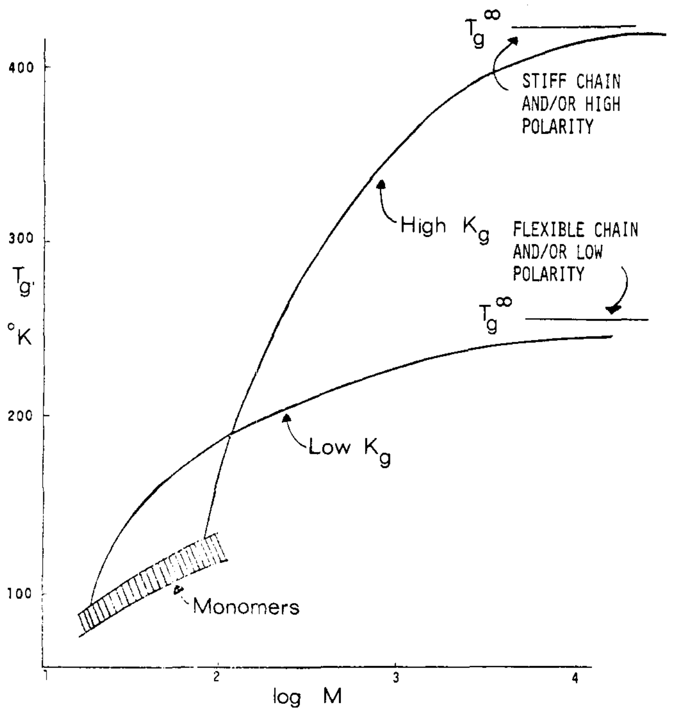

Figure 2. Schematic interpretation of results shown in Figure 1 using a T_g-log M plot. The shaded region shows the expected variation of monomer T_g's with molecular weight judged from data on low molecular weight, nonmonomeric compounds.

slightly dependent on molecular weight ($K_g \sim 1 \times 10^4$ as already stated). If T_g^∞ is very high because, for example, of chain stiffness, it is obvious that K_g must be higher than for a polymer of low T_g^∞.

The functional form of the curve in Figure 1 is still open to precise determination pending additional data and/or a suitable theory. Omitting the single point for $K_g = 0$, a plot of log K_g *vs.* T_g^∞ is a reasonable straight line. We originally believed that K_g *vs.* $(T_g^\infty)^2$ was a reasonable good fit but one reviewer of this paper is convinced that (a) K_g varies as $(T_g^\infty)^4$ and (b) that K_g may well depend on some factor(s) other than T_g^∞.

Several authors[3-5,9,11-13] have tried to interpret K_g in terms of molecular parameters arising from theory, the most common being those of Bueche, Gibbs-DiMarzio, Kanig, and Somcynsky and Patterson. We have not noted any expression for K_g that would lead to a semilogarithmic or a quartic relation between K_g and T_g^∞. The closest is that of Somcynsky and Patterson[14] who give the expression

$$K_g = SM_0T_g^\infty \tag{3}$$

where S is a chain stiffness factor and M_0 is the segment length. It is generally recognized that T_g^∞ increases with chain stiffness. There is also a strong tendency for T_g^∞ to increase with M_0 if one avoids long alkyl side-chain polymers. Hence Somcynsky-Patterson could lead to a K_g-$(T_g^\infty)^3$ correlation but we do not wish to emphasize such a point at this time. We recognize that many other factors affect T_g^∞ such as polarity, symmetry, and tacticity.

Full details, with appropriate literature documentation, are currently being written up. The K_g-T_g^∞ relationship in Figure 1 can be verified provisionally with data assembled by Pezzin *et al.*,[5] which tabulation does not include all of the examples in Figure 1[6,8,9,11,12] nor our modification of literature values of K_g.

Finally, we note that eq 1 is obeyed by a series of lactones with molecular weights ranging from 72 to 240, and leading to a $K_g = 0.45 \times 10^4$. Since these ring compounds lack end groups, it is apparent that molecular weight itself can lead to a lowering of T_g (at least for low molecular weight compounds), possibly through a free volume effects. T_g's of these lactones (but not K_g) have been reported.[15]

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(14) T. Somcynsky and D. Patterson, *J. Polym. Sci.*, **62**, 151 (1962).

(15) J. V. Koleske and R. D. Lundberg, *J. Polym. Sci., Part A-2*, **10**, 323 (1972).

Raymond F. Boyer
The Dow Chemical Company
Midland, Michigan 48640

September 10, 1973

A Double Thermal Helix-Coil Transition

The great majority of thermally induced conformational transitions in polypeptides in mixed organic solvents investigated to date have been found to be of the so-called "inverse" type, in which the ordered conformation is stable at the higher temperature.[1] Nevertheless it has been recognized for many years that in such cases a second transition in the "normal" (helix-to-coil) direction should in principle be observed at a relatively still higher temperature.[2] Up to now such a double transition has not been reported in spite of investigations over quite wide temperature ranges: for example, optical rotation studies of poly(γ-benzyl L-glutamate) have been made in several solvents up to 200° without gaining unequivocal evidence for even the onset of the expected second transition before degradation,[3] attesting to the high stability of the ordered conformation of this particular polypeptide. Such studies are, of course, always limited by the chemical and physical integrity of the polypeptide and solvent system.

Recently the calculation of the effect of solvent composition on helix-coil transition temperatures has been extended in a quantitative manner that has made it possible to delineate polypeptide-solvent combinations in which the possibility of observing a double transition in an experimentally accessible temperature range would be maximized.[4] These results predict that the requirement was for an intrinsically weak polypeptide (*i.e.*, one with a relatively low helix-to-coil transition temperature in pure inert solvents) in combination with a fairly strongly interacting active solvent component which is needed to provide an "inverse" transition in a normally accessible temperature range. These requirements are met by the system poly(β-benzyl L-aspartate) ((BzlAsp)$_n$) and $HCCl_2COOH$. The relative weakness of the (BzlAsp)$_n$ helix is suggested by two observations: (1) the fact that (BzlAsp)$_n$, alone amongst polypeptides of this class, has been shown to undergo both "normal" and "inverse" thermal transitions according to the solvent system used,[5] and (2) the relatively low $HCCl_2COOH$ concentration necessary to induce an isothermal transition from the helical to the random-coil conformation (about 7.5 mol % in $HCCl_2COOH$-$CHCl_3$ mixtures at 25°.[6]) The instability has been attributed to the atypical left-handed chirality of the α-helical

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(1) G. D. Fasman in "Poly-α-Amino Acids," Vol. 1, G. D. Fasman, Ed., Marcel Dekker, New York, N. Y., 1967.

(2) L. Peller, *J. Phys. Chem.*, **63**, 1199 (1959).

(3) J. S. Franzen, J. B. Harry, and C. Bobik, *Biopolymers*, **5**, 193 (1967).

(4) F. E. Karasz and G. E. Gajnos, *J. Phys. Chem.*, **77**, 1139 (1973).

(5) (a) Y. Hayashi, A. Teramoto, K. Kawahara, and H. Fujita, *Biopolymers*, **8**, 403 (1969). (b) R. H. Karlson, K. S. Norland, G. D. Fasman, and E. R. Blout, *J. Amer. Chem. Soc.*, **82**, 2268, (1960).

(6) P. Dubin and F. E. Karasz, *Biopolymers*, **11**, 1745 (1972).

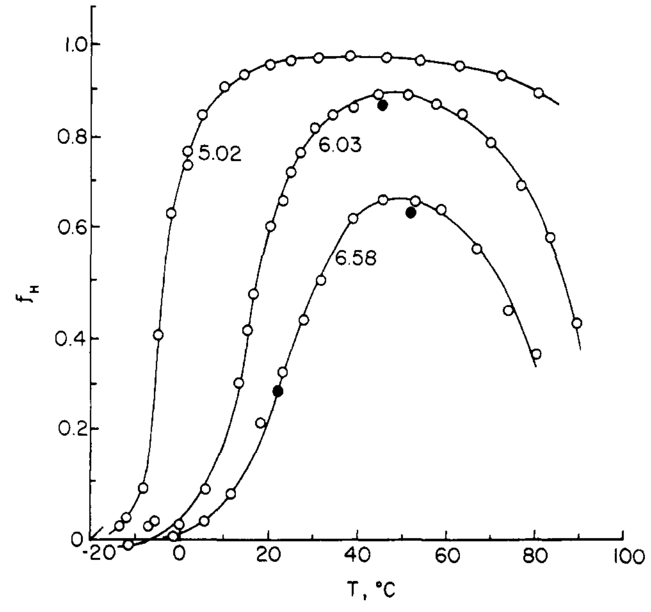

Figure 1. Fractional helical content of $(BzlAsp)_n$ as a function of temperature in $Cl_4C_2H_2$-$HCCl_2COOH$ solutions (0.3% w/v) containing mole per cent $HCCl_2COOH$ as shown. Filled circles: repeat measurements after exposure to highest indicated temperatures; $(BzlAsp)_n$ (Sigma Chemical lot 90C-0560), viscosity av mol wt 230,000.

form of this poly(L-amino acid).[1] The inverse thermal transition in the $(BzlAsp)_n$-$HCCl_2COOH$-$CHCl_3$ system has already been thoroughly investigated in the 0-50° temperature range;[5] no upper transition was observed, though in a similar study using CCl_4 as the inert component there was perhaps some indication of a decrease in f_H, the fractional helical content of the polypeptide, around 60°.[7]

The theory referred to above predicts that in systems of this type the effect of increasing the relative content of the interacting solvent in the solvent mixture is to raise the transition temperature of the lower, "inverse" transition and decrease that of the "normal" process. From the analysis of available data it was predicted that in the case of the $(BzlAsp)_n$-$HCCl_2COOH$ system the two transitions would merge at a "critical" temperature of about 54° and at a "critical" $HCCl_2COOH$ content of about 8.15 mol %.[4] Thus any investigation in solvent compositions containing less than this amount of $HCCl_2COOH$ might be expected to reveal a double transition if the study were to encompass a sufficiently wide temperature range. In fact, the considerable breadth of the transition in the temperature-solvent composition plane makes it desirable to investigate solvent compositions in which the $HCCl_2COOH$ content is somewhat below the critical composition to obtain substantial conversion of the polypeptide to the ordered state.

By substituting 1,1,2,2-tetrachloroethane [bp 146° (760 mm)] for the more volatile chloroform as the inert solvent component in the $(BzlAsp)_n$-$HCCl_2COOH$ system it was possible to extend the accessible temperature region to 85°; further extension was prevented by polymer degradation. However, this range was sufficient to verify the existence of an upper, "normal," transition. This was observed between 60 and 80° according to solvent composition. Figure 1 shows typical f_H *vs.* T curves at several solvent compositions. Helical contents were calculated from optical rotation measurements taken at 436 nm. The latter had been correlated with the Moffitt-Yang b_0 parameter[6] in separate experiments. Such curves were found to be completely reversible if the maximum temperature to which the polypeptide was exposed did not exceed about 85°. Thus, at the lowest $HCCl_2COOH$ concentration shown, the customary coil-to-helix transition is observed with essentially complete transformation to the ordered conformation; the latter is preserved up to the highest temperature used. In a solution containing 6.58 mol % $HCCl_2COOH$, in contrast, an increase in temperature first induces the inverse transition (which is, however, incomplete because of the finite transition width; see Figure 6, ref 4 in regard to this point) and this is followed by a decrease in f_H indicative of the helix-to-coil process. Complete conversion back to the coil is proscribed because of solute degradation; it is estimated that a temperature of about 100° would be required to reduce f_H to 0.1. Also shown is an intermediate case with a $HCCl_2COOH$ content of 6.03 mol %. The phase boundary, corresponding to f_H = 0.5 for the $(BzlAsp)_n$-$HCCl_2COOH$-$Cl_4C_2H_2$ system, is shown in Figure 2. It is seen to be displaced from that for the $(BzlAsp)_n$-$HCCl_2COOH$-$CHCl_3$ system (when the solvent composition is plotted in terms of the molar content of $HCCl_2COOH$) by about 1 mol %, presumably because of the difference in the activity coefficients of $HCCl_2COOH$ in the two solvent mixtures.

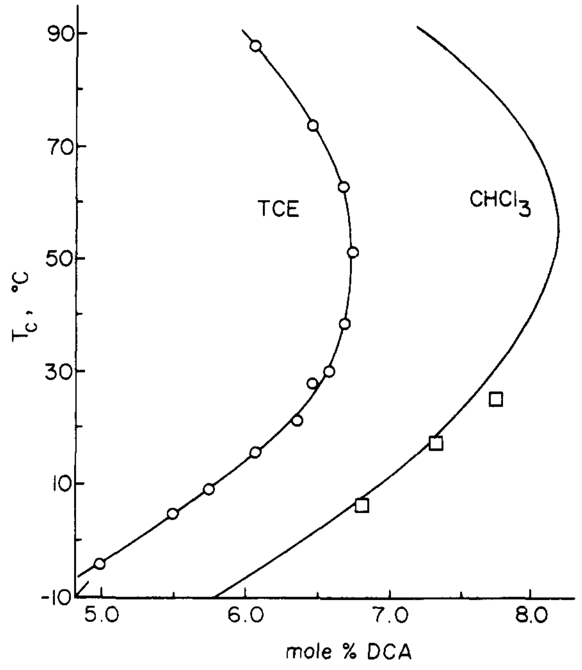

Figure 2. Transition temperatures and phase boundaries (f_H = 0.5) for $(BzlAsp)_n$ in $Cl_4C_2H_2$-$HCCl_2COOH$ and $CHCl_3$-$HCCl_2COOH$ solvent mixtures as function of composition. Helical and random-coil conformations of $(BzlAsp)_n$ predominate on the left and right of the phase boundaries, respectively: $Cl_4C_2H_2$, solid line represents best fit of experimental points (present work); $CHCl_3$, solid line from theory,[4] points from ref 5a and 6.

The present results confirm experimentally the existence of a temperature of maximum stability for the ordered conformation of $(BzlAsp)_n$. Thus by analogy with current concepts of conformational stability in proteins we may interpret the lower transition as an example of "cold denaturation."[8] In contrast to proteins, however, the molecular mechanism responsible for the phenomenon in homopolypeptides is simpler and perhaps less equivocal: the upper transition is made possible by the entropy gain

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(7) T. Norisuye, K. Misumi, A. Teramoto, and H. Fujita, *Biopolymers* (in press).

(8) J. F. Brandts, *J. Amer. Chem. Soc.*, **86**, 4302 (1964); J. F. Brandts and L. Hunt, *ibid.*, **89**, 4826 (1967).

of the ordered polypeptide in the process of destabilization, while in "cold denaturation" this gain is offset and exceeded overall by the loss in solvent entropy accompanying the interaction of the active solvent with the polypeptide backbone. Of course, we do not wish to imply here that the concomitant enthalpic changes are not of equivalent importance in the thermodynamic features of the transition.

It is entirely probable that such double transition will be found in other polypeptide-solvent systems. The helix-to-coil transition temperatures of polypeptides even with intrinsically very stable ordered conformations can be greatly depressed by the presence of a sufficiently strongly interacting solvent and, given a wide enough experimental temperature range, the lower transition might also be observable in many cases.

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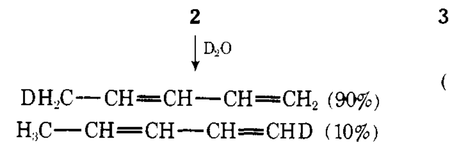
R. P. McKnight and F. E. Karasz*
Polymer Science and Engineering, University of Massachusetts
Amherst, Massachusetts 01002
Received August 22, 1973

Alkali Metal Catalyzed Polymerization of Some Conjugated Dienes Bearing an Allylic Proton

The alkali metal catalyzed polymerization of 1,3-pentadiene in tetrahydrofuran was found to be inhibited completely by triethylamine giving the dienyl alkali metal compound in place of poly(pentadiene). This observation led us to a general method for the preparation of pure dienyl alkali metal compounds:[1] *i.e.*, a conjugated or an unconjugated diene is allowed to react with an alkali metal in tetrahydrofuran in the presence of triethylamine. In this reaction, an unconjugated diene isomerizes to a conjugated one, and the latter reacts with an alkali metal, without any gas evolution, to give the dienyl alkali metal by the abstraction of a hydrogen atom from the methyl or methylene group adjacent to the conjugated system, and by the formation of the reduced dimer(s) of the conjugated diene. This finding gave us further insight into the mechanism of the well-known alkali metal catalyzed polymerization of some conjugated dienes.[2] This paper describes the results obtained with 1,3-pentadiene and 2,3-dimethyl-1,3-butadiene.

cis- and *trans*-pentadiene (**1**), C_5H_8 (1.5 mol), was allowed to react with dispersed potassium (1.0 mol) in an argon atmosphere in a tetrahydrofuran (2.0 mol)-triethylamine (1.0 mol) mixture to give crystalline pentadienylpotassium tetrahydrofuranate (**2**), $C_5H_7K \cdot C_4H_8O$, and a mixture of three reduced dimers (**3**), $C_{10}H_{18}$, following the stoichiometry represented in eq 1.[1]



$$\underset{\mathbf{1}}{4CH_3{-}CH{=}CH{-}CH{=}CH_2} + 2K \xrightarrow[(C_2H_5)_3N]{C_4H_8O} \underset{\mathbf{2}}{2[CH_2{=}CH{=}CH{=}CH{=}CH_2]K \cdot C_4H_8O} + \underset{\mathbf{3}}{C_{10}H_{18}} \xrightarrow{D_2O} \begin{array}{l} DH_2C{-}CH{=}CH{-}CH{=}CH_2\ (90\%) \\ H_3C{-}CH{=}CH{-}CH{=}CHD\ (10\%) \end{array} \tag{1}$$

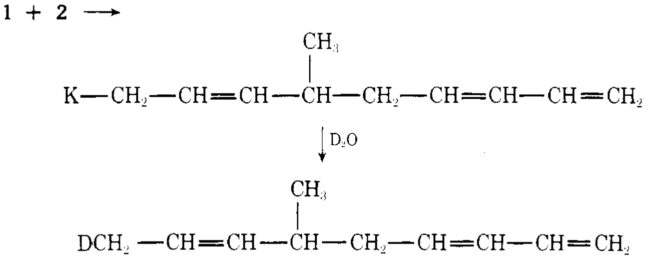
Table I
Polymerization of 1 and 5[a]

Monomer	Catalyst	Yield of Polymer (%)	Microstructure of Polymer		
			1,2 (%)	1,4 (%)	3,4 (%)
1	Metallic K	83	20	80	0
	2	82	25	75	0
5	Metallic K	85	35	65	
	5	85	34	66	

[a] Polymerization condition: catalyst, 1 mol % of monomer; solvent, H_4furan; temperature, 30°; time, 5 days for **1** and 4 days for **5**.

The crystalline dienylpotassium **2** behaved as a catalyst in the polymerization of **1** to give results quite similar to those obtained with a metallic potassium catalyst (Table I). Although this fact seems to suggest that the polymerization of **1** catalyzed by metallic potassium in the absence of amine proceeds *via* **2** as an initiation species, there is no evidence to rule out the possibility that the dimer dianion of potassium forms initially. The reaction between **1** and metallic potassium in which triethylamine was absent depends on a temperature; lower temperature favors the formation of **2** and higher one gives exclusively poly(pentadiene). In these cases, dipotassium dianion and reduced dimers could not be isolated in crystalline forms. These experimental results imply that triethylamine not only inhibits the polymerization of **1**, but also acts the transferrer of hydrogen to result in the formation of reduced dimers (**3**).

The information about the second step in the polymerization was obtained from the equimolar reaction of **2** with **1**, which was carried out in tetrahydrofuran at −35° for 2 hr. Deuteriolysis of this reaction mixture with D_2O gave 6-methyl-1,3,7-nonatriene-9-d_1 (**4**)[3] in 20% yield, in addition to poly(pentadiene). The yield of **4** was about 60% in the case of the corresponding crystalline dienylsodium. In contrast to this, the reaction carried out at 30° gave exclusively poly(pentadiene). These experimental results suggest that the second step of the polymerization is mainly the 1,4 addition of **1** to **2**, in good agreement with the microstructure of poly(pentadiene) (Table I), and that the polymerization proceeds *via* **2** with the monoanionic active species.



$$\mathbf{1} + \mathbf{2} \longrightarrow K{-}CH_2{-}CH{=}CH{-}\overset{\displaystyle CH_3}{\overset{|}{C}H}{-}CH_2{-}CH{=}CH{-}CH{=}CH_2 \xrightarrow{D_2O} DCH_2{-}CH{=}CH{-}\overset{\displaystyle CH_3}{\overset{|}{C}H}{-}CH_2{-}CH{=}CH{-}CH{=}CH_2$$

4

2,3-Dimethyl-1,3-butadiene (**5**), C_6H_{10} (1.0 mol), was allowed to react with dispersed potassium (1.0 mol) in a tetrahydrofuran (2.0 mol)-triethylamine (1.0 mol) mixture in an argon atmosphere at 0° to give the dienylpotassium (**6**), $C_6H_8K_2 \cdot 2(C_4H_8O)$, as a yellowish powdery substance; yield, 60%. The filtrate contained the reduced dimer,

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(1) H. Yasuda, T. Narita, and H. Tani, *Tetrahedron Lett.*, 2443 (1973).

(2) A. V. Tobolsky and C. E. Rogers, *J. Polym. Sci.*, **40**, 73 (1959); *ibid.*, **61**, 155 (1962).

(3) The structure of **4** is supported by following data: ir 1805, 1002, 896 (δ(CH) of terminal vinyl), 1655, 1601 (ν(C=C) of conjugated diene), 949 (δ(CH) of conjugated diene), 965 (δ(CH) of trans CH=CH); nmr δ 0.46 (d, 3 H, CH_3—C—) 1.08 (d, 2 H, CH_2D—C=C) 1.58 (t, 2 H, =C—CH_2—C=) 4.65, 5.58 (m, 7 H, CH_2=, —CH=CH—); *Anal.* Calcd for $C_{10}H_{15}D$: 137. Found: 137 (mol wt by ms).