Heterotactic Enthalpic Interactions of Amino Acids with Butanol in Aqueous Solutions

Yan Zhu,[†,‡] Li Yu,*[,†] and Xianhong Pang[‡]

Key Laboratory of Colloid and Interface Chemistry, Ministry of Education (Shandong University), Jinan, 250100, China, and Department of Chemistry and Chemical Engineering, Taishan Medical College, Taian, 271016, China

The enthalpies of mixing of L-serine and L-threonine with butanol and their respective enthalpies of dilution in aqueous solutions at 298.15 K and those of amino acids (glycine, L-alanine, L-serine, L-threonine, and L-proline) with butanol in aqueous solutions at 310.15 K were determined as a function of the mole fraction by flow microcalorimetric measurements. These experimental results have been analyzed to obtain the heterotactic enthalpic interaction coefficients (h_{xy}) according to the McMillan−Mayer theory. The results obtained in the present work are compared with the data of our previous work at 298.15 K. It has been found that the h_{xy} coefficients between the amino acid molecules studied and butanol molecules in aqueous solutions at (298.15 and 310.15) K are all positive. The h_{xy} coefficients at 310.15 K are more positive than those of 298.15 K for the same system studied. The results are discussed in terms of solute−solute interaction and solute−solvent interaction.

Introduction

The folding, structural stability, and dynamics of globular proteins are thought to be extensively controlled by the interactions of the macromolecule with water. Various added substances affect these interactions and consequently alter the structural stability of proteins.[1] Since proteins have a complex structure and show some intricate effects on their structure, investigations on various thermodynamic properties of amino acids and simple peptides in aqueous solutions of organic or inorganic substances are of current interest due to their importance in the better understanding of the nature and mechanisms taking place in biological cells.[2,3] Organic solvents strongly affect the solubility and denaturation of proteins.[4] There is considerable interest in aqueous solutions of the lower aliphatic alcohols because of their unique structural relationship to pure water.[5]

In our previous studies, the enthalpies of mixing of amino acids with 2-chlorethanol,[6] 1,2-ethanediols,[7] and 2-butanone[8] in aqueous solution were measured by the method of microcalorimetry. This present work reports the enthalpic interaction coefficients between L-serine, L-threonine, and butanol in aqueous solution at 298.15 K and those of various amino acids (glycine, L-alanine, L-serine, L-threonine, and L-proline) and butanol in aqueous solution at 310.15 K according to the McMillan−Mayer theory.[9] These coefficients reflect the sum of the enthalpic effects of interactions between the components in aqueous solutions. In addition, the results obtained in the present work together with those reported in an earlier paper,[10] about the enthalpic interaction of amino acids (glycine, L-alanine, and L-proline) with butanol at 298.15 K, have been used to investigate the difference between the enthalpic pairwise interaction coefficients of amino acids with butanol at different temperatures.

* Corresponding author. E-mail: ylmlt@sdu.edu.cn.
† Shandong University.
‡ Taishan Medical College.

Materials and Methods

Five amino acids, glycine (Gly), L-alanine (Ala), L-serine (Ser), L-threonine (Thr), and L-proline (Pro), were obtained from Shanghai Chemical Co., China, and used after recrystallization from a water−methanol mixture and dried in vacuum desiccators until their weights became constant. Analytical reagent grade butanol was used without further purification. The water used in the experiments was deionized, distilled, and degassed. Both the aqueous amino acid solutions and aqueous butanol solution were prepared by mass using a Mettler AE 200 balance with a precision of ± 0.0001 g. All the solutions were degassed and used within 12 h after preparation to avoid possible bacterial contamination.

The measurements of enthalpies of mixing and dilution were carried out with a flow microcalorimeter (2277 Thermal Activity Monitor, made in Sweden) at (298.15 and 310.15) K. The calorimeter has a high temperature control accuracy (± 0.001 K). The baseline stability (over a period of 24 h) of it is 0.2 μW. The solutions were pumped through the mixing-flow vessel of the calorimeter using a pair of LKB-2132 microperpex peristaltic pumps. The flow rates were determined from the mass of the samples delivered in 8 min. The variation of flow rates was less than 0.1 % both before and after a complete experiment. The relative mean deviation of the thermal power determined was 0.3 %, and that of the enthalpies of mixing and dilution was less than 1 %. The apparatus and procedure used were the same as those described in earlier work.[6−8]

The enthalpies of mixing and dilution can be treated to determine the enthalpic interaction coefficients based on McMillan−Mayer theory.[9] The excess enthalpy $H^E(m_x, m_y)$ of a solution containing two solute species x and y can be expressed as a virial expansion of solute molalities using the following equation

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$$H^{E}(m_x, m_y)/w_1 = H(m_x, m_y)/w_1 - h^{*}_{w} - m_x H^{\infty}_{x,m} - m_y H^{\infty}_{y,m} = h_{xx}m_x^2 + 2h_{xy}m_x m_y + h_{yy}m_y^2 + h_{xxx}m_x^3 + 3h_{xxy}m_x^2 m_y + 3h_{xyy}m_x m_y^2 + h_{yyy}m_y^3 + \ldots \quad (1)$$

where $H^{E}(m_x, m_y)$ and $H(m_x, m_y)$ represent the excess enthalpy and the total enthalpy of a solution containing m_x mol of x and m_y mol of y species in w_1 kilograms of water, respectively; h^{*}_{w} is the standard enthalpy of 1 kg of pure water; $H^{\infty}_{x,m}$ and $H^{\infty}_{y,m}$ are the limiting partial molar enthalpies of solutes x and y, respectively; and the various h_{ij} and h_{iij} are the interaction coefficients that represent the contribution of solute−solute interactions between pairs, triplets, and higher order interactions of solvated solute molecules in a binary solution.

The dilution enthalpies ΔH_{dil} ($J \cdot kg^{-1}$) are determined by measuring thermal power P (μW) and flow rates of solution and solvent (f_A and f_B, $mg \cdot s^{-1}$)

$$\Delta H_{dil} = P/(f_A + f_B - m_{x,i} M_x f_A) \quad (2)$$

where M_x is the mole mass of solute ($kg \cdot mol^{-1}$) and $m_{x,i}$ is initial molality ($mol \cdot kg^{-1}$).

The final molality m_x ($mol \cdot kg^{-1}$) may be calculated from the equation

$$m_x = m_{x,i} f_A/[f_B(m_{x,i} M_x + 1) + f_A] \quad (3)$$

The mixing enthalpy ΔH_{mix}($J \cdot kg^{-1}$) of aqueous x solution and aqueous y solution is calculated from the equation

$$\Delta H_{mix} = P^{*}/(f_x + f_y - m_{x,i} M_x f_x - m_{y,i} M_y f_y) \quad (4)$$

where P^{*} is the mixing thermal power (μW); $f_x f_y$ are the flow rates of solutions x and y; and $m_{x,i} m_{y,i}$ are the initial molalities of solutions x and y before mixing, respectively.

To facilitate the calculation, an auxiliary functionΔH^{*} is introduced

$$\Delta H^{*} = \Delta H_{mix} - \Delta H_{dil}(x) - \Delta H_{dil}(y) = H^{E}(m_x, m_y) - H^{E}(m_x) - H^{E}(m_y) \quad (5)$$

Therefore the equation for the heterotactic interaction coefficients can be evaluated for the combination of eqs 1 and 5

$$\Delta H^{*}/w_1 = 2h_{xy}m_x m_y + 3h_{xxy}m_x^2 m_y + 3h_{xyy}m_x m_y^2 + \ldots \quad (6)$$

If the mixing experiments are carried out at different values of m_x and m_y, then the pairwise and triplet enthalpic interaction coefficients can be evaluated.

Results and Discussion

Tables 1 and 2 list the heterotactic enthalpic interaction coefficients at (298.15 and 310.15) K, respectively, which have been calculated from eq 6 based on the calorimetric results. Since there are difficulties in the interpretation of the higher order coefficients due to the fact that they also contain pairwise interaction terms, the analysis is restricted to the pairwise coefficients h_{xy}. The enthalpic interaction coefficients represent a measure of interactions between two hydrated solutes and depend on the interactions between the solute molecules and the solvent water. With the aim to conveniently compare data with previous work[10] for amino acids (glycine, L-alanine, L-proline)/butanol in the aqueous solutions at 298.15 K, the trend of h_{xy} values of the two systems at (298.15 and 310.15) K is gathered in Figure 1.

By and large, the global effects between amino acids and butanol in the aqueous solutions reflect three superimposed processes. The first is the partial dehydration of the hydration shell of the amino acid zwitterions (endothermic process). The second is the partial dehydration of the hydration shell of butanol (endothermic process), and the third is the direct interaction between the molecules of amino acids and butanol, which plays the dominant role in the overall interaction process.

Since both the amino acid and butanol molecules have hydrophobic and hydrophilic groups, the direct interaction between them can be summarized as: (a) the hydrophobic−hydrophobic interaction (endothermic process, making positive contributions to h_{xy}); (b) the hydrophobic−hydrophilic interaction (endothermic process, making positive contributions to h_{xy}); (c) the hydrophilic−hydrophilic interaction (exothermic process, making negative contributions to h_{xy}).

As can be seen from Figure 1, the experimentally observed positive values of h_{xy} at (298.15 and 310.15) K testify to the predominance of endothermic processes over the effect of direct interaction of amino acid with butanol molecules. Whether the experimental temperature is (298.15 or 310.15) K, h_{xy} values change in the same order as follows: h_{xy}(glycine) < h_{xy}(L-serine) < h_{xy}(L-alanine) < h_{xy}(L-proline) < h_{xy}(L-threonine). This indicates that the structure of amino acid molecules has a more important effect on the interaction between amino acid and butanol. It is the R-group that can be responsible for the observed variation trends of h_{xy} because the amino acid molecules have the same charged groups ($-NH_3^+$, $-COO^-$).

The direct interactions between glycine and butanol in aqueous solution include interaction types (b) and (c). Since interaction type (c) is dominant, the h_{xy} coefficient between

Table 1. Heterotactic Enthalpic Interation Coefficients between Amino Acids and Butanol in Aqueous Solution at 298.15 K

solutes x + y	h_{xy} / $J \cdot kg \cdot mol^{-2}$	h_{xxy} / $J \cdot kg^2 \cdot mol^{-3}$	h_{xyy} / $J \cdot kg^2 \cdot mol^{-3}$	R^{2a}	SD[b]
L-serine + butanol	720 ± 75[c]	5466 ± 1270	−5454 ± 1332	0.9983	1.41
L-threonine + butanol	836 ± 78	3161 ± 1317	−3083 ± 1374	0.9984	1.46

[a] Square of correlation coefficient. [b] Standard deviation. [c] The estimated deviation.

Table 2. Heterotactic Enthalpic Interation Coefficients between Amino Acids and Butanol in Aqueous Solution at 310.15 K

solutes x + y	h_{xy} / $J \cdot kg \cdot mol^{-2}$	$h_{xxy} \cdot 10^{-4}$ / $J \cdot kg^2 \cdot mol^{-3}$	$h_{xyy} \cdot 10^{-4}$ / $J \cdot kg^2 \cdot mol^{-3}$	R^{2a}	SD[b]
glycine + butanol	854 ± 138[c]	70.5 ± 63.6	−75.3 ± 67.8	0.9996	0.37
L-alanine + butanol	984 ± 349	149 ± 16.8	−159 ± 179	0.9966	1.21
L-serine + butanol	942 ± 294	20.6 ± 20.4	−2.24 ± 2.15	0.9988	0.77
L-threonine + butanol	1071 ± 320	−20.0 ± 14.5	2.06 ± 1.52	0.9989	0.84
L-proline + butanol	923 ± 258	−15.5 ± 12.9	1.62 ± 1.36	0.9993	0.67

[a] Square of correlation coefficient. [b] Standard deviation. [c] The estimated deviation.

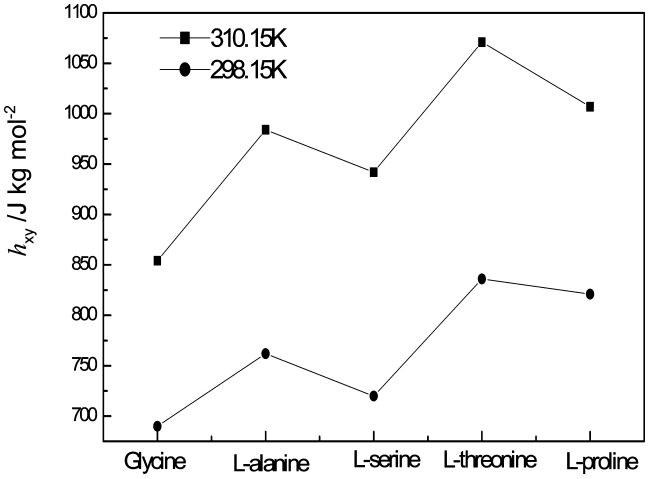

Figure 1. Comparisons between the values of the heterotactic enthalpic pairwise interaction coefficients of amino acids with butanol in aqueous solutions at 298.15 K and those at 310.15 K.

glycine and butanol is positive. Compared to glycine, for L-alanine and L-valine, one hydrogen atom on the α-carbon has been replaced by a methyl and an isopropyl, respectively, which results in the interaction (a) strengthening notably with the prolongation of nonpolar side chains. So the h_{xy} coefficients increase in the following sequence: h_{xy}(glycine) < h_{xy}(L-alanine) < h_{xy}(L-valine).

The hydroxyl group, which can participate in hydrogen bonding interaction, of amino acid can have a great influence on the h_{xy} values. L-Serine is similar to L-alanine except that it has an OH group replacing a hydrogen atom of the methyl group. The dominant role played by interaction (c) causes h_{xy}(L-serine) < h_{xy}(L-alanine).

As for L-serine, the $-CH_2OH$ group can be considered as a substitute for one hydrogen atom of the α-carbon of glycine. In butanol aqueous solutions, there exists h_{xy}(glycine) < h_{xy}(L-serine), which shows that the interactions (a) and (b) yield very strong interaction effects.

Thus, for the above series of amino acids, there exists the following rule: h_{xy}(glycine) < h_{xy}(L-serine) < h_{xy}(L-alanine) < h_{xy}(L-threonine).

L-Proline is a natural amino acid that has one pyrrole ring. Its special structure makes it important to the properties of polypeptide. The cyclic structure weakens the interactions (a) and (b) between L-proline and butanol molecules due to the steric effect. However, the L-proline molecule still shows a relative hydrophobicity. As can be seen from Figure 1, the values of h_{xy} coefficient for the interaction between L-proline and butanol in aqueous solution at (298.15 and 310.15) K are approximates to those of L-alanine at the two temperatures, respectively. This behavior may be interpreted as similarity of the hydrophobic behavior of L-proline to L-alanine. Evidences for this similarity are also found from the studies on amino acids/urea and amino acids/2-chlorethanol systems in aqueous solutions at 298.15 K. The h_{xy} values for the interactions between L-proline and urea[11] and 2-chlorethanol[6] in aqueous solutions at 298.15 K are also close to those of L-alanine.

It can be found that the changing trend is similar for the h_{xy} coefficient between each amino acid and butanol in aqueous solution at (298.15 and 310.15) K. However, the h_{xy} coefficient magnitude of the latter is more than that of the former. This can be interpreted as the result of the following two contributions. On one hand, the hydrophobic interaction (endothermic process) between amino acid and butanol molecules intensifies, and the relative strength of hydrogen bond interactions (exothermic process) between OH groups of amino acids and butanol molecules attenuates as temperature increases. On the other hand, the partial dehydration of the solvation layers for amino acid and butanol molecules enhances with increasing temperature, and the endothermic effect increases. Therefore, as a consequence of a different balance of the involved interactions for the amino acid/butanol system in the aqueous solution, the h_{xy} coefficient shows a more positive magnitude with the increase of temperature.

Supporting Information Available:

Tables S1 and S2. This material is available free of charge via the Internet at http://pubs.acs.org.

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