

The β -Conformation of Poly(*S*-[(3-hydroxypropyl)-carbamoylmethyl]-L-cysteine) in Aqueous Mixtures of D₂O, Aliphatic Alcohols, and Acetic Acid

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ABSTRACT: Poly(*S*-[(3-hydroxypropyl)-carbamoylmethyl]-L-cysteine) [poly(Cys[CamPrOH])] has been prepared by the amidation of poly(*S*-carboboxyethyl-L-cysteine) with 3-amino-propanol, and the properties of poly(Cys[CamPrOH]) whose number-average molecular weight is 12,000 have been investigated in various aqueous mixtures. Although the conformation of poly(Cys[CamPrOH]) in H₂O is close to a random coil, it can be converted into the β -conformation in D₂O. The β -conformation is formed in aqueous mixtures of CH₃OH, C₂H₅OH, iso-C₃H₇OH and *n*-C₃H₇OH, respectively, and its residue ellipticity at 200 nm at the highest alcohol content of the solubility limit becomes higher as the alcohol is more hydrophobic. In a 97% acetic acid solution poly(Cys[CamPrOH]) takes on the β -conformation.

KEY WORDS Poly(*S*-[(3-hydroxypropyl)-carbamoylmethyl]-L-cysteine) / β -Conformation / Aliphatic Alcohols / Deuterium Oxide / Infrared Spectra / Circular Dichroism / Optical Rotatory Dispersion /

It has been demonstrated¹⁻³ that poly(*S*-carboxymethyl-L-cysteine) and poly(*S*-carboxyethyl-L-cysteine) can take on the β -conformation of an antiparallel chain arrangement in an aqueous solution when their side-chain carboxyl groups are at low ionizations. The conformations of these polypeptides were determined by spectroscopic methods, but they must be intermolecularly associated in aqueous solutions when hydrogen-bonded β -conformation is developed. In order to observe the association behavior of these ionized polypeptides in solution, we have to consider the effects of the electric charges of their side-chains on various solution properties such as light scattering.⁴

In this connection we have prepared a water-soluble but non-ionizable polypeptide capable of forming the β -conformation in solution, by a method analogous to the derivation of poly(*N*⁵-[3-hydroxypropyl]-L-glutamine) from poly(γ -benzyl L-glutamate).^{5,6} The polypeptide is poly(*S*-[(3-hydroxypropyl)-carbamoylmethyl]-L-cysteine), which will be abbreviated below as poly(Cys[CamPrOH]).

In a previous communication,⁷ we reported some

results on the measurement of circular dichroism of solutions of poly(Cys[CamPrOH]) in D₂O and its temperature dependence. While randomly coiled in H₂O at temperatures ranging from 1.5 to 80°C, poly(Cys[CamPrOH]) is in the β -conformation in D₂O at 1.5°C and is gradually transformed into a random coil with raising temperature.

In the present paper we will describe the synthesis of poly(Cys[CamPrOH]) in some detail and also the results of circular dichroism measurements of poly(Cys[CamPrOH]) dissolved in mixtures of D₂O with H₂O. The circular dichroism of poly(Cys[CamPrOH]) dissolved in mixtures of various aliphatic alcohols with H₂O is also measured at different compositions. Poly(Cys[CamPrOH]) is shown to have a conformation close to a random coil containing a small amount of ordered parts in H₂O, and, as the H₂O content decreases, the β -conformation comes about. In an 80% *n*-C₃H₇OH mixture the β -conformation is formed most perfectly among the solutions examined. The optical-rotatory-dispersion and intrinsic-viscosity measurements indicate that in CH₃COOH-H₂O mixtures the β -conformation is stabilized at CH₃COOH

contents exceeding 90%.

EXPERIMENTAL

Materials

Poly(*S*-carbobenzoymethyl-L-cysteine) was prepared as described previously.^{8,9} 2-Aminoethanol and 3-aminopropanol were purchased from Tokyo Kasei Kogyo Co., Inc., and were dried over BaO and vacuum-distilled before use. Spectrograde D₂O (99.75%) was obtained from Merck. CH₃OH and iso-C₃H₇OH were spectrograde reagents from Tokyo Kasei Kogyo Co., Inc., and C₂H₅OH, *n*-C₃H₇OH and CH₃COOH were reagents of special grade from Hayashi Junyaku Kogyo Co., Inc. CH₃COOD was obtained from Merck.

Preparation of Solutions

Solutions of poly(Cys[CamPrOH]) in D₂O or alcoholic mixtures were prepared by dissolving the polypeptide sample in H₂O (or D₂O in the case of D₂O-rich solutions) to bring about a concentration of about 0.2 g dl⁻¹ and then by diluting the solution with D₂O or alcohol (or H₂O) to give a desired solvent composition. Solutions of poly(Cys[CamPrOH]) in CH₃COOH-H₂O mixtures were prepared first by dissolving the polypeptide sample in CH₃COOH and then diluting the solution with H₂O. The solvent composition is expressed by the volume ratio of D₂O, alcohol or CH₃COOH with H₂O. Solutions of poly(Cys[CamPrOH]) in CH₃COOD-D₂O were similarly prepared and their compositions are also given in the volume ratio.

Measurements

The infrared spectra were measured on a Jasco Infrared Spectrophotometer DS 402-G at 25 ± 2°C. Films of polypeptides were cast from aqueous solutions on AgCl plates, and 1% solutions of poly(Cys[CamPrOH]) in CH₃COOD-D₂O mixtures were put in a cell of CaF₂ windows having path length 0.025 mm.

The circular dichroism was measured on a Jasco Circular Dichrometer J-20, using a jacketed cell having path length, 1 or 2 mm. The concentration of solutions was around 0.02 g dl⁻¹ or 10⁻³ M in residue, and the residue ellipticity is expressed by $[\theta]$ (degree cm² decimol⁻¹). The temperature was controlled, and usually kept at 25 ± 0.01°C by circulating water of constant temperature through

the jacket, and it was read by a Takara Thermistor Thermometer.

The optical rotatory dispersion was recorded on the same instrument used for measuring the circular dichroism, over the wavelength region of 600 to 280 nm, at 25 ± 0.01°C, using a jacketed cell of path length 2 cm. The concentration of solutions was about 0.1 g dl⁻¹, and the specific rotation is given by $[\alpha]$.

The relative viscosity of the poly(Cys[CamPrOH]) solutions in CH₃COOH-H₂O mixtures was measured by an Ubbelohde viscometer whose flow time for water was 180 s at 25 ± 0.01°C.

RESULTS

Synthesis and Solid Conformation

The reaction of poly(*S*-carbobenzoymethyl-L-cysteine)^{8,9} with 3-aminopropanol gave poly(Cys[CamPrOH]). To 6 g or poly(*S*-carbobenzoymethyl-L-cysteine) 48 ml of 3-aminopropanol were added, and this mixture was stirred for half a day at 60°C. Then 12 ml of chloroform were added and the mixture was kept at 60°C for 5 days. Swollen gels dissolved after one day, at which time the reaction was actually completed. The mixture was poured into about 500 ml of 3% acetic acid solution, and the solution was then dialyzed against distilled water with its frequent changes. Following filtration, the solution was lyophilized and 3.2 g of poly(Cys[CamPrOH]) were isolated. Sample code: TX-4071.

Anal. Calcd for C₈H₁₄O₃N₂S: C, 44.04%; H, 6.42%; N, 12.84%; S, 14.68%. Found: C, 43.44%; H, 6.61%; N, 13.30%; S, 13.45%.

Poly(Cys[CamPrOH]) was soluble in dichloroacetic acid, trifluoroacetic acid, *m*-cresol, dimethyl sulfoxide and hexamethylphosphoramide. It was also soluble in water up to about 0.3 g dl⁻¹, and this solubility did not change with temperature up to 70°C. Poly(Cys[CamPrOH]) was also soluble in D₂O, but its solubility in D₂O was lower than that in H₂O. The polypeptide was soluble in CH₃COOH-H₂O mixtures of CH₃COOH contents less than 98%.

The polypeptide sample used had a number-average molecular weight of 12,000 in 0.1% CH₃COOH solution, when measured by osmotic pressure. The sample had an intrinsic viscosity of 0.31 dl g⁻¹ in dichloroacetic acid at 25°C.

When poly(Cys[CamPrOH]) was stored over P_2O_5 *in vacuo*, its solubility in H_2O decreased with time, as was also the case for poly(N^5 -[3-hydroxypropyl]-L-glutamine). It was possible, however, to retard the decrease in solubility in H_2O by storing the polypeptide sample over P_2O_5 but not *in vacuo*.

The infrared spectra of poly(Cys[CamPrOH]) in the film cast from aqueous solution are shown in Figure 1. The amide I band is located at 1625 cm^{-1} , and the amide II band is at 1528 cm^{-1} , indicating that the polypeptide is in the β -structure in the solid state, even though the peptide groups are also on its side chains.

In order to prepare poly(*S*-[(2-hydroxyethyl)-carbamoylmethyl]-L-cysteine), poly(*S*-carbobenzyloxymethyl-L-cysteine) was reacted with 2-aminoethanol at 60°C . The reaction mixture became brown in color after overnight and readily soluble in water. Following dialysis against distilled water, the insoluble materials were removed by centrifugation, and the supernatant was lyophilized. The lyophilized material was cast in film from its aqueous solution, and the infrared spectra were examined. A broad amide I band appeared around 1650 cm^{-1} and amide II bands were at 1555 and 1580 cm^{-1} . These bands indicate that the polypeptide is randomly coiled in the solid state. In addition, no circular dichroism could be observed down to the peptide ultraviolet region. Thus we conclude that the amidation reaction of poly(*S*-carbobenzyloxymethyl-L-cysteine) with 2-aminoethanol brought about racemization and possibly partial peptide scission.

Circular Dichroism and Conformation in Solutions

The circular dichroism of poly(Cys[CamPrOH]) in D_2O - H_2O mixtures at 1.5°C has a broad negative band around 225 nm and a band around 198 nm changing its sign according to the solvent composition. With increasing D_2O content, the band at 198 nm changes its sign from negative to positive. The value of residue ellipticity at 198 nm at 1.5°C is plotted against D_2O content in Figure 2. In these solvent mixtures poly(Cys[CamPrOH]) should have been subjected to partial deuterium substitution to a degree dependent on the solvent composition, but the observed results clearly show the coil-to- β transition of poly(Cys[CamPrOH]) with increasing D_2O content.

The conformational transition occurs sharply from 0 to 10% D_2O content but proceeds gradually with increasing D_2O content beyond 10%. The deuterium substitution brings about the coil-to- β transition in two steps, and this is similar to the coil-to- β transition of poly(*S*-carboxymethyl-L-cysteine) induced by titration with aqueous HCl solution.³

Figure 3 shows the circular dichroism of poly(Cys[CamPrOH]) in CH_3OH - H_2O mixtures. With increasing CH_3OH content, the negative band at 225 nm becomes deeper and shifts slightly to the red, and the band at 200 nm changes its sign from negative to positive. The negative ellipticity at 228 nm is very large at the solvent composition of the solubility limit, *i.e.*, at 97.6% CH_3OH , but the positive ellipticity at 200 nm is of the same order of magnitude as that in D_2O at 1.5°C . These results can be taken to mean that the coil-to- β transition of

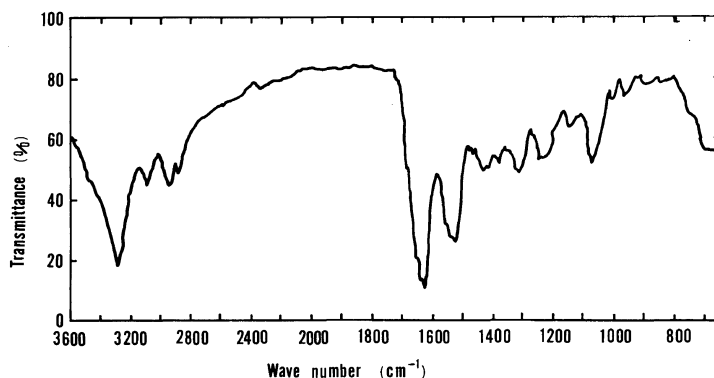


Figure 1. Infrared spectra of poly(Cys[CamPrOH]) in the film cast from aqueous solution.

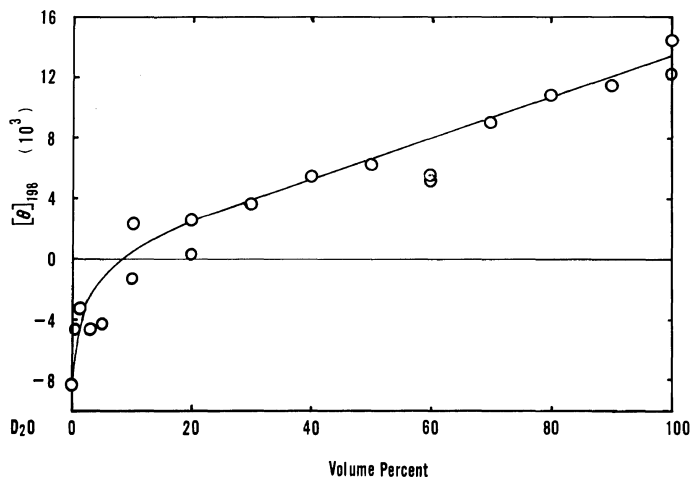


Figure 2. Relation of residue ellipticity at 198 nm to the volume percent of D₂O in D₂O-H₂O mixtures at 1.5°C.

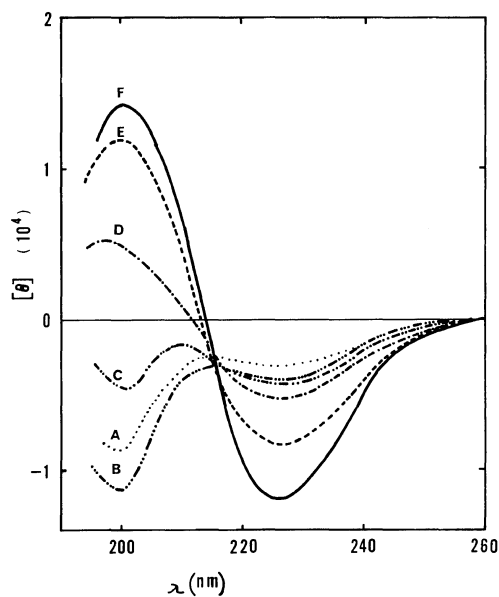


Figure 3. Circular dichroism of poly(Cys[CamPrOH]) in CH₃OH-H₂O mixtures at 25°C. CH₃OH content (%): A, 0; B, 0.1; C, 10; D, 30; E, 80; F, 97.6.

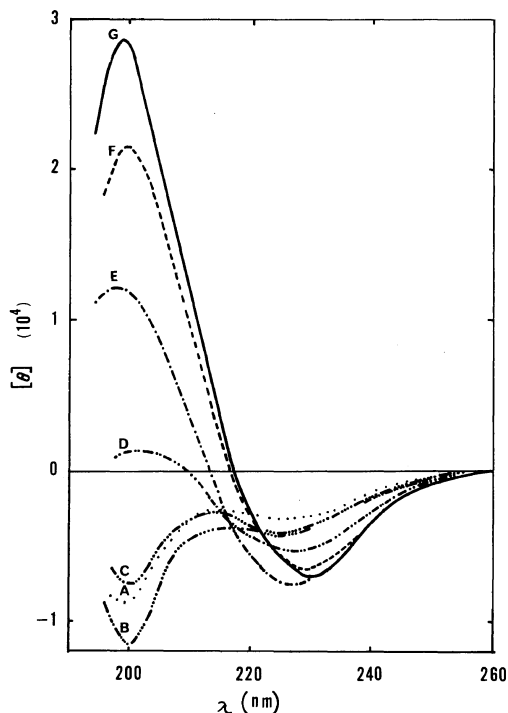


Figure 4. Circular dichroism of poly(Cys[CamPrOH]) in C₂H₅OH-H₂O mixtures at 25°C. C₂H₅OH content (%): A, 0; B, 0.1; C, 20; D, 30; E, 60; F, 80; G, 85.

poly(Cys[CamPrOH]) occurs also with increasing CH₃OH content.

Figures 4 and 5 show the circular dichroism of poly(Cys[CamPrOH]) in C₂H₅OH-H₂O and *n*-C₃H₇OH-H₂O mixtures, respectively. The circular

dichroism changes similarly to that in the CH₃OH-H₂O mixtures cited above, when the solvent

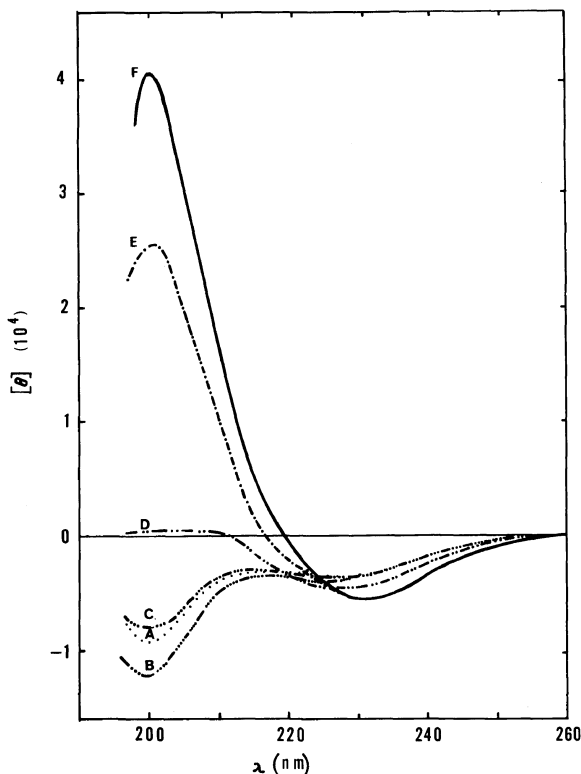


Figure 5. Circular dichroism of poly(Cys [CamPrOH]) in n -C₃H₇OH-H₂O mixtures at 25°C. n -C₃H₇OH content (%): A, 0; B, 0.1; C, 50; D, 60; E, 70; F, 80.

composition is altered. At the highest alcohol content, the negative ellipticities at 225 nm are smaller than those in CH₃OH-H₂O mixtures, but the positive ellipticities at 200 nm are much larger, especially, in n -C₃H₇OH-H₂O mixtures. In iso-C₃H₇OH-H₂O mixtures the circular dichroism behaves similarly. The observed changes in circular dichroism with increasing alcohol content is considered to show the coil-to- β transition of poly(Cys[CamPrOH]).

In all these alcoholic mixtures, it can be noticed that at an alcohol content of 0.1% the negative ellipticity at 200 nm is the lowest, in spite of the gradual increase in negative ellipticity at 225 nm. We may interpret the results as disruption of some ordered structure, possibly, the β -conformation, in H₂O by the addition of a small amount of alcohol. The random coil conformation of poly(Cys[CamPrOH]) in 0.1% alcohol solutions gives residue ellipticities, about -12,000 at 200 nm

and about -4,000 at 225 nm, and the circular dichroism of this random coil has a greater similarity to random coil poly(*S*-carboxyethyl-L-cysteine) than to random coil poly(*S*-carboxymethyl-L-cysteine).³

Figure 6 shows the residue ellipticity at 200 nm as a function of alcohol content in various solvent mixtures. While the conformation of poly(Cys[CamPrOH]) in CH₃OH-H₂O mixtures changes into the β -conformation asymptotically with increasing CH₃OH content, it undergoes a sharper transition to the β -conformation by the addition of other alcohols in increasing content.

Table I summarizes the values of residue ellipticity of poly(Cys[CamPrOH]) in D₂O and aqueous alcoholic mixtures having the highest alcohol content. The high value of the negative ellipticity at 228 nm in 97.6% CH₃OH should be attributed to the environmental effect of high CH₃OH content, since the corresponding value at 85% CH₃OH is as much

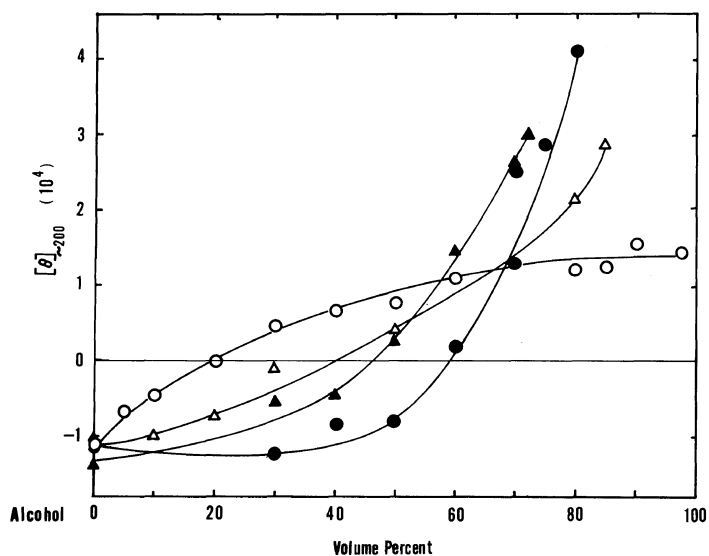


Figure 6. Relation of residue ellipticity at 200 nm to the volume percent of alcohol in aqueous alcoholic mixtures at 25°C: ○, CH₃OH-H₂O; △, C₂H₅OH-H₂O; ▲, iso-C₃H₇OH-H₂O; ●, n-C₃H₇OH-H₂O.

Table I. Circular dichroism of poly (Cys [CamPrOH]) in various solvent mixtures

Solvent	Temperature	Volume % of alcohol	λ	$[\theta]$	λ	$[\theta]$
	°C		nm	(degree cm ² decimol ⁻¹)	nm	(degree cm ² decimol ⁻¹)
D ₂ O	1.5		197.5	17,000	227.5	- 5,400
CH ₃ OH-H ₂ O	25	97.6	200	16,000	227.5	- 12,000
	1.5		200	22,000	227.5	- 13,500
C ₂ H ₅ OH-H ₂ O	25	85.0	199	28,000	230	- 7,800
iso-C ₃ H ₇ OH-H ₂ O	25	72.0	200	29,000	231.5	- 8,000
n-C ₃ H ₇ OH-H ₂ O	25	80.0	202	41,000	227.5	- 5,000

as those in the other alcoholic mixtures at the solubility limit. On the other hand, the value of positive ellipticity at 200 nm becomes larger as a higher alcohol is added, but the content of alcohol is lower at the solubility limit.

In Figures 2 and 6, we may note two features in the formation of β -conformation. When D₂O or CH₃OH is added to H₂O, the formation of the β -conformation is gradual and the residue ellipticity reaches, at most, 22,000. On the other hand, when higher alcohols are added to H₂O, the coil-to- β transition occurs gradually at a low alcohol

content and then the transition proceeds sharply when a certain alcohol content has been exceeded. In n-C₃H₇OH-H₂O mixtures, the conformation is scarcely influenced by the addition of a small amount of alcohol, but the coil-to- β transition occurs sharply above a 50% alcohol content, and the residue ellipticity reaches 41,000 at a 80% n-C₃H₇OH content.

It seems likely that the coil-to- β transition proceeds more sharply and a more perfect β -conformation is attained, as the more hydrophobic alcohol is added.

Behavior in Aqueous Acetic Acid Solutions

As mentioned earlier, poly(Cys[CamPrOH]) is soluble in aqueous acetic acid solutions of various acetic acid contents up to 98%. The infrared spectra of poly(Cys[CamPrOH]) in CH₃COOD–D₂O mixture at low CH₃COOD contents revealed that the amide I band locates at a frequency, 1625, 1629, 1630, and 1635 cm⁻¹, in 0.01, 1, 2 and 5% CH₃COOD, respectively. Figure 7 illustrates the infrared spectra of the solution of poly(Cys[CamPrOH]) and the solvent of 0.01% CH₃COOD at the amide I band region. At a certain frequency region the absorption of solvent exceeds that of the solution, and this is caused by the highly absorbing CH₃COOD and its substitution for poly(Cys[CamPrOH]). The observed frequency at low CH₃COOD contents clearly indicates the induction of β -conformation by D₂O, and its shift toward higher values can be interpreted as the destruction of the β -conformation by the added CH₃COOD.

The conformation of poly(Cys[CamPrOH]) in CH₃COOH–H₂O mixtures can be investigated over a wider region of compositions by means of optical rotatory dispersion. Figure 8 shows the optical rotatory dispersion of poly(Cys[CamPrOH]) in these mixtures of different compositions. The levorotation increases at low CH₃COOH contents,

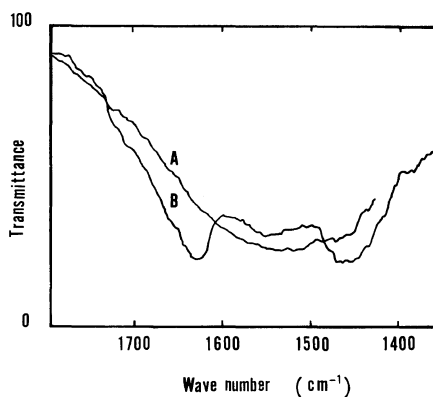


Figure 7. Infrared spectra of poly(Cys[CamPrOH]) in CH₃COOD–D₂O mixtures of 0.01% CH₃COOD content: A, solvent; B, solution.

but a large increase in dextrorotation occurs at the highest CH₃COOH content. It was found that the optical rotatory dispersion obeys the Moffitt–Yang equation^{10,11}

$$[\alpha] = \frac{n^2 + 2}{3} \frac{100}{M_o} \left\{ \frac{212^2 a_o}{\lambda^2 - 212^2} + \frac{212^4 b_o}{(\lambda^2 - 212^2)^2} \right\}$$

where M_o is the residue molecular weight (218), n the refractive index, λ the wavelength in nm, and a_o and b_o are the dispersion constants, respectively.

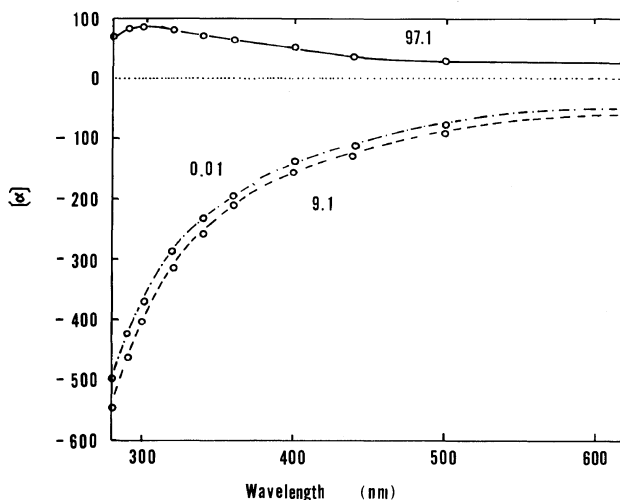


Figure 8. Optical rotatory dispersion of poly(Cys[CamPrOH]) in CH₃COOH–H₂O mixtures at 25°C. Values indicate the volume % of CH₃COOH.

Figure 9 shows the dispersion constants as a function of the CH_3COOH content, and it can be seen that the value of a_0 changes greatly at both extrema, while the value of b_0 remains zero within ± 20 , except at the highest CH_3COOH content. The large decrease in a_0 by the addition of a small amount of CH_3COOH indicates the presence of a small amount of ordered conformation, possibly the β -conformation, in H_2O and its disruption by added CH_3COOH . Such an effect has also been observed on the addition of alcohols to a 0.1% content. The large increase in a_0 as well as that of dextrorotation at the highest CH_3COOH content can be attributed to the formation of β -conformation. Only at the intermediate compositions of the solvent mixture is the random-coil conformation stable.

In Figure 9 the intrinsic viscosity, $[\eta]$, of poly(Cys[CamPrOH]) in $\text{CH}_3\text{COOH}-\text{H}_2\text{O}$ mixtures is also given as a function of the solvent composition, and it is clear that it behaves quite similarly to a_0 . The steep changes in intrinsic

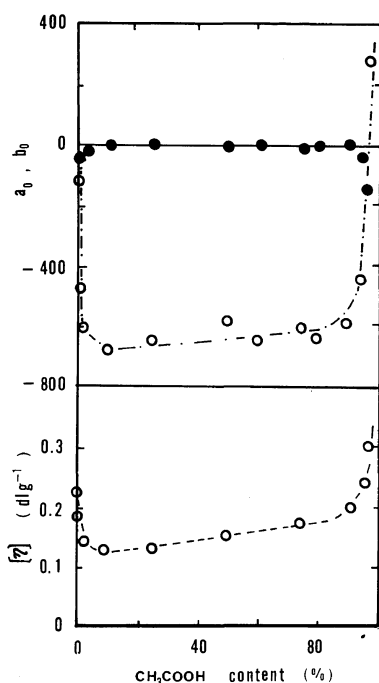


Figure 9. Relation of dispersion parameters and intrinsic viscosity to the volume percent of CH_3COOH in $\text{CH}_3\text{COOH}-\text{H}_2\text{O}$ mixtures at 25°C : top, \circ , a_0 and \bullet , b_0 ; bottom, $[\eta]$.

viscosity also represent conformational transitions of poly(Cys[CamPrOH]).

DISCUSSION

There may be two causes for the conformational effects on polypeptides: one is the effect of deuteration of peptide groups, and the other is the environmental effect of the deuterated solvent component.

In organic solvent mixtures containing dichloroacetic acid or water, it was observed that the α -helices of poly(γ -benzyl L-glutamate)¹²⁻¹⁴ and poly(β -benzyl L-aspartate)¹⁵ were slightly stabilized with respect to temperature when they were deuterated. However, it was inferred that the stability of the α -helix of poly(β -benzyl L-aspartate) did not change on deuteration when it was examined in dioxane-dimethyl sulfoxide mixtures.¹⁵ From these results it seems likely that deuteration does not influence the intrinsic stability of α -helices appreciably but the apparent increase in the stability of α -helices arises from alteration of the properties of the solvent.

In an aqueous salt solution of poly(L-glutamic acid) or poly(L-lysine) the helix-coil transition occurs at a more alkaline pH (pD) on deuteration, and this seems to indicate that the helix of poly(L-glutamic acid) is stabilized by deuteration while that of poly(L-lysine) is weakened by deuteration.¹⁶ These apparently conflicting results were attributed to the reduced acidity of side-chain carboxyl and ammonium groups, respectively, by deuteration, and Appel and Yang¹⁶ concluded that the intrinsic stability of α -helices is not influenced by deuteration.

It is thus considered that deuteration has only a small effect on the stability of polypeptide conformation. In this respect, the marked effect of D_2O on the conformation of poly(Cys[CamPrOH]) as observed here is unique. There has never been observed such a large effect of D_2O on the conformation of poly(L-lysine)¹⁷⁻¹⁹ and poly(*S*-carboxymethyl-L-cysteine)¹ to form the β -conformation in aqueous solutions.

The effect of deuteration on the peptide hydrogen bonding of *N*-methylacetamide in CCl_4 solutions has been noted.²⁰ Measurements of sedimentation equilibrium demonstrated that deuterated *N*-methylacetamide gave a weight-average molecular

weight about twice as high as the protiated material, indicating that the self-association in CCl_4 was extensively promoted by deuteration. From these results we may infer that deuteration of peptide groups promotes the self-association of molecules through the hydrogen-bonding of these groups even in aqueous solutions.

The effect of D_2O on the micelle formation has been observed in aqueous solutions of dodecylpyridinium iodide; the critical micelle concentration of the cationic surfactant was shown to be slightly lowered in D_2O than in H_2O .²¹ Thus association through hydrophobic interaction is also promoted by D_2O than it is by H_2O .

For the stabilization of the β -conformation of polypeptides the interaction between the side-chain groups, whether it is hydrogen-bonding or hydrophobic, must be strongly operative, as compared with that of α -helices. Consequently, self-association through hydrogen-bonding between side-chain peptide groups and hydrophobic interaction strengthened in D_2O could induce the coil-to- β transition of poly(Cys[CamPrOH]) in aqueous solutions.

The effect of added aliphatic alcohols on poly(L-glutamic acid) and poly(L-ornithine) in water has been investigated by potentiometric titration, and it was found that the helix is formed by the addition of alcohol.^{22,23} The effect of alcohols on the coil-to-helix transition is more effective and cooperative, as the alcohol is more hydrophobic. However, it was also observed from the rotation measurements that poly(L-lysine hydrochloride) was subject to a sharp coil-to-helix transition even in $\text{CH}_3\text{OH-H}_2\text{O}$ mixtures at 88% CH_3OH content.²⁴ Similarly, the helix-coil transition was induced by the addition of 75% iso- $\text{C}_3\text{H}_7\text{OH}$ in the case of poly(L-lysine hydrobromide).²⁵ Thus there seems to be no difference in the sharpness of the transition induced by the two alcohols.

The effect of aliphatic alcohols on poly(N^5 -[ω -hydroxyalkyl]-L-glutamine) in water is the promotion of the helix formation,^{5,6,26,27} but the sharpness of this formation is seen to depend on the degree of polymerization of the polypeptide.²⁸ In connection with the formation of β -conformation of poly(Cys[CamPrOH]) in a concentrated CH_3COOH solution, it should be noted that acetic acid behaves as a helix-forming solvent for poly(N^5 -[3-hydroxypropyl]-L-glutamine).⁵

While the addition of aliphatic alcohols induces the helix formation of polypeptides, we do not have much data on the effect of these alcohols on the formation of β -conformation. It was observed that low-molecular-weight poly(L-serine) soluble in water was subject to the coil-to- β transition in an aqueous solution when CH_3OH or trifluoroethanol was added.^{29,30} The residue ellipticity of poly(L-serine) in 80% CH_3OH was as high as 60,000 at 200 nm.

The optical activity measurements of water-soluble block copolymers of L-valine sandwiched between two poly(DL-lysine hydrochloride) blocks gave some conflicting results. Epand and Scheraga³¹ reported that the poly(L-valine) part in their copolymers was in the β -conformation in H_2O but was converted into the α -helix in 98% CH_3OH . Kubota and Fasman³² found, however, that the poly(L-valine) part in their block copolymer had the β -conformation in both water and aqueous CH_3OH solutions.

The circular dichroism measurements of deca(L-alanine)-poly(oxyethylene) ester, $\text{H}(\text{Ala})_{10}\text{O}[\text{POE}]$, showed evidence for a greater preference to the β -conformation in trifluoroethanol than in H_2O .³³ However, it was found that hexa- and hepta-(L-methionine)- ω -amino-poly(oxyethylene)amide, Boc-(Met)_{6 or 7}-NH[POE], could take on the β -conformation in H_2O , whereas they were in the disordered conformation in CH_3OH and $\text{C}_2\text{H}_5\text{OH}$.³⁴

These polypeptides having "aliphatic" side chains can take on the α -helix as well as the β -conformation, and this fact would be the reason for giving such complex conformational stability to these polypeptides. Our results on the effect of aliphatic alcohols to bring about the coil-to- β transition of poly(Cys[CamPrOH]) are similar to those of poly(L-serine),^{29,30} and they will apply to polypeptides which do not take on the α -helix.

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