

of weight loss in TGA. The fragmentation of the PB chain has now become predominant.<sup>4</sup> These explanations have been offered by us in ref 2.

When the luminescence emissions of the two random copolymers are compared with those of PB, we see that the intensity at a given temperature decreases with decreasing butadiene content. Furthermore, the inflection points shift to about 130 °C (Figure 1). These inflection temperatures again agree approximately with the onsets of DSC exotherm and of weight gain in TGA. We note that a shift in the inflection temperature with composition has also been observed in compatible mixtures of PS and poly(vinyl methyl ether).<sup>2</sup> At higher temperatures, the plateau or the broad maximum reached in the luminescence curves at 155 to 160 °C likewise coincides reasonably well with weight loss data.

For the block copolymer, the DSC and TGA results resemble closely those of polybutadiene. The luminescence curves of the two polymers are indistinguishable except at temperatures above 135 °C. The similarity between the luminescent behavior of the block polymer and the polybutadiene suggests that the surface composition of the block copolymer resemble that of polybutadiene.

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## Effect of Main-Chain Length and Nature of Side Chain on Specific Heat Variations of Polypeptides

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Specific heat measurements at low temperatures, where only the acoustic modes are appreciably excited thermally, can give valuable information about the solid-state structure of macromolecules. By this method the conformation of polypeptides can be ascertained<sup>2-5</sup> and, in particular, an estimate of the proportion of  $\alpha$  to  $\beta$  structure may be obtained.<sup>6</sup>

In this context, we have<sup>2,3</sup> recently demonstrated that L-valine, L-alanine, and tri(L-alanine) present a specific heat variation characteristic of a tridimensional molecular solid which is correctly fit by the Kitaigorodskii empirical rule.<sup>7</sup> For the L-valine and L-alanine homopolypeptides in the  $\beta$  form the predicted three- and two-dimensional vibrational behaviors, due to the intermolecular hydrogen bondings responsible for the sheet structure, were observed. For the  $\alpha$  form of poly(L-alanine), conversely, a one-dimensional pattern at higher temperature was shown, since each polypeptide chain vibrates separately. The Debye-Tarasov model<sup>8</sup> accounts well for the experimental results of all polypeptides.

In this note we report the specific heat variations of two monodispersed homooligopeptides of the L-valine and L-alanine series, namely, *t*-Boc-(L-Val)<sub>6</sub>-OMe and *t*-Boc-(L-Ala)<sub>6</sub>-OMe (*t*-Boc = *tert*-butoxycarbonyl, OMe = methoxy). We hoped the intermediate molecular weight

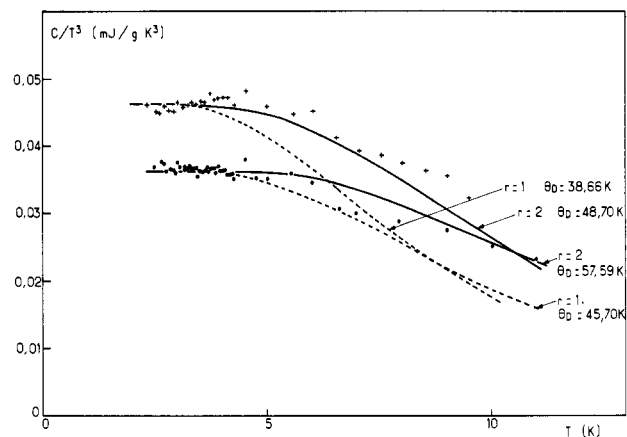


Figure 1. Specific heat divided by the cube of temperature vs. temperature for *t*-Boc-(L-Val)<sub>6</sub>-OMe (+) and *t*-Boc-(L-Ala)<sub>6</sub>-OMe (●). Dashed lines represent the standard Debye law ( $r = 1$ ) and solid lines the Kitaigorodskii model ( $r = 2$ ).

of the two peptides would allow us to obtain deeper insight into the effect of the main-chain length on the long-range order of polypeptides in the solid state.<sup>2</sup> In addition, comparison of the identical main-chain-length homopolypeptides derived from L-valine, containing a  $\beta$ -branched side chain, and L-alanine, characterized by a small and linear side chain, would allow the effect of the bulkiness of the lateral group of the constituent amino acid residue to be investigated.<sup>3</sup>

The details of the synthesis of *t*-Boc-(L-Val)<sub>6</sub>-OMe and *t*-Boc-(L-Ala)<sub>6</sub>-OMe are reported in ref 9 and 10, respectively. Rigorously monodispersed, chemically and optically pure, solvent-free, and well-crystallized homooligopeptides were obtained. The samples present a cross- $\beta$  structure with a defined unit cell, as shown by recent crystallographic work.<sup>11,12</sup> The heat capacity was measured between 2 and 10 K by a standard adiabatic method on a small polycrystalline sample as previously described.<sup>2</sup> The two hexapeptides were compressed to give a pellet which was placed on an alumina disk. The heat capacity of the experimental set up, i.e., alumina disk, germanium thermometer, constantan pressure gauge used as a resistor, and GE 7031 glue, was determined in a previous experiment. The heat capacity of the compounds was obtained by difference between the full and the empty cells which is around 50% of the empty cell with a mean accuracy of about 5%. The two hexapeptides after compression and thermal treatment are still in the  $\beta$  form,<sup>13</sup> as verified by infrared absorption spectroscopy.

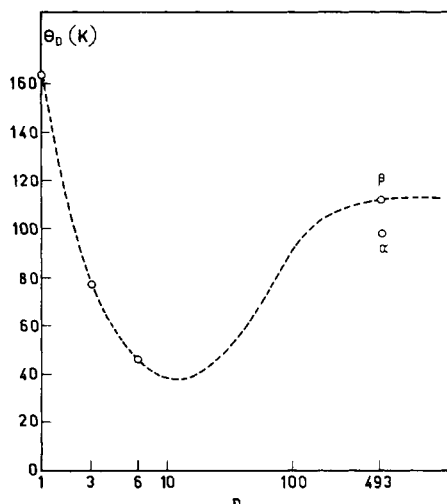
The experimental results in the form of the usual  $C_p/T^3 = f(T)$  plot are presented in Figure 1. In both cases a broad maximum followed by a monotonous decrease is found. At low temperatures the Debye law for tridimensional phonons gives the standard relation

$$C = \frac{12}{5} R \pi^4 (T/\theta_D)^3$$

where  $C$  is the heat capacity per mole,  $R$  is the ideal gas constant,  $r$  is the number of vibrators, and  $\theta_D$  is the Debye temperature.

To fit the experimental data we can employ the usual Debye approximation for atomic solids which takes into consideration the acoustic modes only ( $r = 1$ ; see Figure 1).

However, to get a better fit we can apply the Kitaigorodskii molecular method, which takes into account six degrees of freedom for each molecule, three translations, and three rotations, approximated by the same Debye function, with the number of vibrators  $r = 2$  per unit cell. As already shown for small peptides a better agreement



**Figure 2.** Debye temperatures  $\theta_D$  (for  $r = 1$ ) vs.  $n$  (the chain length) for the L-alanine series.  $\alpha$  and  $\beta$  refer, respectively, to the  $\alpha$ -helical structure and the  $\beta$ -pleated-sheet form of poly(L-alanine). The dashed line is simply an interpolation.

is found (Figure 1), whereas the Debye-Tarasov model, which introduces a vibrational anisotropy, does not work, as demonstrated in polypeptides.<sup>2,3</sup>

Now we can make a comparison of the characteristic vibrational temperatures found for the two series of compounds, namely, the alanine and the valine amino acids and peptides.

The ratio of the Debye temperatures  $k = \theta_{D_{Val}}/\theta_{D_{Ala}}$  gives the following: amino acids,<sup>3</sup>  $k = 0.84$ ; hexapeptides,  $k = 0.84$ ; polypeptides ( $\beta$  sheets),<sup>3</sup>  $k = 0.74$ .

Thus, the hexapeptides are homologous with the amino acids in that they are isomorphous molecular solids with a Debye temperature smaller for the valine series because of a steric effect due to the bulky isopropyl group. This result has to be compared with those from vacuum-ultraviolet circular dichroism<sup>14</sup> and infrared absorption<sup>13,15</sup> studies which have already shown that in the  $t$ -Boc-(L-X)<sub>n</sub>-OMe (X = Ala and Val) series in the solid state the  $\beta$  conformation is well developed at the alanine tripeptide ( $n = 3$ ) and its development at the pentapeptide ( $n = 5$ ) is essentially complete; conversely, the  $\beta$  conformation is fully obtained in the valine series only at the heptapeptide ( $n = 7$ ).

This apparent discrepancy can be explained in the following way: the spectroscopic observations on small homopeptides give information about the local order in these molecular solids, whereas in specific heat experiments only the long-range order is detected. At very low temperatures only the long-wavelength acoustic phonons are excited and we detect structural details of the order of these wavelengths. Conversely, spectroscopic experiments<sup>15</sup> indicate the presence of new internal vibrations in the hexapeptides. Since the number of vibrational degrees of freedom is constant for a given solid, we have a progressive change from the molecular acoustical modes to those of polymerically associated forms.

This fact is confirmed by the Debye temperature variation vs. the number of peptide units for the L-alanine series (Figure 2). The  $\theta_D$  values have been calculated by taking into consideration the acoustical phonons only. On the basis of all the results that we have collected on this series (see also ref 2 and 3), we conclude that there exists a minimum in the Debye temperature for an oligopeptide of higher rank. This means that the unit entropy goes through a maximum value on going from the molecular solids to the polymerically associated states because a more

disordered state exists between them, around the hexapeptide level. To observe a new increasing order between peptide chains (characterized by a slightly different value of the given ratio) one should examine an oligopeptide with  $n > 12$ . It is of interest to mention that  $n \approx 12$  is the critical chain length for  $\alpha$ -helix formation in the solid state for homopeptides derived from L-alanine, as shown by Fujie et al. by infrared absorption and X-ray diffraction techniques.<sup>16</sup> As pointed out above, the normal modes associated with the polymerically associated form become predominant around that stage of chain length.

In conclusion, the present specific heat measurements at low temperatures have confirmed the influence of a bulky lateral group on the cohesive energy of small peptides together with a similar vibrational behavior of these homopeptides. Furthermore, by analyzing the Debye temperatures variation vs. chain length in the alanine series we have shown that a polymerically associated structure may be present for the dodecapeptide: such a prediction has to be verified experimentally.

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## Thin-Layer Chromatographic Comparison between Deuterated and Hydrogenous Polystyrenes

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Thin-layer chromatography (TLC) is a powerful method for separating molecules by the small structural differences which are often difficult to detect by other methods. Thus far, it has been successfully applied to macromolecules having differences in molecular weight, stereoregularity, microstructure, terminal group, and composition and sequential arrangement in copolymers.<sup>1</sup> It is then tempting to ask whether TLC is capable of separating isotopically different polymers.

Recently, extensive use of the deuterium-labeling techniques has been made in neutron scattering experiments.<sup>2-4</sup> In most cases, the scattering data are explained by assuming deuteration to give no thermodynamic effects on the system. On the other hand, there have already been several observations showing that deuterio and protio systems are not identical. Strazielle and Benoit<sup>5</sup> have