic)	1	0.06	5.75	15.75	a
CH (aromatic)	2	0.08	4.45	19.00	a
O (hydroxyl)	1	2.55	4.85	40.60	b
H (hydroxyl)	1	4.75	4.30	43.95	a
O^- (carboxyl)	3	5.05	5.85	29.75	b
O (carboxyl)	1	3.55	5.20	18.50	b
H (carboxyl)	1	3.55	5.20	18.50	b
		b. Acetic Acid[a]			
N (sp^2)	2	1.95	6.60	48.75	b
C (sp^2)	2	2.85	5.60	39.80	a
O (carbonyl)	2	3.35	5.80	96.50	b
H (amide)	1	5.85	4.90	47.35	a
CH_3 (aliphatic)	6	−0.08	6.20	31.60	a
CH_2 (aliphatic)	4	−0.08	6.20	35.83	a
CH (aliphatic)	1	0.06	5.95	18.98	a
CH (aromatic)	2	0.08	4.65	22.07	a
O (hydroxyl)	1	2.55	5.80	48.60	b
H (hydroxyl)	1	4.55	4.90	67.40	a
O^- (carboxyl)	3	4.96	6.60	49.50	b
O (carboxyl)	1	3.45	6.10	33.70	b
H (carboxyl)	1	3.45	6.10	33.70	b
		c. Methanol[a]			
N (sp^2)	2	0.23	4.70	41.6	
C (sp^2)	2	0.18	5.20	53.6	
O (carbonyl)	2	1.45	4.80	43.9	
H (amide)	1	0.30	4.10	30.5	
CH_3 (aliphatic)	4	0.38	5.90	19.5	
CH_2 (aliphatic)	3	0.32	5.70	21.6	
CH (aliphatic)	2	0.32	5.70	21.6	
CH (aromatic)	2	0.46	4.70	28.5	
O (hydroxyl)	1	0.85	3.65	38.8	
H (hydroxyl)	1	0.85	3.65	38.8	
O^- (carboxyl)	2	2.80	4.80	48.5	
O (carboxyl)	1	1.30	4.35	28.6	
H (carboxyl)	1	1.15	4.35	29.3	
		d. Ethanol[a]			
N (sp^2)	2	0.18	6.20	49.8	
C (sp^2)	2	0.15	5.80	57.5	
O (carbonyl)	2	1.18	6.20	49.1	
H (amide)	1	0.28	5.50	45.6	
CH_3 (aliphatic)	4	0.41	7.10	31.3	
CH_2 (aliphatic)	3	0.39	7.00	31.8	
CH (aliphatic)	2	0.39	6.90	33.0	
CH (aromatic)	2	0.40	6.35	42.9	
O (hydroxyl)	1	0.57	4.15	51.6	
H (hydroxyl)	1	0.57	4.20	51.6	
O^- (carboxyl)	2	2.45	6.35	60.7	
O (carboxyl)	1	1.10	5.90	43.5	
H (carboxyl)	1	0.88	5.90	43.5	
		e. Water[a]			
N (sp^2)	2	0.63	4.33	35.8	
C (sp^2)	2	0.63	3.90	14.3	
O (carbonyl)	2	1.88	3.94	67.6	
H (amide)	2	0.31	3.54	31.3	
CH_3 (aliphatic)	8	−0.13	5.50	41.8	
CH_2 (aliphatic)	4	−0.10	5.50	60.8	
CH (aliphatic)	2	−0.13	5.50	104.8	
CH (aromatic)	3	0.11	3.90	3.3	

Table I *(Continued)*

Atom or Group	n	Δf (kcal/mol)	R_V (Å)	V_f (Å^3)	Mol Species
		e. Water[a]			
O (hydroxyl)	2	1.58	3.94	55.2	
H (hydroxyl)	2	0.31	3.54	54.7	
O$^-$ (carboxyl)	4	4.20	4.10	42.5	
O (carboxyl)	2	4.20	4.10	64.1	
H (carboxyl)	2	0.31	3.54	54.7	

[a] For section a, V_s = 33.8 Å^3 and the molecular species are a = $HCOO^-$ and b = HCOOH; for section b, V_s = 51.6 Å^3 and the molecular species are a = H_3CCOO^- and b = H_3CCOOH; for section c, V_s = 42.8 Å^3 and the molecular species is CH_3OH; for section d, V_s = 63.7 Å^3 and the molecular species is CH_3CH_2OH; for section e, V_s = 21.2 Å^3 and the molecular species is H_2O.

of ϕ and ψ. The Davidon technique[17] was employed in the energy minimization. The geometry of the L-alanine residue was that suggested by Pauling and Corey[18] while the geometry of the L-proline residue was that found by Sasisekharan.[19]

The conformational energy maps of alanine are reported in digitalized form in order to emphasize the large variations in total conformational energy with choice of solvent. The proline conformational maps are reported in the same format as those of Krimm and Venkatachalam[8] so as to allow direct comparison. Rotations about the imide bonds in the L-proline chains were subject to the equivalence condition and occurred in 10° increments in ω. The values of ϕ in the L-proline chains were fixed at that value which yielded the deepest global minimum as a function of ψ and ω for each choice of solvent.

Results

Alanine. In Figure 3 are shown the digitalized conformational maps for two planar-peptide units of L-alanine and for a long ordered chain of L-alanine planar-peptide units (lc-Ala). The dipeptide calculations correspond to the least-ordered chain, *i.e.*, it is a model for the "random coil" of L-alanine (rc-Ala)[20] while the long chain corresponds to the completely ordered polymer. Thus these two maps, for each choice of solvent, should describe the conformational properties of the least- and most-ordered forms of poly(L-alanine). For the sake of space, the other conformational energy maps are not presented in this paper. However, in Figure 4 we report the relationship between the mean-square residue length, $\langle l_k^2 \rangle$, and k, the number of residues in an ordered chain segment, for the five solvents, and *in vacuo* studies. These curves reflect the conformational properties of L-alanine chains as a function of both solvent and ordered chain length. We will discuss Figure 4 shortly, but now turn our attention to studying the conformational energy maps of rc-Ala and lc-Ala.

The rc-Ala maps indicate that the most probable conformations of the dipeptide unit of L-alanine is either β or left-handed 3_1 helical in all solvents. *In vacuo*, as noted in other studies,[21] the 2_1 helix (ϕ = −80°, ψ = 100°) is the most probable structure. An interesting observation, which persists for longer oligomeric chains, is that the larger solvent molecules, ethanol and acetic acid, promote the formation of the left-handed 3_1 helical structure while water, methanol, and formic acid, less bulky solvent molecules, aid in the formation of β conformations. In this regard, solvent polarity has little effect. These findings would suggest that a polymer of rc-Ala could be rather unordered, but yield CD–ORD spectra indicative of a β or left-handed 3_1 helical structure. We are presently investigating the effect of solvent on the CD spectra of unordered polypeptide chains.

The overall topology of the conformational energy surfaces of the dipeptides of L-alanine are very similar in all solvents. There is a significantly greater degree of conformational freedom in these maps as compared to those for lc-Ala. The interactions between polymer and solvent have a large stabilizing effect for acetic acid and formic acid as would be expected. Water, which is relatively polar when compared to methanol and ethanol, surprisingly has approximately the same stabilization polymer–solvent interaction energy with the dipeptide of L-alanine as these two less polar solvents. L-Alanine oligomers have more favorable interactions with aqueous solution than with methanol and ethanol as the length of the chain increases.

The energy maps of lc-Ala in methanol and ethanol are nearly identical. The right-handed α helix, located near ϕ = −60°, ψ = −40°, is the preferred conformation for both solvents. Relative minima at ϕ = −160°, ψ = 100°, and ϕ = −80°, ψ = 140°, on both maps, are indicative of the possible existence of isolated stable β and left-handed 3_1 helical conformations, respectively. For ethanol, in fact, the right-handed α helix and the left-handed 3_1 helix are approximately equally stable.

The conformational energy map of lc-Ala *in vacuo* contains more significant variations in energy than the methanol and ethanol maps. This suggests that lc-Ala *in vacuo* can adopt fewer conformational states than in methanol and ethanol. This map indicates less flexibility of the biopolymer than computed by other workers.[21,22] The reason for this is due to an increase in the stabilization energy given to the hydrogen bond. In these calculations we insisted, as already mentioned, that the ΔH of breaking the hydrogen bonds of the α helix in water be 1.5 kcal/mol. This required a pairwise intrachain hydrogen-bonding energy of −4.6 kcal/mol as compared to the −3.5 kcal/mol normally used. These calculations indicate that in this case lc-Ala *in vacuo* would prefer the right-handed α-helical conformation.

lc-Ala in aqueous solution also is most stable as a right-handed α helix. However, the conformational flexibility of the polymer is much enhanced over what it was *in vacuo*. There are relative minima located at points on the map corresponding to β and left-handed 3_1 helical structures just as for methanol and ethanol.

The conformational map of lc-Ala in acetic acid indi-

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Figure 3. Digitalized conformational energy maps for dipeptide L-alanine and for (L-alanine)$_{50}$ which is chosen as a model for the polymer in five solvents and *in vacuo*. ϕ is plotted along the abscissa and ψ along the ordinate. The total conformational energy was minimized with respect to χ_1 at each point. The energies left blank are greater than 5 kcal/mole per residue above the minimum. (Figure continues for six pages.)

Part a. The Lowest Energy Is −9.8 kcal/Mole per Dipeptide, Located at (−160,140), for the Dialanine Chain in Methanol

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180	−7.5	−9.0	−8.7	−8.0	−7.9	−7.9	−6.6						−5.6	−1.8				−2.7	−7.5
160	−8.0	−9.4	−8.8	−8.3	−8.3	−8.3	−8.0	−0.5					−1.5	−1.8				−3.1	−8.0
140	−8.4	−9.8	−9.1	−8.4	−8.4	−8.4	−8.3	−6.9						−1.3				−3.2	−8.4
120	−8.0	−9.5	−8.7	−8.1	−8.3	−8.5	−8.4	−7.5	−0.3					−0.7				−2.7	−8.0
100	−7.6	−9.1	−8.4	−7.9	−8.4	−8.9	−8.0	−5.5	−1.2				−1.7	−0.6				−2.3	−7.6
80	−7.3	−8.8	−8.1	−7.7	−8.5	−7.9						−3.1	−4.9	−0.5				−2.0	−7.3
60	−7.1	−8.5	−7.9	−7.6	−8.5	−5.3					−1.1	−6.6	−5.6	−0.6				−1.9	−7.1
40	−6.9	−8.4	−7.7	−7.4	−8.1	−6.0						−7.2	−6.2	−0.9				−1.5	−6.9
20	−4.5	−7.4	−7.2	−6.8	−7.3	−7.2							−6.5	−1.0					−4.5
0		−3.4	−6.1	−6.2	−6.6	−7.1	−4.3						−3.8	−1.4					
−20	−5.0	−5.2	−6.2	−6.6	−7.0	−7.3	−7.6							−2.0				−1.6	−5.0
−40	−7.8	−8.9	−8.0	−7.4	−7.6	−7.7	−7.8	−7.4						−1.3				−2.9	−7.8
−60	−8.0	−9.1	−8.1	−7.3	−7.4	−7.4	−7.3	−6.8	−1.7					−0.6				−3.1	−8.0
−80	−7.5	−8.5	−7.5	−6.7	−6.7	−6.7	−5.8	−2.7						−2.4				−2.6	−7.5
−100	−6.5	−7.5	−6.5	−5.6	−5.6	−5.4	−1.4					−4.9	−6.1	−1.9				−1.6	−6.5
−120	−5.9	−6.9	−5.8	−4.9	−5.0	−4.6						−5.9	−5.4	−0.4				−1.0	−5.9
−140	−6.8	−7.9	−6.8	−5.9	−6.0	−5.7						−5.6	−5.7	−0.8				−1.8	−6.8
−160	−7.3	−8.7	−8.1	−7.2	−7.2	−7.1	−1.7					0.0	−6.2	−1.5				−2.2	−7.3
−180	−7.5	−9.0	−8.7	−8.0	−7.9	−7.9	−6.6						−5.6	−1.8				−2.7	−7.5

Part b. The Lowest Energy Is 2.6 kcal/Mole per Residue, Located at (−60, −40), for the Infinite Alanine Chain in Methanol

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180	6.6	5.8	4.8	5.4	5.3	6.6													6.6
160	5.9	4.4	4.7	5.1	4.3	4.7	5.9												5.9
140	4.4	3.8	4.3	5.0	4.2	3.6	4.5	6.0										7.0	4.4
120	4.6	4.0	4.3	4.9	4.3	4.7	5.4	4.9											4.6
100	6.0	3.5	3.9	4.3	4.8	5.6	5.9												6.0
80	5.1	5.4	5.8	4.1	5.0	6.5													5.1
60	6.0	5.0	6.0	7.8	7.2							4.6							6.0
40	6.2	5.1	5.2	5.2	5.9								3.8						6.2
20			6.1	6.3	5.9	5.5							4.1						
0					7.1	5.2	7.1						7.4						
−20					6.9	5.0	3.4												
−40	5.2	5.0	5.7				2.6											7.5	5.2
−60	5.1	4.9	5.4	6.1				4.2										7.5	5.1
−80	5.0	4.7																	5.0
−100													7.2						
−120												6.7	7.7						
−140													6.4						
−160		6.1	5.4	6.1	6.1	6.2							6.9						
−180	6.6	5.8	4.8	5.4	5.3	6.6													6.6

Part c. The Lowest Energy Is −11.0 kcal/Mole per Dipeptide, Located at (−80,120), for the Dialanine Chain in Ethanol

180	−7.3	−8.7	−8.4	−8.2	−9.0	−9.8	−7.4						−6.4	−3.5				−2.4	−7.3
160	−7.7	−8.9	−8.6	−8.4	−9.2	−9.8	−9.1	−1.7					−2.1	−2.5				−2.8	−7.7
140	−7.8	−9.0	−8.5	−8.4	−9.2	−9.8	−10.5	−8.0						−1.8				−2.7	−7.8
120	−7.9	−9.0	−8.7	−8.7	−9.7	−11.0	−10.3	−9.1	−2.3					−1.6				−2.7	−7.9
100	−7.9	−9.3	−8.9	−9.0	−10.2	−10.7	−10.1	−7.8	−3.3				−2.7	−1.9				−3.1	−7.9
80	−8.3	−9.8	−9.4	−9.6	−10.6	−10.2				−1.8	−5.2	−6.4	−2.2					−3.0	−8.3
60	−7.8	−9.4	−9.0	−9.3	−10.2	−7.4				−2.6	−8.3	−7.3	−2.7					−2.5	−7.8
40	−7.5	−9.0	−8.6	−9.0	−9.9	−7.9					−8.6	−8.7	−2.6					−2.1	−7.5
20	−5.1	−8.1	−8.1	−8.2	−9.0	−9.1					−1.7	−8.0	−2.9						−5.1
0		−3.9	−6.8	−7.3	−8.4	−9.1	−6.3					−5.2	−2.9						
−20	−5.1	−5.3	−6.4	−7.3	−8.3	−9.2	−9.1	−1.8					−3.0					−1.7	−5.1
−40	−8.0	−9.0	−8.4	−8.2	−8.9	−9.5	−10.3	−9.1					−2.3					−3.2	−8.0
−60	−8.4	−9.5	−8.8	−8.4	−9.0	−9.7	−9.0	−8.9	−3.7				−1.9					−3.6	−8.4
−80	−8.3	−9.4	−8.7	−8.3	−9.0	−8.7	−7.6	−5.2	−2.2				−4.1					−3.5	−8.3
−100	−6.8	−8.3	−7.5	−7.2	−7.8	−7.2	−2.8			−1.8	−6.9	−8.2	−3.5					−1.9	−6.8
−120	−5.9	−7.1	−6.4	−6.4	−7.1	−6.1					−7.6	−7.7	−2.9					−1.0	−5.9
−140	−6.7	−7.8	−7.2	−6.9	−8.1	−7.1					−7.2	−8.6	−2.6					−1.7	−6.7
−160	−7.1	−8.5	−8.0	−7.8	−8.5	−8.3	−2.6				−1.6	−7.6	−3.1					−2.1	−7.1
−180	−7.3	−8.7	−8.4	−8.2	−9.0	−9.8	−7.4					−6.4	−3.5					−2.4	−7.3
	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180

Part d. The Lowest Energy Is 3.3 kcal/Mole per Residue, Located at (−80,140) and (−60,−40), for the Infinite Alanine Chain in Ethanol

180	7.1	6.3	5.4	5.6	4.8	5.7													7.1
160	6.6	5.2	5.3	5.5	4.0	4.2	6.3												6.6
140	5.4	5.0	5.4	5.7	4.2	3.3	4.1	6.9										7.9	5.4
120	5.2	4.6	5.0	5.2	3.9	3.5	5.3	5.1											5.2
100	6.2	3.9	4.2	4.3	4.2	5.0	5.7	7.8										7.5	6.2
80	4.7	5.0	5.3	3.5	4.3	5.5												7.6	4.7
60	6.0	5.0	6.1	7.6	7.1						6.1								6.0
40	6.6	5.5	5.7	5.4	5.8	7.8						4.5							6.6
20		7.0	7.0	7.0	6.2	5.7						4.6							
0				8.3	7.2	5.7	7.6					8.2							
−20					6.8	5.4	4.6												
−40	6.0	5.9	6.7				3.3											8.2	6.0
−60	5.5	5.4	6.1	7.0				5.3										7.8	5.5
−80	4.9	4.7																	4.9
−100												7.1							
−120											7.7	7.6							
−140												5.6	7.5						
−160	7.2	6.4	5.6	5.9	5.3	6.0						7.4	7.9						7.2
−180	7.1	6.3	5.4	5.6	4.8	5.7													7.1
	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180

Figure 3 *(Continued).*

Part e. The Lowest Energy is −2.3 kcal/Mole per Dipeptide, Located at (−80,100), for the Dialanine Chain *in Vacuo*

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180	0.0	−1.0	−1.0	−0.5	−0.8	−0.9	0.4						0.9	4.7				4.9	0.0
160	−0.3	−1.2	−1.2	−0.8	−1.1	−1.4	−1.1	6.3					5.1	5.0				4.6	−0.3
140	−0.6	−1.5	−1.3	−0.9	−1.3	−1.7	−1.7	−0.2						5.6				4.6	−0.6
120	−0.4	−1.5	−1.3	−1.0	−1.5	−2.0	−2.0	−1.2	5.9					6.0				4.8	−0.4
100	−0.2	−1.3	−1.2	−0.9	−1.7	−2.3	−1.8	0.9	5.1				4.6	6.0				5.1	−0.2
80	0.1	−1.0	−0.9	−0.8	−1.8	−1.6					6.7	3.4	1.5	6.0				5.3	0.1
60	0.2	−0.8	−0.7	−0.6	−1.7	1.0					5.3	0.1	0.6	5.8				5.5	0.2
40	0.5	−0.6	−0.5	−0.4	−1.4	0.3						−0.4	0.0	5.5				5.8	0.5
20	2.8	0.3	0.0	0.2	−0.6	−0.9						6.8	−0.3	5.4					2.8
0		4.3	1.1	0.8	0.1	−0.7	1.8						2.4	5.0					
−20	2.4	2.6	1.2	0.5	−0.1	−0.8	−1.2	6.6						4.5				5.9	2.4
−40	−0.2	−0.9	−0.5	−0.2	−0.6	−1.0	−1.4	−1.0						5.3				4.6	−0.2
−60	−0.6	−1.2	−0.8	−0.3	−0.5	−0.8	−0.9	−0.5	4.6					5.8				4.3	−0.6
−80	−0.2	−0.9	−0.4	0.2	0.0	−0.1	0.7	3.4	6.6					3.8				4.7	−0.2
−100	0.8	0.2	0.7	1.3	1.2	1.3	5.1				6.0	1.6	−0.3	4.3				5.7	0.8
−120	1.6	0.9	1.4	2.1	1.9	2.2						0.5	0.4	5.7				6.4	1.6
−140	0.8	0.1	0.6	1.2	1.0	1.2						0.9	0.2	5.4				5.8	0.8
−160	0.1	−0.8	−0.5	0.1	−0.2	−0.2	5.1					6.5	−0.2	4.8				5.1	0.1
−180	0.0	−1.0	−1.0	−0.5	−0.8	−0.9	0.4						0.9	4.7				4.9	0.0

Part f. The Lowest Energy Is −6.3 kcal/Mole per Residue, Located at (−60,−40), for the Infinite Alanine Chain *in Vacuo*

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180					−1.3														
160					−2.2	−1.9													
140		−1.4	−1.3		−2.4	−3.2	−2.3	−1.3											
120		−1.4	−1.5	−1.4	−2.6	−2.4	−1.7	−2.4											
100		−2.0	−2.0	−2.0	−2.0	−1.6	−1.4												
80			−0.2	−2.2	−1.8						−4.1								
60												−5.2							
40				−1.4	−1.4							−4.4							
20					−1.9	−2.4													
0					−1.7	−3.3													
−20					−2.7	−4.1	−5.0												
−40					−2.1		−6.3												
−60						−2.7		−4.7											
−80							−1.5												
−100																			
−120																			
−140																			
−160																			
−180					−1.3														

Part g. The Lowest Energy Is −8.2 kcal/Mole per Dipeptide, Located at (−160,140), for the Dialanine Chain in Water

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180	−6.5	−7.8	−7.8	−7.2	−7.2	−7.0	−5.2						−3.5	−0.2				−1.6	−6.5
160	−6.9	−8.0	−8.0	−7.5	−7.6	−7.4	−6.6	1.1					0.5	0.0				−1.8	−6.9
140	−7.1	−8.2	−8.1	−7.6	−7.8	−7.6	−7.1	−5.2						0.5				−1.8	−7.1
120	−6.9	−8.1	−8.0	−7.5	−7.8	−7.7	−7.2	−5.9	1.4					0.9				−1.5	−6.9
100	−6.6	−7.8	−7.7	−7.3	−7.8	−7.9	−6.8	−3.7	0.8				0.0	1.0				−1.2	−6.6
80	−6.3	−7.5	−7.4	−7.1	−7.8	−7.1						−0.8	−3.0	0.9				−1.0	−6.3
60	−6.1	−7.2	−7.2	−6.9	−7.6	−4.4					1.3	−4.0	−3.8	0.8				−0.8	−6.1
40	−5.9	−7.1	−7.0	−6.6	−7.4	−5.2						−4.4	−4.3	0.6				−0.6	−5.9
20	−3.6	−6.3	−6.5	−6.1	−6.6	−6.4							−4.5	0.6					−3.6
0		−2.3	−5.5	−5.6	−6.1	−6.3	−3.1						−1.8	0.3					
−20	−4.0	−4.0	−5.5	−6.0	−6.4	−6.5	−6.2							−0.2				−0.5	−4.0
−40	−6.7	−7.6	−7.3	−6.8	−6.9	−6.8	−6.5	−5.4						0.6				−1.7	−6.7
−60	−7.0	−7.9	−7.5	−6.8	−6.9	−6.7	−6.2	−5.1	0.6					1.3				−1.9	−7.0
−80	−6.6	−7.5	−7.1	−6.3	−6.4	−6.1	−4.8	−1.6						−0.8				−1.6	−6.6
−100	−5.7	−6.5	−6.0	−5.3	−5.3	−4.8	−0.6					−2.1		−0.3				−0.7	−5.7
−120	−5.0	−5.8	−5.3	−4.5	−4.5	−3.9						−3.2		0.9				0.0	−5.0
−140	−5.8	−6.7	−6.2	−5.4	−5.5	−4.9						−3.0	−4.0	0.5				−0.7	−5.8
−160	−6.4	−7.6	−7.3	−6.6	−6.6	−6.3	−0.7						−4.5	−0.2				−1.4	−6.4
−180	−6.5	−7.8	−7.8	−7.2	−7.2	−7.0	−5.2						−3.5	−0.2				−1.6	−6.5

Part h. The Lowest Energy Is −5.0 kcal/Mole per Residue, Located at (−60,−40), for the Infinite Alanine Chain in Water

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180	−1.1	−1.7	−2.7	−2.2	−2.5	−0.9													−1.1
160	−1.5	−3.0	−3.0	−2.5	−3.5	−2.7	−1.3												−1.5
140	−2.8	−3.4	−3.2	−2.7	−3.7	−4.1	−2.7	−1.0										−0.2	−2.8
120	−2.7	−3.3	−3.3	−2.9	−3.8	−3.2	−2.1	−2.2											−2.7
100	−1.4	−3.7	−3.7	−3.4	−3.1	−2.4	−1.7												−1.4
80	−2.3	−1.8	−1.8	−3.5	−3.0	−1.8													−2.3
60	−1.4	−2.2	−1.5									−2.7							−1.4
40	−1.2	−2.1	−2.4	−2.6	−2.5	−0.6							−4.0						−1.2
20		−1.2	−1.8	−2.1	−2.9	−3.0							−3.8	−0.8					
0				−1.5	−2.6	−3.6	−0.9						−0.6	−0.9					
−20					−2.5	−4.0	−4.3							−0.5					
−40	−2.1	−2.3	−1.8				−5.0												−2.1
−60	−2.4	−2.6	−2.3	−1.8	−1.7			−3.6										0.0	−2.4
−80	−2.8	−3.0	−1.0	−1.4	−1.6														−2.8
−100													−0.4						
−120												−0.1							
−140													−0.8						
−160		−1.7	−2.4	−1.8	−1.9	−1.6													
−180	−1.1	−1.7	−2.7	−2.2	−2.5	−0.9													−1.1

Figure 3 *(Continued).*

Part i. The lowest Energy is −28.0 kcal/Mole per Dipeptide, Located at (−80,120), for the Dialanine Chain in Acetic Acid

180	−22.9	−24.2	−23.9	−23.8	−25.1	−26.9	−23.7						−22.3	−19.7					−22.9
160	−23.0	−24.2	−23.7	−23.7	−24.9	−26.6	−25.8						−18.2	−19.6					−23.0
140	−24.4	−24.8	−24.8	−24.1	−25.4	−27.2	−27.9	−23.8						−18.9				−18.5	−24.4
120	−24.3	−25.1	−24.4	−24.0	−26.4	−28.0	−27.8	−26.7	−18.3					−19.1				−18.7	−24.3
100	−23.9	−24.6	−23.6	−23.4	−25.8	−26.5	−24.6	−21.8					−19.6						−23.9
80	−21.6	−22.4	−22.6	−22.4	−24.8	−23.7						−19.0	−21.7						−21.6
60	−22.5	−23.2	−22.4	−23.6	−24.7	−20.9						−23.1	−25.0	−18.0					−22.5
40	−21.6	−22.7	−23.0	−23.0	−23.9	−21.9						−24.1	−25.2	−19.1					−21.6
20	−18.7	−21.3	−21.2	−21.4	−23.5	−22.6							−23.6	−19.1					−18.7
0			−20.4	−20.8	−23.0	−24.0	−20.1						−20.0	−18.4					
−20	−19.4	−19.2	−20.2	−20.8	−22.5	−25.4	−24.1												−19.4
−40	−22.0	−22.6	−21.8	−21.2	−22.4	−25.2	−25.4	−23.2											−22.0
−60	−23.6	−23.2	−22.2	−21.5	−24.2	−24.5	−25.5	−22.5											−23.6
−80	−21.8	−23.0	−22.3	−21.6	−22.7	−24.3	−22.1	−18.1											−21.8
−100	−22.6	−22.6	−21.8	−22.1	−23.3	−23.0	−18.8					−20.8	−22.4	−18.3					−22.6
−120	−22.0	−23.0	−22.8	−22.1	−23.5	−23.2						−24.0	−24.0	−18.5					−22.0
−140	−22.8	−23.8	−23.0	−23.0	−24.3	−23.9						−21.5	−24.5	−18.0					−22.8
−160	−23.0	−24.4	−24.3	−23.6	−25.6	−26.5	−19.2						−24.1	−19.1					−23.0
−180	−22.9	−24.2	−23.9	−23.8	−25.1	−26.9	−23.7						−22.3	−19.7					−22.9
	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180

Part j. The Lowest Energy is −7.1 kcal/Mole per Residue, Located at (−100,120), for the Infinite Alanine Chain in Acetic Acid

180	−3.0	−3.6	−4.1	−3.2	−4.1	−3.1													−3.0
160	−3.7	−4.8	−4.3	−3.5	−5.0	−4.8													−3.7
140	−5.8	−6.1	−5.3	−4.3	−5.9	−6.8	−4.6											−2.9	−5.8
120	−6.2	−6.6	−6.0	−5.0	−7.1	−6.7	−3.7											−2.5	−6.2
100	−4.5	−6.8	−6.1	−5.4	−5.8	−3.6													−4.5
80	−2.9	−2.4	−3.1	−4.5	−4.0														−2.9
60		−2.7																	
40																			
20																			
0																			
−20																			
−40																			
−60																			
−80	−3.0	−2.9																	−3.0
−100		−2.4	−3.0																
−120																			
−140																			
−160	−2.9	−3.5	−3.8	−3.0	−4.2														−2.9
−180	−3.0	−3.6	−4.1	−3.2	−4.1	−3.1													−3.0
	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180

Part k. The Lowest Energy is −15.4 kcal/Mole per Dipeptide, Located at (−140,100), (−180,−80), (−140,−80), for the Dialanine Chain in Formic Acid

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180	−12.8	−13.0	−13.6	−12.4	−13.2	−13.7	−12.3						−10.6	−6.6				−7.4	−12.8
160	−12.9	−12.9	−13.3	−12.1	−12.8	−13.0	−14.5						−5.8	−7.0				−7.5	−12.9
140	−13.8	−13.7	−13.9	−12.6	−13.2	−13.3	−14.6	−12.7						−7.8				−8.2	−13.8
120	−14.8	−13.6	−13.8	−13.4	−13.1	−13.7	−15.1	−13.9	−5.3					−7.5				−8.9	−14.8
100	−15.0	−15.0	−15.4	−14.0	−14.3	−14.6	−13.7	−10.8	−7.9				−8.9	−7.8				−9.2	−15.0
80	−14.6	−14.6	−14.8	−13.9	−14.0	−13.8						−8.5	−11.6	−7.2				−9.0	−14.6
60	−12.9	−13.0	−14.4	−13.7	−14.1	−10.6					−5.7	−13.6	−13.0	−6.4				−7.2	−12.9
40	−11.2	−11.4	−12.0	−12.4	−11.8	−9.3						−11.9	−8.4	−5.4				−5.4	−11.2
20	−9.3	−10.9	−11.8	−11.2	−12.1	−11.6								−6.7					−9.3
0		−6.8	−10.6	−10.3	−11.9	−12.3	−7.8							−7.0					
−20	−9.6	−8.5	−10.5	−10.4	−10.5	−12.2	−13.1	−9.4						−6.6				−5.7	−9.6
−40	−12.5	−12.2	−12.4	−11.2	−11.0	−12.0	−13.3	−11.9										−7.1	−12.5
−60	−14.6	−14.4	−14.5	−13.2	−13.8	−13.7	−14.3	−14.1	−6.0					−6.4				−9.1	−14.6
−80	−15.4	−15.3	−15.4	−14.0	−13.6	−13.8	−12.3	−8.4						−8.7				−9.7	−15.4
−100	−14.0	−13.9	−14.0	−12.6	−12.5	−13.0	−8.3				−6.1	−8.8	−11.7	−7.4				−8.5	−14.0
−120	−12.9	−12.6	−12.6	−11.3	−11.3	−12.0						−10.7	−12.0	−5.3				−6.2	−12.9
−140	−12.5	−12.6	−13.8	−12.3	−12.3	−13.2						−9.6	−11.6					−6.8	−12.5
−160	−12.4	−12.6	−13.0	−12.8	−12.6	−13.4	−6.8						−12.1	−5.4				−6.7	−12.4
−180	−12.8	−13.0	−13.6	−12.4	−13.2	−13.7	−12.3						−10.6	−6.6				−7.4	−12.8

Part l. The Lowest Energy is −5.0 kcal/Mole per Residue, Located at (−60,−40), for the Infinite Alanine Chain in Formic Acid

	−180	−160	−140	−120	−100	−80	−60	−40	−20	0	20	40	60	80	100	120	140	160	180
180			−0.7	−0.6	−1.1														
160		−0.6	−0.8	−0.7	−1.9	−1.7													−1.0
140	−1.0	−1.0	−1.0	−0.8	−2.0	−2.9	−1.4	−0.5											−2.1
120	−2.1	−1.2	−1.3	−1.1	−2.2	−2.1	−0.5	−1.3											−0.2
100	−0.2	−2.1	−1.8	−1.1	−0.8	−0.6	−0.4												−0.7
80	−0.7			−1.0	−0.8							−1.3							
60		−0.3											−4.4						
40		−0.3	−0.3	−1.2	−1.1								−3.1	−0.4					
20					−1.5	−2.4							−0.6						
0					−1.1	−2.7	−0.9												
−20					−1.8	−3.5	−4.6												−0.7
−40	−0.7	−0.3					−5.0												0.0
−60	0.0							−1.6											
−80																			
−100																			
−120													−0.4						
−140																			
−160			−0.5	−0.2	−0.4	−0.3													
−180			−0.7	−0.6	−1.1														

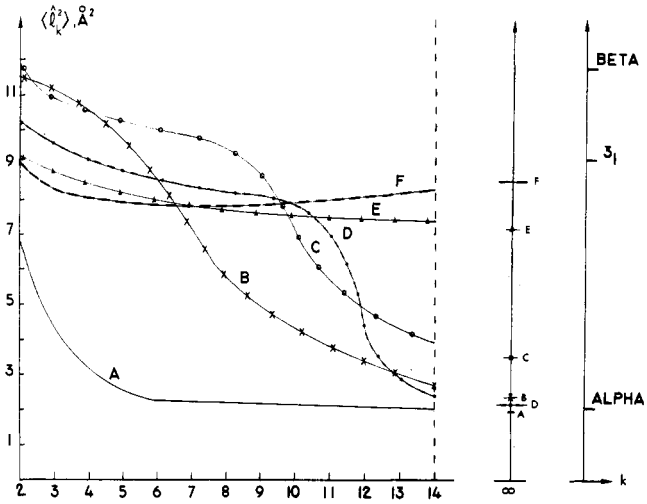

Figure 4. Plots of $\langle \hat{l}_k^2 \rangle$ *vs.* k for ordered chains of L-alanine in five solvents and in *in vacuo* studies. The values of $\langle \hat{l}_k^2 \rangle$ for various standard secondary structures and for $k \rightarrow \infty$ are shown at the right in the figure: (A) *in vacuo*, (B) methanol, (C) water, (D) formic acid, (E) ethanol, and (F) acetic acid.

cates a radical departure in the conformational properties of the biopolymer. Only conformations in the upper left-hand corner of the map, β and left-handed 3_1-helical-type structures, are permitted. The global minimum is at $\phi = -100°$, $\psi = 120°$. A relative minimum is also noted near the β position on the map. This radical change in the conformational properties is not too surprising since one expects the polar groups of the solvent molecules to strongly interact with lc-Ala. Interactions involving the carbonyl oxygen and amide hydrogen with solvent would be especially stabilizing.[23] The net effect of such interactions would be to "pull" the α helix apart in order to expose the carbonyl oxygens and amide hydrogens to solvent. Thus extended structures should be preferred.

An extended structure, a near left-handed 3_1 helix, presently called the *extended coil*, has been identified for the charged form of poly(L-glutamic acid) in aqueous solution.[12,24] This conformation is mainly stabilized by the interaction of the COO^- groups with solvent, and the minimizing of the intrachain electrostatic energy by maximizing the distance between COO^- groups. Solvent interactions with backbone carbonyl and amide groups make favorable contributions to the promotion of the extended coil, but are small in comparison to the interactions involving COO^- groups. This new conformation was discovered, in part, because its CD-ORD spectra are similar to poly(L-proline) II spectra. Recently, a number of workers studying more exotic polymer-solvent systems have reported CD spectra similar to that of poly(L-proline) II. A list of these studies is given in ref 25, several examples are: $[N^5$-(2-hydroxyethyl)-L-glutamine$]_n$[26,27] in water, (γ-ethyl L-glutamate)$_n$ in sulfuric acid–water mixtures,[28] (γ-methyl L-glutamate)$_n$ in fluor *gem*-dials,[25] and (phenylalanine)$_n$ in methanesulfonic acid–water mixtures.[29] If these polymers adopt the extended coil conformation it cannot be stabilized by the strong electrostatic repulsions present in poly(L-glutamic acid). In view of the calculations reported here, such structures are stable due to highly favorable interactions between carbonyl oxygens and/or amide hydrogens in the peptide backbone with the highly polar solvent molecules.

However, the conformational map of lc-Ala in formic acid contradicts the conclusion of the preceding paragraph. The first thought one should have is to expect lc-Ala in formic acid to behave nearly the same as in acetic acid. An inspection of the map of lc-Ala in formic acid indicates that the biopolymer has conformational properties very similar to lc-Ala in aqueous solution. From an inspection of the polymer-solvent parameters presented in Table I it is seen that the Δf's of formic acid and acetic acid are nearly identical while the R_V's of formic acid are about midway between the R_V's of water and acetic acid. The first five and the seventh V_f listed in Table I are necessary to describe interactions between solvent and (Ala)$_n$. In four out of six of these cases the V_f for formic acid are closer in value to the V_f of water than that of acetic acid. This includes the important carbonyl oxygen and amide hydrogen values.

We conclude from these observations that in this case the size and shape of the solvent molecule overrides the free-energy interactions between solvent and solute species in specifying conformation. This is a consequence of our model; whether or not such a volume effect due to size and topology of solvent molecules can be such a critical factor in dictating conformation is not known.

Note that chain aggregration, or chain folding, which could provide stabilization free energy through inter- or intrachain hydrogen-bond formation, respectively, is not taken into consideration in our calculations. The extent of polymer–solvent interactions could also be seriously modified by chain aggregation, or chain folding, through excluded volume effects. Our discussion is valid only for isolated chains of L-alanine which presumably corresponds to very dilute solutions.

Figure four demonstrates how the mean-square residue length, $\langle l_k^2 \rangle$, of an L-alanine unit depends upon solvent and ordered chain length. Two general observations can be made from Figure 4; first, all five solvents promote the unfolding of the right-handed α helix, and, secondly, the ordered secondary structures for $k > 14$, *i.e.*, chains longer than 14 residues, have $\langle \hat{l}_k^2 \rangle$ values nearly identical with the polymer. This suggests that L-alanine oligomers composed of 14 or more residues are very nearly identical with the polymer in ordered secondary structure in all five solvents considered here.

Oligomeric L-alanine chains *in vacuo*, water, methanol, and formic acid undergo an extended conformation $\rightarrow$ right-handed α-helix transition as a function of increasing k. At some specific chain length, different for each solvent, the energy gained by the formation of the intrachain hydrogen bonds overcomes the stabilization energy resulting from the polymer-solvent interactions. For acetic acid the polymer-solvent interaction energy stabilizes extended secondary structures to such a degree that an α helix is not realized. In ethanol there is a balanced equilibrium between the formation of α helix and the retention of extended conformations leading overall to a large value for $\langle \hat{l}_k^2 \rangle$.

***trans*-Proline.** In Figure 5 are shown the conformational energy maps, using the notation of Krimm and Venkatachalam,[8] for two peptide units of *trans*-L-proline, and for a long ordered chain of *trans*-L-proline peptide units (lct-Pro). The former is the model for the least-ordered, or random, form of *trans*-poly(L-proline) (rct-Pro), and the

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(29) E. Peggion, L. Strasorier, and A. Cosani, *J. Amer. Chem. Soc.*, **92**, 381 (1970).

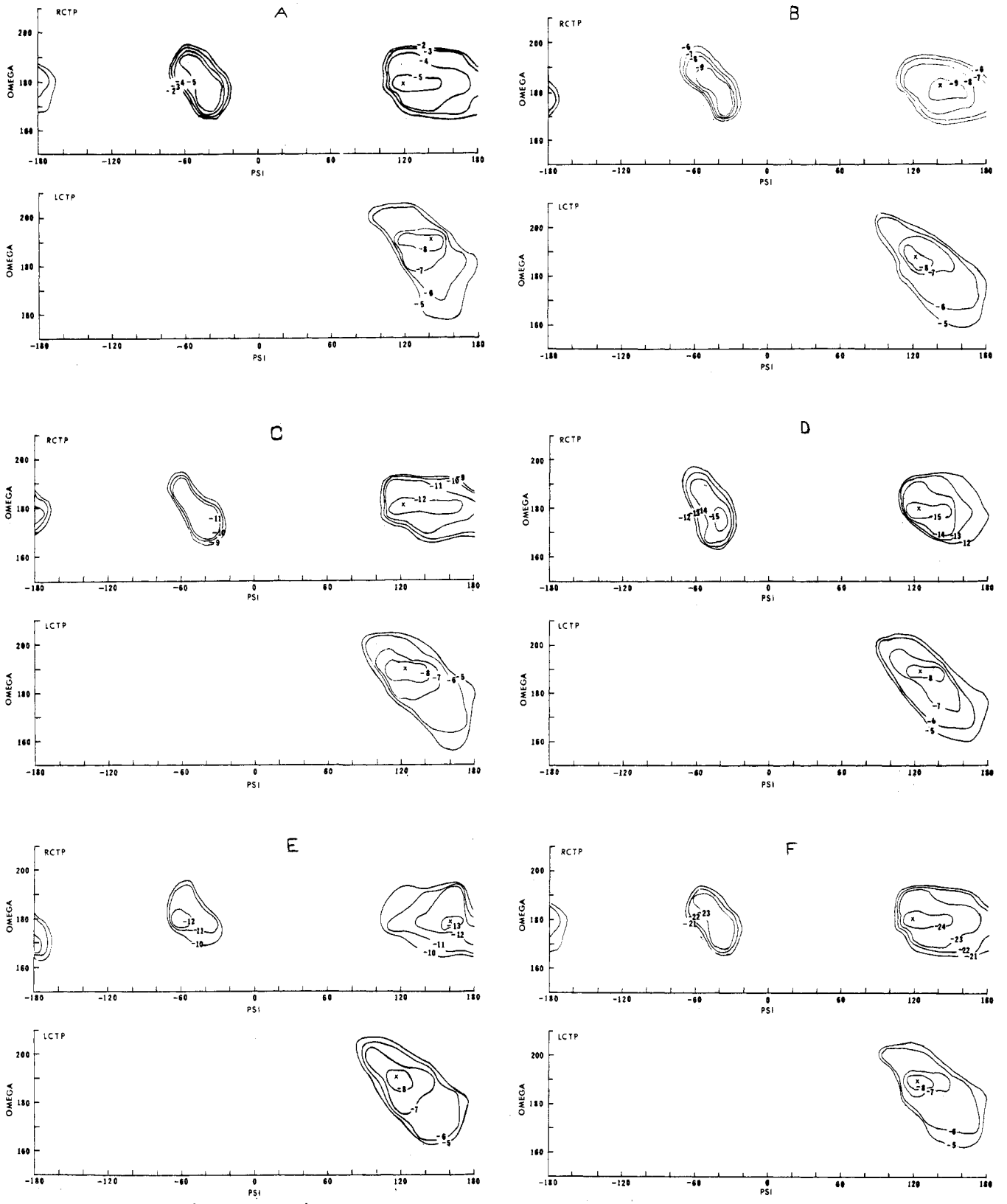

Figure 5. Conformational energy maps of ψ *vs.* ω (rotation about the C′–N imide bond) for rct-Pro and lct-Pro. The torsional rotation barrier about the C′–N is: $V(\omega) = (A/2)(1 - \cos 2\omega)$, $A \sim 20$ kcal/mol. This is from F. K. Winkler and J. D. Dunitz, *J. Mol. Biol.*, **59**, 169 (1971). The energy contours are in kcal/mole per residue. The X's denote the global minima: (A) *in vacuo*, (B) aqueous solution, (C) methanol, (D) ethanol, (E) formic acid, and (F) acetic acid.

latter is a model for the perfectly ordered polymer. Figure 5 contains conformational energy maps for each of the five solvents considered in this report. Note that both Krimm and Venkatachalam and ourselves compute the polymer-solvent *free* energy for each trial conformation.

rct-Pro has two major energy minima. The deepest and broadest lies in the region ψ = 120–180°, and ω = 160–200°. This minimum corresponds to the left-handed 3_1

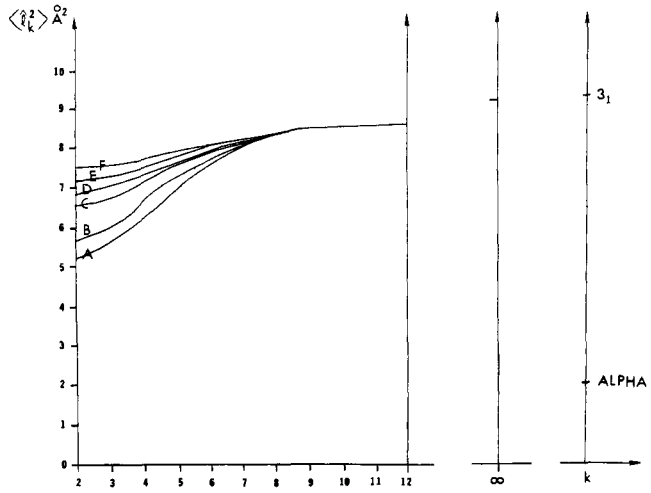

Figure 6. Plots of $\langle \hat{l}_k^2 \rangle$ *vs.* k for ordered chains of *trans*-L-proline in five solvents and in *in vacuo* studies. The values of $\langle \hat{l}_k^2 \rangle$ for two standard secondary structures and for $k \rightarrow \infty$ are shown at the right in the figure: (A) methanol, (B) *in vacuo*, (C) water, (D) formic acid, (E) ethanol, and (F) acetic acid.

helix observed in the solid state.[19] The other minimum occurs in the region of the right-handed α helix, $\psi = -60°$. This particular conformation has been observed for proline residues in several globular proteins.[30] There is, at present, considerable debate whether this minimum is possible for a dimer of *trans*-L-proline. Some workers feel it is an artifact of inaccurate potential functions. rct-Pro is very sensitive to solvent in the sense that the magnitude of the polymer-solvent interaction varies considerably with the choice of solvent. The stable secondary structures of rct-Pro, however, are rather insensitive to solvent. Both methanol and ethanol have a greater stabilizing interaction with rct-Pro than water. The reason for this is due to the absence of the NH group and the addition of the CH_2 groups from the pyrrolidine ring. The only highly favorable interaction of the water molecules with the proline residue is through the carbonyl oxygen. However, some of the stabilization free energy gained through this interaction is lost to unfavorable interactions between the water molecules and the ring CH_2 groups. Methanol and ethanol, on the other hand, have a moderately favorable interaction with the carbonyl oxygen, through the OH group, and with the CH_2 units, through hydrophobic bonding with the CH_3 or CH_2CH_3 groups of methanol or ethanol, respectively. Thus it appears that these two moderately favorable interactions of methanol and ethanol with rct-Pro are more stabilizing than the difference between one highly stabilizing and several slightly destabilizing interactions as takes place when water molecules interact with rct-Pro. The absence of the NH group prevents a water molecule from acting as a binary-binding solvent species. The interaction of the carbonyl oxygen of rct-Pro with the neutral forms of acetic and formic acids is sufficiently stabilizing so that both these solvents interact more favorably with rct-Pro than the other three solvents considered here.

In all solvents torsional rotations about the imide bond up to ±10° are energetically possible. The global minima, however, always occur for $\omega = 180°$. The shape of the minima, specifically their broadness with respect to ω, suggests fluctuations in ω occur at room temperature in all five solvents.

(30) a) Subtilisin, Pro-225; b) Staphylococcal Nuclease, Pro-47; c) Lysozyme, Pro-70; d) Carboxypeptidase-A, Pro-288; e) Hemoglobin, Pro-37, Pro-44, Pro-77, Pro-95.

Table II
Characteristic Polymer–Solvent Interactions of lct-Pro and lcc-Pro[a]

Solvent	lct-Pro	lcc-Pro
Water	−0.5	+0.1
Methanol	−0.2	+0.2
Ethanol	−0.6	0.0
Formic acid	−0.4	−0.1
Acetic acid	−0.8	−0.3

[a] See text for definitions, in five solvents. These values were obtained at the global minima in total conformational energy. Energies are in kcal/mole per residue.

The conformational energy maps of lct-Pro possess a single minimum in conformational energy which corresponds to the left-handed 3_1 helix. Each of the five solvents interact with lct-Pro in such a way that the total resultant polymer-solvent interaction is small. In each instance, the stereochemistry of the left-handed 3_1 helix is apparently sufficiently restrictive to preferential solvent binding so that the stabilization free energy, realized through carbonyl oxygen–polar group of the solvent molecule interactions plus either favorable, or unfavorable, CH_2–solvent molecule interactions, results in total interaction free energies between lct-Pro and solvent which are only slightly stabilizing. At first this would appear contrary to the findings of Krimm and Venkatachalam[8] who conclude that lct-Pro is very much stabilized as a left-handed 3_1 helix through interactions with water. However, we find the same if we consider *only* the interactions of water molecules with the carbonyl oxygens as done in their calculations. Nevertheless, there is a substantial decrease in stabilization free energy when the remainder of the lct-Pro atoms are allowed to interact with water. The $CH_2 \cdots H_2O$ polymer solvent interactions are the primary source of the destabilization free energy.

Torsional rotations about the imide bond up to ±10° may occur in lct-Pro. The global minima, for all solvents, occur at $\omega \approx 187°$. This is the first instance in which a nonplanar configuration of a peptide unit has been shown to be energetically more favorable than the corresponding planar geometry.

In Figure 6 is a plot of $\langle \hat{l}_k^2 \rangle$ *vs.* k for *trans*-L-proline molecules in the five solvents and *in vacuo* studies. An analysis of this plot indicates that only the small, $k < 4$, oligomers of *trans*-L-proline are conformationally sensitive to solvent. Further, only these small oligomers have the capacity to adopt secondary structures other than the left-handed 3_1 helix. The low-energy region near $\psi = -60°$, $\omega = 180°$ provides this added chain flexibility up to $k = 4$. For *trans*-L-proline oligomers consisting of four or more peptide residues the left-handed 3_1 helix is the preferred secondary structure of lct-Pro in all five solvents and *in vacuo* studies. These findings are in agreement with earlier experimental studies[31] where, in aqueous solution, the left-handed 3_1 helix was preferentially adopted starting with the tetra (*trans*-L-proline) oligomer.

***cis*-Proline.** Figure 7 contains conformational energy maps of rcc-Pro, random-chain *cis*-L-proline, and lcc-Pro, long-chain *cis*-L-proline, *in vacuo* and aqueous solution. The rcc-Pro maps contain two minimum energy regions. The deepest, in the vicinity of $\omega = 0°$ and $\psi = 160°$, corresponds to the secondary structure observed in the solid state.[32] The second minimum corresponds to a conformation in which the second planar peptide unit lies in plane

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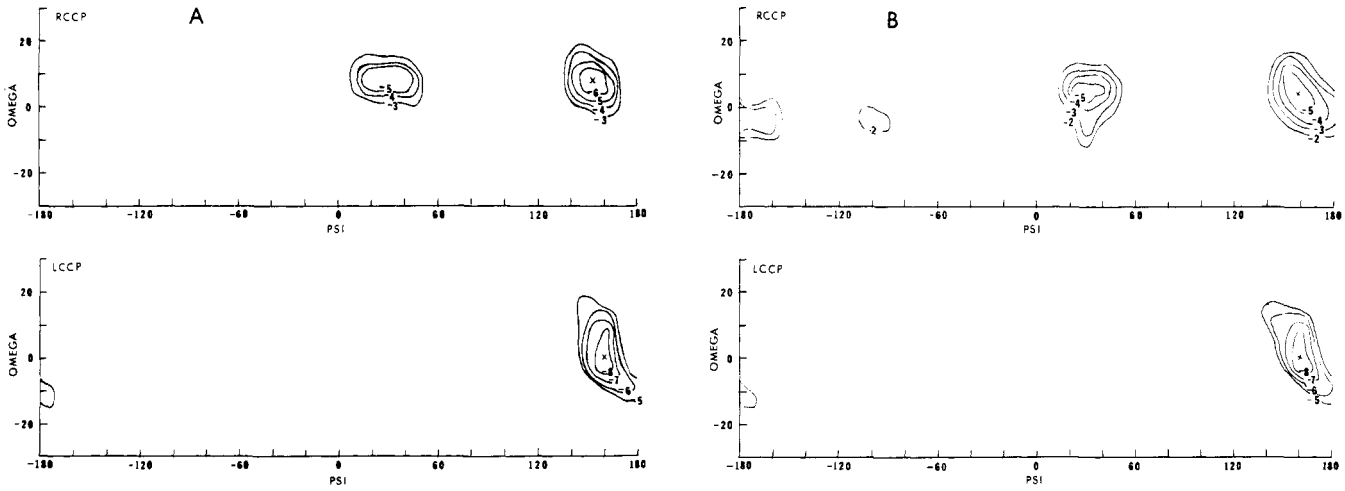

Figure 7. Conformational energy maps of ψ *vs.* ω for rcc-Pro and lcc-Pro in (A) *in vacuo* and (B) aqueous solution. The energy contours are in kcal/mole per residue and the X's denote global minima.

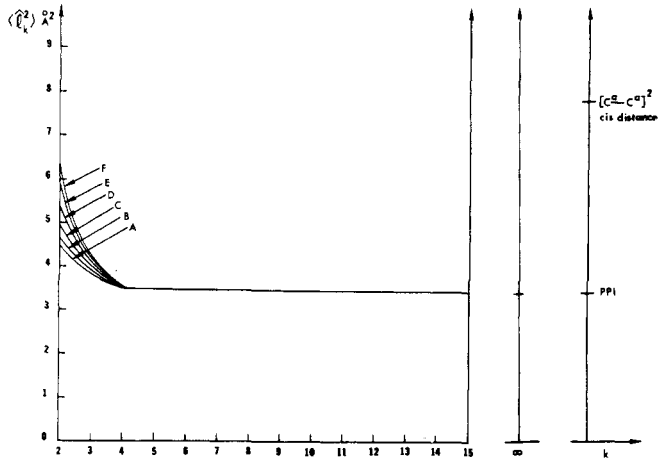

Figure 8. Plots of $\langle \hat{l}_k^2 \rangle$ *vs.* k for ordered chains of *cis*-L-proline in five solvents and in *in vacuo* studies. The values of $\langle \hat{l}_k^2 \rangle$ for the polymeric form in the solid state, PPI, is listed at the far right along with the $(C^\alpha\text{–}C^\alpha)^2$ distance of the cis residue for $\omega = 0°$. The value of $\langle \hat{l}_k^2 \rangle$ is also listed at the right of the figure: (A) *in vacuo*, (B) methanol, (C) formic acid, (D) water, (E) ethanol, and (F) acetic acid.

which nearly bisects the $N\text{–}C^\alpha\text{–}C^\beta$ bond angle of the pyrrolidine ring of the first planar-peptide unit. The carbonyl oxygen bond of the second residue is pointed away from the pyrrolidine ring of the first residue. rcc-Pro shows about the same conformational sensitivity to solvent as does rct-Pro. This is demonstrated in Figure 8 where $\langle \hat{l}_k^2 \rangle$ of *cis*-L-proline is plotted as a function of k, the number of residue units in an ordered chain. The maximum variation in $\langle \hat{l}_k^2 \rangle$ as a function of solvent takes place for $k = 2$.

The conformational properties of lcc-Pro is extremely insensitive to solvent. The severe stereochemical restrictions in this biopolymer, as reflected in the *in vacuo* and aqueous solution conformational energy maps, does not allow polymer–solvent interactions to modify secondary chain structure. In Table II are listed the values of the polymer–solvent interactions of lct-Pro and lcc-Pro for each of the respective global energy minima. It is noted that the interactions of lcc-Pro with solvent are smaller in magnitude than the corresponding polymer–solvent interactions of lct-Pro. This is in qualitative agreement with the findings of Krimm and Venkatachalam.[8] Also, the cis form of the biopolymer exhibits a less polar, more hydrophobic character in the polar solvents than does the trans form. This finding is in agreement with experiment.[33] The enhanced hydrophobic character of the cis form is due to, as postulated,[33] greater exposure of the pyrrolidine rings to solvent, and less exposure of the carbonyl oxygens. Values of $|\omega| \leq 10°$ are possible for both rcc-Pro and lcc-Pro with the energy minima always occuring at $\omega = 0°$.

Summary

Both the polarity and the *size* of the solvent molecule play important roles in the extent to which solvent will dictate the conformation of a polypeptide chain. For $(Ala)_n$, the balance between polymer–solvent interactions and the formation of intrachain hydrogen bonds in the right-handed α' helix results in a specific minimum chain length in order to maintain the α-helical secondary structure. Very polar solvents prevent the α helix from forming regardless of chain length. The absence of the NH group in both *cis*- and *trans*-$(Pro)_n$ coupled with the presence of the pyrrolidine ring leads to relatively small overall interactions with solvent as compared to $(Ala)_n$. *trans*-Poly(L-proline), in the ordered form, has larger interactions with solvent than the ordered cis form of the biopolymer. The conformational properties of both *trans*- and *cis*-L-proline oligomers is less sensitive, for all solvents, to the length of the ordered chain, k, than L-alanine oligomers. This is due to the fact that L-alanine chains with $k \geq 4$ have the capacity to adopt one of two possible conformations, extended or right-handed α helical, while both *trans*- and *cis*-L-proline chains are limited to a single secondary structure for $k \geq 4$.

Significant deviations from planarity are possible in both the *cis*- and *trans*-L-proline residue units as a result of rotations about the imide bond. The global energy minima for lct-Pro occurs for $\omega \approx 187°$ while the global minima for lcc-Pro is found at $\omega = 0°$. The polymer–solvent interactions are relatively insensitive to rotations about the imide bond. Work is now underway to evaluate the effect of torsional rotations about *amide* bonds upon the conformational properties of the corresponding polypeptide chains.

Acknowledgments. Kenneth H. Forsythe is pleased to acknowledge fellowship aid from The Southern Fellowships Fund. Part of this work was supported by a National Institute Dental grant (Grant Number DE-02587-06).

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(33) See L. Mandelkern in "Poly-α-Amino Acids," G. D. Fasman, Ed., Marcel Dekker, New York, N. Y., 1967, p 675.