Macromolecules **1991**, *24*, 6435–6438

Thermodynamics of the Melting Point Depression in Poly(β-hydroxybutyrate-*co*-β-hydroxyvalerate) Copolymers

William J. Orts and Robert H. Marchessault*,†

Department of Chemistry, University of Toronto, Toronto, Ontario, Canada

Terry L. Bluhm

Xerox Research Centre of Canada, Mississauga, Ontario, Canada

Received February 19, 1991; Revised Manuscript Received July 11, 1991

ABSTRACT: Both small-angle X-ray scattering (SAXS) and wide-angle X-ray diffraction (WAXD) data were used to establish relationships between the degree of crystallinity, melting temperature, and lamellar thickness for random copolymers of poly(β-hydroxybutyrate-*co*-β-hydroxyvalerate) (PHB/V). It was shown that the predictions of Sanchez and Eby that lamellar thickness increases with both cyrstallization temperature and comonomer content are correct. The Sanchez–Eby model for inclusion of both comonomer units within crystalline regions was used to describe the thermodynamics of the melting point depression in isodimorphic systems such as PHB/V and predictions from this model were contrasted with predictions from the Flory exclusion model. It was found that for isodimorphic systems the Flory exclusion model does not correctly predict the melting point depression, however, the Sanchez–Eby inclusion model does provide a good fit to the experimental data.

1. Introduction

Poly(β-hydroxybutyrate) (PHB)[1,2] and its random β-hydroxyvalerate (HV) copolymers (PHB/V)[3,4] are biodegradable thermoplastics[5] with unique cocrystallization behavior.[6–8] The properties of PHB/V, such as energy to break, melting temperature, and heat of fusion, vary widely with copolymer composition, yet the crystallinity remains at a high level over the full range of compositions due to cocrystallization. This phenomenon is known as isodimorphism,[6] i.e., HV units pack into the PHB lattice, and vice versa, causing two morphic forms to be detected. As expected for isodimorphic copolymers, PHB and PHV chains have similar crystalline conformations; 2_1 helices with fiber repeats of 0.596 and 0.556 nm, respectively. The melting point with respect to composition decreases sharply to reach a broad minimum at approximately 40 mol % HV, at which point the crystal switches from a PHB-type lattice to a PHV crystal lattice. X-ray[6] and electron diffraction[8,9] studies have shown that the unit cell does not expand significantly as the HV composition increases, suggesting that there is sufficient room to accommodate the other counit in the unit cell. The consistently high degree of crystallinity at all compositions for these random copolymers means that some degree of inclusion of the comonomer within each of the two crystalline phases must occur. Determining the extent of inclusion (i.e., partitioning), the effect of thermal history on counit inclusion, as well as the role of inclusion in determining the physical properties of these polyesters is an ongoing challenge.

Models of the crystalline state of random copolymers have been based on two extremes: in the uniform exclusion model proposed by Flory,[10] the crystalline phase is composed only of crystallizable A units, while B units are excluded. Alternatively, inclusion models allow for B units in both the amorphous and crystalline phases, resulting in defects in the crystalline phase and uneven partitioning of B units between the crystalline and amorphous phases. Sanchez and Eby[11] extended the uniform inclusion model to allow for a full range of partitioning of B units between the two phases.[12] Their model has been applied to interpret the observed calorimetric properties of randomly chlorinated polyethylene (PE), with respect to the varying degree of chlorine inclusion in the PE crystalline phase.[13] Experiments using the Sanchez–Eby approach were carried out at low (mol %) compositions since inclusion was not extensive. In general, inclusion was still accompanied by decreases in the degree of crystallinity and expansion of the unit cell. Copolymers of bacterial, poly(β-hydroxybutyrate-*co*-β-hydroxyvalerate), are a better example of the Sanchez–Eby model since significant inclusion is thought to occur over the entire range of copolymer compositions.

It has been shown that the Flory copolymer equation underestimates the melting point depression of PHB/V random copolymers.[6,14] Sanchez and Eby showed that corrections for finite crystallite size can be made for both the Flory exclusion model and their inclusion model. On the basis of these arguments (an application of the kinetic theory[11]), they predicted that for isothermal crystallization, crystal thickness should increase with increasing concentration of the noncrystallizable counit. This prediction holds for both inclusion and exclusion models. At first thought this may seem unreasonable for the exclusion model, since it would be increasingly difficult to have long uninterrupted sequences of crystallizable A sequences with increasing B concentrations. However, it should be remembered that in the Flory model increased lamellae thickness must be accompanied by a sharp decrease in crystallinity.

In this work, data from calorimetric analysis, WAXD, and SAXS will be used to establish relationships between the degree of crystallinity, melting temperature, and lamellar thickness for PHB/V random copolymers. It will be shown that the lamellar thickness increases not only with crystallization temperature, but also with HV content, as predicted by Sanchez and Eby. The Sanchez–Eby model for uniform inclusion of counits between crystalline phases will be used as a framework to discuss the trends in HV inclusion within the PHB lattice and describe the thermodynamics of the melting point depression with copolymer composition.

---

† Present address: McGill University, 3420 University St., Montreal, Quebec, Canada H3A 2A7.

0024-9297/91/2224-6435$02.50/0

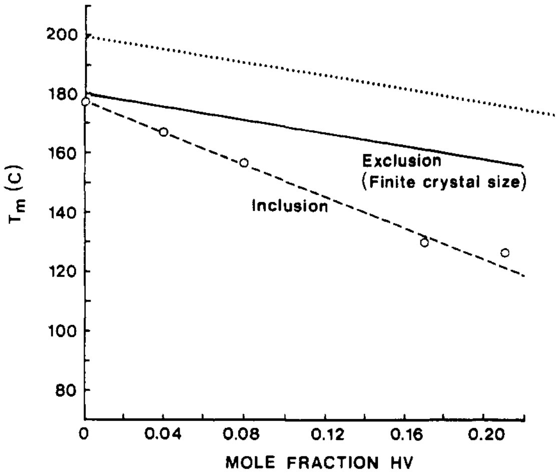

Figure 1. Melting point (T_m) vs mol % HV, X_{HV}, for PHB/V. The dotted curve is from the Flory equation (exclusion model) for melting point depression, the solid curve is derived from the exclusion model corrected for finite crystal size, and the dashed curve is derived assuming uniform inclusion of random comonomer units.

2. Experimental Section

Samples of PHB/V produced in pilot-scale batch fermentations of *Alcaligenes eutrophus* were kindly provided by Imperial Chemicals Industries (ICI), U.K. These polymers were obtained by solvent extraction from bacterial cultures. Similar materials are available from ICI under the trade name Biopol. Molecular weights of these isotactic random copolymers ranged from 300 000–500 000. Samples were melt pressed using a Carver press at their melt temperatures. A pressure of 2500 psig was applied, and the molding time was 4 min. Immediately following molding the samples were crystallized in vacuo at various temperatures reached either by cooling from the melt or by heating after quenching in liquid nitrogen. Samples were maintained at an oven crystallization temperature (T_c) until they reached an equilibrium degree of crystallinity (as measured by WAXD). Crystallization times of up to 500 h were required for some samples.

After the samples were removed from the oven, calorimetry data were recorded on a Perkin-Elmer DSC-2C instrument calibrated with an indium standard immediately before use. DSC traces were recorded at 20 deg/min. This rapid heating rate was used to minimize the effect of annealing during melting which could lead to lamellar thicknesses and T_m not being measured on the same crystals. Where multiple endotherms were detected, the melting point from the higher peak was taken as the true melting point.

X-ray crystallinity measurements were made on a Philips powder diffractometer Model PW 1710 equipped with a graphite monochromator and pulse height analyzer. Nickel-filtered Cu Kα radiation (λ = 0.1542 nm) was used with a voltage of 40 kV and current of 30 mA. The percentage of crystallinity was calculated from diffracted intensity data in the range 2θ = 10–40° using Ruland's method.[15] The amorphous scattering component was estimated by measuring the scattering curve from a sample immediately after quenching in liquid nitrogen. Small-angle X-ray scattering (SAXS) data was collected using a Kratky camera mounted on the same X-ray source. The scattered beam intensities were measured using a proportional counter at a distance of 20 cm from the sample. The entrance collimating slit was 100 μm and the receiving slit 75 μm. Corrections for slit smearing and Lorentz factor were made by multiplying the observed intensity (I) by the scattering vector ($s = 2\theta/\lambda$). thus lamellar spacings were determined from plots of sI vs s.

3. Results and Discussion

The arguments of Sanchez and Eby are applied to describe the melting point depression of PHB/V copolymers. Figure 1 shows the melting point (T_m) as a function of mole fraction (X_{HV}) for different annealing temperatures. The dotted line in Figure 1, which clearly underestimates the melting point depression, was derived by the Flory copolymer equation based on counit exclusion of equilibrium crystals, accordingly, the melting temperature is

$$\frac{1}{T_m} - \frac{1}{T_m{}^\circ} = -\frac{R \ln X_{HB}}{\Delta H_m{}^\circ} \qquad (1)$$

where T_m is the observed melting point and R the gas constant. Reference values of $T_m{}^\circ$, the equilibrium melting temperature of PHB homopolymer, and $\Delta H_m{}^\circ$, the molar heat of fusion, $T_m{}^\circ$ = 198 °C and $\Delta H_m{}^\circ$ = 3100 cal/mol, were obtained by best-fit extrapolation of DSC data to infinite crystal thickness and were comparable to those already reported.[16,18]

It can be argued that the Flory treatment is inappropriate since it describes equilibrium crystals and not copolymers crystallized rapidly. On the basis of kinetic arguments, the exclusion model has been rederived to account for finite crystal size[11,12,17] such that

$$T_m = T_m{}^\circ\left(1 - \frac{RT_m}{\Delta H_m{}^\circ} X_{HB} - \frac{2\sigma_e}{\Delta H_m{}^\circ l}\right) \qquad (2)$$

where σ_e is the surface free energy of the crystal surface and l is the lamellar thickness. One clearly sees that the surface correction term ($2\sigma_e/l$) causes a parallel shift of the Flory curve (solid line in Figure 1) to a more reasonable initial temperature, but does not account for the relatively steep decline in T_m with X_{HV}. Regardless of the value of σ_e, the PHB/V melting behavior can not be described by copolymer exclusion.

The melting point depression is more aptly described by the Sanchez–Eby inclusion model, as shown by the dashed line in Figure 1. The melting point decrease with respect to X_{HV} is accompanied by changes in the crystal packing energy due to a defect free energy ϵ. T_m is then described by

$$T_m = T_m{}^\circ\left(1 - \frac{\epsilon}{\Delta H_m{}^\circ} X_{HV} - \frac{2\sigma_e}{\Delta H_m{}^\circ l}\right) \qquad (3)$$

The reference value of $\sigma_e = 38 \times 10^{-3}$ J m^{-2} was obtained from Barham et al.[16] Exclusion must be accompanied by a decrease in the degree of crystallinity (X_{cr}) with increasing X_{HV} since it becomes increasingly difficult to find long uninterrupted sequences of crystallizable HB counits even at low X_{HV}. The equilibrium (X_{cr}) was measured by WAXD for samples ranging from 0 to 21 mol % HV, as shown in Figure 2 (the data shown are for crystallization at an undercooling of ΔT = 60 °C for all samples). Under no conditions was the degree of crystallinity for PHB/V less than 53%, whether crystallization started from the melt or from the glassy state. Crystallinity measured by DSC, i.e. $\Delta H_m(X_{HV})/\Delta H_m{}^\circ$, over the same composition range decreases significantly (see Figure 2). The Eby–Sanchez model explains the discrepancy between measurement of the crystallinity by X-ray versus by calorimetry. A fundamental assumption of the inclusion model is that a drop in the observed heat of fusion, $\Delta H_m(X_{HV})$, from $\Delta H_m{}^\circ$ is given by

$$\frac{\Delta H_m(X_{HV})}{X_{cr}} = \Delta H_m{}^\circ - \Delta H_D X_{HV} - 2\frac{\Delta H_e}{l} \qquad (4)$$

i.e. the enthalpy varies with lamellar thickness (l) as well as with the defect heat of fusion (ΔH_D) the copolymer composition, X_{HV} and the excess enthalpy to form the end surface of the crystal, ΔH_e. To validate the use of DSC to measure crystallinity, $\Delta H_m/X_{cr}$ must be independent of X_{HV}. Yet, as predicted by the Sanchez–Eby

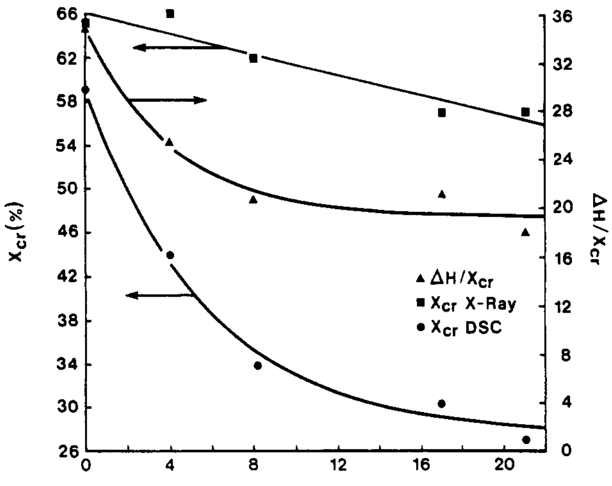

Figure 2. Crystallinity (X_{cr}) measured by WAXD and DSC as well as $\Delta H_m/X_{cr}$ as a function of copolymer composition (X_{HV}).

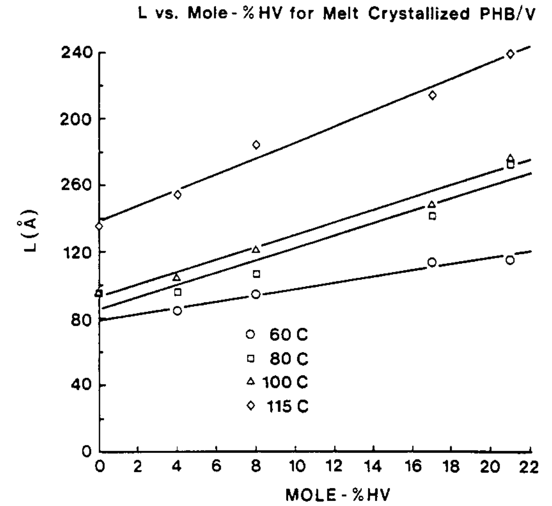

Figure 3. Bragg spacing (L) as a function of the copolymer composition for PHB/V samples crystallized at different crystallization temperatures, T_c.

treatment, $\Delta H_m/X_{cr}$ is clearly not constant, but decreases as X_{HV} increases (see Figure 2). Thus, in the case of PHB/V a decrease in the observed heat of fusion is not due to a decrease in crystallinity, but is rather due to changes in the crystalline structure, such as increased defects, smaller crystallites, and generally greater disorder within the region.

The lamellar thickness of PHB/V samples was measured by SAXS to determine the relationship between l and copolymer composition, as well as between l and the crystallization temperature (T_c). Figure 3 shows that for PHB/V at a given crystallization isotherm the Bragg spacing increases with increasing X_{HV}. Increasing T_c causes an increase in the positive slope of this curve, as well as shifting the intercept to a higher initial l value. The data support the often-discussed inverse relationship between lamellar thickness and the degree of undercooling, $\Delta T = T_m - T_c$. Copolymers such as PHB/V are interesting cases since the degree of undercooling is affected not only by changing T_c but also by varying copolymer composition due to the melting point depression. This relationship, which is not generally discussed when dealing with copolymers, was predicted by the Sanchez–Eby treatment.

Better insight into the applicability of the uniform inclusion model can be gained by using a plot of T_m vs $1/l$.

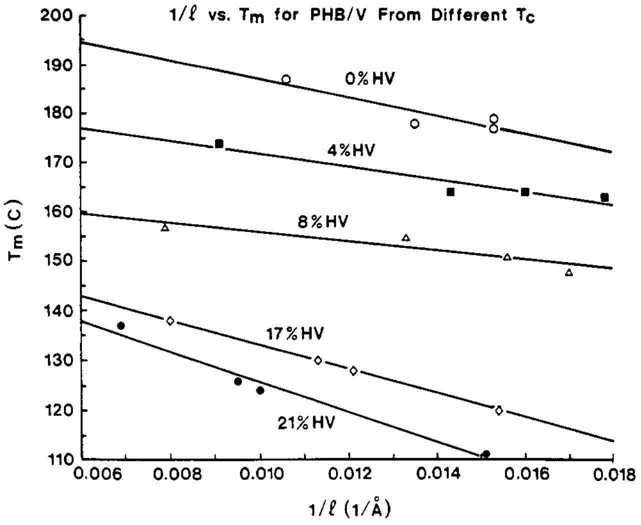

Figure 4. Variation in melting temperature (T_m) as a function of the inverse lamellar thickness ($1/l$) for 0, 4, 8, 17, and 21 mol % HV samples of PHB/V.

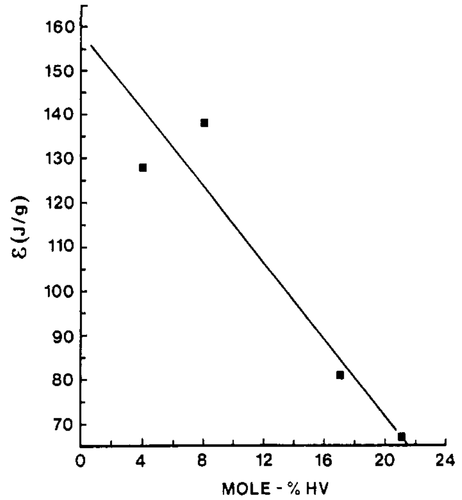

Figure 5. Variation in defect free energy (ϵ) as a function of HV content of PHB/V copolymers (X_{HV}).

Table I
Surface Free Energy (σ_e) and Defect Free Energy (ϵ) for Various Compositions of PHB/V

X_{HV}	σ_e, J m^{-2} × 10^3	ϵ, J g^{-1}
0	33 ± 3	
4	28 ± 4	128
8	24 ± 3	138
17	45 ± 4	81
21	60 ± 5	67

For example, if equation 3 is rearranged such that

$$T_m = T_m^\circ\left(1 - \frac{\epsilon}{\Delta H_m^\circ}X_{HV}\right) - \frac{2\sigma_e}{\Delta H_m^\circ}T_m^\circ\,(1/l) \qquad (5)$$

then the final term describes a constant slope regardless of X_{HV}. The data in Figure 4 reveal that for low X_{HV} the slope is constant. However as X_{HV} approaches 17% the slope is increasingly negative, indicating a greater value of $\sigma_e/\Delta H_m^\circ$. Furthermore, ϵ calculated from the intercepts decreases significantly with increasing X_{HV}, as shown in Table I and Figure 5. Since ϵ is expected to approach zero if the comonomers are perfectly interchangeable in the lattice, the diminishing ϵ with X_{HV} reveals that inclusion is favored with increasing HV counit composition.

Clearly one is seeing the results of the delicate balance between crystal packing energy, measured here by ϵ, and the surface free energy. Figure 4 (T_m vs $1/l$) is better interpreted as the change in melting temperature with the surface-to-volume ratio (s/v) of the crystalline phase. The steeper slopes with increasing X_{HV} suggest a more significant role of the surface free energy at higher HV compositions. This is in conjunction with the data from Figure 3 in which increasing copolymer composition at a given T_c leads to thicker crystals (or lower s/v). Interestingly, previously reported studies of PHB/V[7] show that increasing X_{HV} significantly slows the rate of crystallization. PHB can reach full crystallinity in a matter of minutes, whereas the 21% HV sample may require several weeks to reach its maximum under the same conditions. Thus, the rate of crystallization ultimately determines the morphology.

The trend in slopes shown in Figure 4 is different from those reported by Mitomo[8] for solution grown PHB/V crystals where values of $\sigma_e/\Delta H_m^\circ$ are relatively unvarying at low X_{HV}, but decrease at higher copolymer composition. The differences between solution grown and melt crystallized samples are not surprising considering the contrast in $\sigma_e/\Delta H_m^\circ$ reported by Marchessault et al.[18] between solution-grown PHB crystals and melt-crystallized oriented fibers. These differences are in keeping with what is known about the contrasting chain folding mechanisms and the role of entanglements between the two crystal types.[19,20]

4. Conclusions

The results of thermodynamic analysis of PHB/V based on uniform inclusion reveal that the inclusion of HV counits in the PHB-type lattice increases with increasing X_{HV} and the defect energy (ϵ) decrease correspondingly. This implies that the number of defects in the crystalline region also increases. Despite such changes in the crystalline microstructure, crystallinity is maintained with increasing copolymer composition. For isothermal crystallization this corresponds to increases in lamellar thickness with X_{HV}.

The copolymer exclusion model is inappropriate to describe the melting point depression of PHB/V since it relies on the exclusion of counits (an entropic effect) and does not consider the specific interaction, or enthalpic effect, between comonomers. Deviations from experimental data can not be ascribed to the effect of finite crystallite size. The uniform inclusion model accounts for the free energy involved in PHB/V cocrystallization, which is extensive for relatively similar comonomers such as HB and HV. Chain rearrangement during crystallization was not considered in this treatment. This is in keeping with the NMR results of Doi et al.,[21] showing that PHB and PHV not only are similar in their crystalline forms but also take on similar conformations in solution.

The uniform inclusion model utilized here is a useful simplification of a rather complex crystallization process. For example, it provides no a priori explanation of the differences in the surface free energy observed for the different copolymer compositions, nor does it take account of partitioning of counits between phases which would lead to a broadening of the melting endotherm in the midrange of composition.[22] The keys to gaining a clear picture of the thermodynamics of the melting point depression, i.e. the role of chain rearrangement during PHB/V crystallization, are (1) quantify the degree of inclusion and (2) elucidate the effect of inclusion on the crystalline microstructure. For example, solid-state NMR has shown that the HV ethyl group can exist in three different chemical environments: within the amorphous phase, within the PHV crystalline phase, and within the PHB crystalline phase.[6] Furthermore, the ethyl side group in PHV can exist in two possible conformational positions. More intensive studies utilizing NMR, SAXS, density measurements, high-resolution electron microscopy, or small-angle neutron scattering of selectively deuterated samples are critical for describing this cocrystallization phenomenon.

References and Notes

(1) Lemoigne, M. *Ann. Inst. Past.* **1925**, *39*, 144.

(2) Dawes, E. A.; Senior, P. J. *Adv. Microbiol. Physiol.* **1973**, *10*, 135.

(3) (a) Holmes, P. A.; Wright, L. F.; Collins, S. H. *Eur. Pat. Appl. 54,459*, 1981. (b) Wallen, L. L.; Rohwedder, W. K. *Environ. Sci. Technol.* **1974**, *8*, 576.

(4) Doi, Y.; Tamaki, A.; Kunioka, M.; Soga, K. *Appl. Microbiol. Biotechnol.* **1988**, *28*, 330.

(5) Ellar, D.; Lundgren, D. G.; Okamura, K.; Marchessault, R. H. *J. Mol. Biol.* **1968**, *35*, 489.

(6) Bluhm, T. L.; Hamer, G. K.; Marchessault, R. H.; Fyfe, C. A.; Veregin, R. P.; *Macromolecules* **1986**, *19*, 2861.

(7) Bloembergen, S.; Holden, D. A.; Hamer, G. K.; Bluhm, T. L.; Marchessault, R. H. *Macromolecules* **1986**, *19*, 2865.

(8) Mitomo, H.; Barham, P. J.; Keller, A. *Sen-i Gakkaishi* **1986**, *42*, 589; Mitomo, H.; Barham, P. J.; Keller, A. *Polym. J.* **1987**, *19*, 1241.

(9) Marchessault, R. H.; Bluhm, T. L.; Deslandes, Y.; Hamer, G. K.; Orts, W. J.; Sundararajan, P. R.; Taylor, M. G.; Bloembergen, S.; Holden, D. A. *Makromol. Chem. Macromol. Symp.* **1988**, *19*, 235.

(10) Flory, P. J. *Trans. Faraday Soc.* **1955**, *51*, 848.

(11) Sanchez, I. C.; Eby, R. K. *J. Res. Natl. Bur. Stand. Sect. A* **1973**, *77*, 353.

(12) Sanchez, I. C.; Eby, R. K. *Macromolecules* **1975**, *8*, 638.

(13) Gaddy, G. D.; Eby, R. K. *Polym. Commun.* **1990**, *31*, 298.

(14) Bloembergen, S.; Holden, D. A.; Marchessault, R. H. *Am. Chem. Soc., Div. Polym. Chem. Polym. Prepr.* **1988**, *29*, 594.

(15) (a) Alexander, L. E. *X-ray Diffraction Methods in Polymer Science*; Wiley: New York, NY, 1969; Chapter 3. (b) Ruland, W. *Acta Crystallogr.* **1961**, *14*, 1180.

(16) Barham, P. J.; Keller, A.; Otun, E. L.; Holmes, P. A. *J. Mater. Sci.* **1984**, *19*, 2781.

(17) (a) Gutzler, F.; Wegner, G. *Colloid Polym. Sci.* **1980**, *258*, 776. (b) Kalepky, U.; Fischer, E. W.; Herchenroeder, P.; Schelten, J.; Lieser, G.; Wegner, G. *J. Polym. Sci., Polym. Phys.* **1979**, *17*, 2117. (c) Roe, R. J.; Gieniewski, C. *Macromolecules* **1973**, *6*, 212.

(18) Marchessault, R. H.; Coulombe, S.; Morikawa, H.; Okamura, K.; Revol, J. F. *Can J. Chem.* **1981**, *59*, 38.

(19) Flory, P. J.; Yoon, D. Y. *Nature* **1978**, *272*, 226.

(20) Sadler, D. M.; Keller, A. *Science* **1979**, *203*, 263.

(21) Doi, Y.; Kunioka, M.; Nakamura, Y.; Soga, K. *Macromolecules* **1986**, *19*, 2860.

(22) Allegra, G. *Am. Chem. Soc., Div. Polym. Chem. Polym. Prepr.* **1988**, *29*, 615.

Registry No. PHB/V (copolymer), 80181-31-3; $H_3C(CH_2)_3CO_2H$, 109-52-4; $H_3C(CH_2)_2CO_2H$, 107-92-6.