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Enthalpies of dilution of glycine, *l*-serine and *l*-valine in mixtures of water and *N*,*N*-dimethylformamide at 298.15 K

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Abstract

Enthalpies of dilution of glycine, *l*-serine and *l*-valine dissolved in mixtures of water and *N*,*N*-dimethylformamide (DMF) with mass percent of DMF 0, 5, 15, 25, 35 and 45, respectively, have been measured with an LKB 2277 Bio Activity Monitor at 298. 15 K. It was found that pairwise enthalpic interaction coefficients h_2 of glycine and *l*-serine decrease with increasing DMF content in mixed solvents, while the influence of DMF on h_2 values of *l*-valine is not obvious. © 2000 Elsevier Science B.V. All rights reserved.

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1. Introduction

Amino acids are monomers to constitute proteins. Solute–solute interactions are significant to investigate thermodynamic properties of amino acids solutions [1,2]. Correspondingly, the enthalpy of dilution of amino acids is very important. Studies on the enthalpy of dilution of amino acids focus on that in water [3–7]. The majority of proteins exist in aqueous mixed solvents containing many organic substances. They do not exist in pure water. Organic solvents affect strongly the solubility and denaturalization of proteins, and enzyme activity. The interactions occurring between solutes in water are different from that in mixed solvents. Aqueous solutions of *N*,*N*-dimethylformamide (DMF) is a model mixed solvent which represents an environment of protein's interiors [8]. It is worthwhile to explore the effect of changing DMF solvent medium on amino acid molecular interactions. Glycine is the most simple amino acid in nature. *l*-Serine is the amino acid with polar side-chain –CH_2OH. *l*-Valine is the amino acid with apolar side-chain –$C(CH_3)_2$. Based on the research of polar and apolar amino acids, it is well understand the effect of hydroxyl and alkyl group on the interactions of protein's interiors. The present work reports the enthalpic interactions of these representative amino acids in mixtures of water and DMF by using LKB 2277 Bio Activity Monitor.

2. Experimental

Analytical reagent grade glycine, *l*-serine and *l*-alanine were recrystallized two times from ethanol + -water mixtures. The materials were used after drying at 100°C for 6 h and then in vacuo over silica gel at

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room temperature for a minimum of 24 h. Analytical regent grade DMF was dried by storage over molecular sieves 0.4 nm for 2 days and then was distilled under reduced pressure. The refracting prism of the collected cut was $n_D^{25} = 1.4282$ (1.42817 in the work of Riddick and Bunger [9]). Freshly prepared twice-distilled water was used in all experiments. Mixtures of water and DMF were prepared by mass. The mass percentage of DMF in these mixtures ranged in 10% increments from 5% to 45%. Solutions of amino acids were made by mass on the molality concentration scale.

The enthalpies of dilution of glycine, *l*-serine and *l*-valine in mixtures of water and DMF were measured with an LKB 2277 Bio Activity Monitor operating at 298.15 K. The solutions were pumped regularly into the mixed cell of microcalorimeter using two LKB BROMMA 2132 pumps. The total flow rate of solutions through the calorimeter was kept fixed at ca. 12.5 mg s^{-1}. Errors in the determinations of the molar enthalpies of dilution were estimated to be < 1%.

3. Results and discussion

The molar excess enthalpy of a solution with molality m can be written as a power series of m with coefficients h_n by [10–13]

$$H_m^E = H^E/m = L_{2\Phi} = h_2 m + h_3 m^2 + h_4 m^3 + \cdots \quad (1)$$

where $L_{2\Phi}$ is the relative apparent molar enthalpy. The enthalpic interaction coefficients h_n which are related to the McMillan–Mayer B coefficients measure the enthalpic interactions, mediated by the solvent, occurring between pairs, triplets and higher order multiplets of solute species. The molar enthalpy of dilution $\Delta_{dil} H_m$ in terms of the excess enthalpy at initial molality m_i and final molality m_f. is

$$\Delta_{dil} H_m = H^E_{m,m_f} - H^E_{m,m_i} \quad (2)$$

Combining Eqs. (1) and (2) yields

$$\Delta_{dil} H_m = h_2(m_f - m_i) + h_3(m_f^2 - m_i^2) + h_4(m_f^3 - m_i^3) + \cdots \quad (3)$$

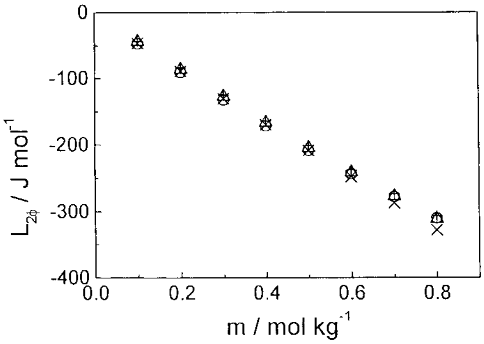

Fig. 1. Relative apparent molar enthalpy of glycine in water plotted against the molality of glycine at 298.15 K: (×) this work; (O) Sturtevant [6]; (Δ), Gucker et al. [7]; (+) Humphrey et al. [4].

In an actual experiment, $\Delta_{dil} H_m$ and m_f are determined by measuring heat power W and flow rates of solution and solvent (f_u and f_v):

$$m_f = m_i f_u / [f_v(m_i M + 1) + f_u] \quad (4)$$

$$\Delta_{dil} H_m = -W/(f_u c_i) \quad (5)$$

where c_i is the initial molar amount of solute (mol) in 1 kg solution.

Tables 1–3 give the experimental results of enthalpies of dilution. The dilutions of glycine and *l*-serine in mixtures of water and DMF are endothermic processes (Tables 1–2). The dilutions of *l*-valine in mixtures of water and DMF are exothermic processes (Table 3).

Relative apparent molar enthalpy of glycine in water obtained in this work is in good agreement with those of Sturtevant [6], Gucker et al. [7] and Humphrey et al. [4] (see Fig. 1).

Fig. 2 shows the variation of pairwise enthalpic interaction coefficients h_2 of three amino acids with

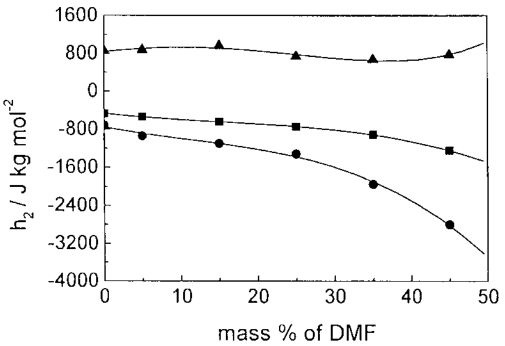

Fig. 2. Variation of pairwise enthalpic interaction coefficients h_2 of amino acids with DMF content in mixed solvents at 298.15 K: (■) gly; (●) *l*-ser; (▲) *l*-val. The h_2 values of *l*-serine and *l*-valine in water can be found in [4].

Table 1
Enthalpies of dilution of glycine in mixtures of water and DMF at 298.15 K

m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})	m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})
0% DMF					
0.4001	0.1304	111.39	0.3000	0.1479	64.14
0.4001	0.1702	94.94	0.3000	0.1975	43.12
0.4001	0.1968	83.14	0.2499	0.0821	72.49
0.4001	0.2239	71.84	0.2499	0.1232	54.62
0.4001	0.2635	55.36	0.2499	0.1650	36.53
0.3500	0.1146	98.87	0.2001	0.0659	59.58
0.3500	0.1722	73.84	0.2001	0.0988	44.74
0.3500	0.2302	49.57	0.2001	0.1322	30.04
0.3000	0.0983	85.77			
5% DMF					
0.4016	0.1975	91.55	0.2997	0.1467	71.13
0.4016	0.2640	61.07	0.2997	0.1966	47.63
0.4016	0.1302	123.30	0.2997	0.0973	95.76
0.4016	0.2240	78.85	0.2498	0.1224	60.81
0.4016	0.1695	104.41	0.2498	0.1642	40.71
0.3496	0.1710	81.77	0.2498	0.0810	81.34
0.3496	0.2296	54.54	0.2002	0.0983	50.04
0.3496	0.1132	109.68	0.2002	0.1317	33.62
0.3496	0.1948	70.14	0.2002	0.0652	66.73
0.3496	0.1475	92.32			
15% DMF					
0.4001	0.1958	107.49	0.2999	0.0976	112.76
0.4001	0.2625	72.19	0.2502	0.1226	71.20
0.4001	0.1300	146.39	0.2502	0.1648	48.37
0.3498	0.1716	95.73	0.2502	0.0816	96.20
0.3498	0.2299	64.14	0.2000	0.0983	59.30
0.3498	0.1138	128.24	0.2000	0.1315	39.80
0.2999	0.1471	84.13	0.2000	0.0653	78.57
0.2999	0.1970	56.79			
25% DMF					
0.3499	0.1712	112.72	0.2500	0.0813	114.97
0.3499	0.2293	77.72	0.2007	0.0984	70.97
0.3499	0.1133	155.01	0.2007	0.1320	48.10
0.2999	0.1464	100.48	0.2007	0.0652	95.16
0.2999	0.1968	68.34	0.1499	0.0735	53.99
0.2999	0.0961	136.43	0.1499	0.0986	37.32
0.2500	0.1226	85.86	0.1499	0.0491	72.02
0.2500	0.1644	57.80			
35% DMF					
0.3000	0.1480	122.10	0.2018	0.0665	115.55
0.3000	0.1982	82.97	0.2000	0.0990	86.98
0.3000	0.0986	168.31	0.2000	0.1322	58.81
0.2508	0.1240	102.71	0.2000	0.0659	117.00
0.2508	0.1658	71.75	0.1000	0.0496	45.56
0.2508	0.0827	141.57	0.1000	0.0663	31.50
0.2018	0.0999	87.95	0.1000	0.0330	60.47
0.2018	0.1334	59.26			
45% DMF					
0.2000	0.0992	111.68	0.1798	0.1013	86.78
0.2000	0.1326	76.73	0.1500	0.0745	88.32

Table 1 (*Continued*)

m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})	m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})
0.2000	0.0661	151.81	0.1500	0.0992	60.54
0.2000	0.1128	94.54	0.1500	0.0496	118.44
0.1798	0.0893	103.44	0.1000	0.0497	60.99
0.1798	0.1191	69.53	0.1000	0.0663	41.98
0.1798	0.0593	136.69	0.1000	0.0331	81.82

Table 2
Enthalpies of dilution of *l*-serine in mixtures of water and DMF at 298.15 K

m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})	m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})
5% DMF					
0.4004	0.1988	153.45	0.3003	0.0992	161.30
0.4004	0.2646	100.55	0.2500	0.1241	102.01
0.4004	0.1311	206.20	0.2500	0.1656	66.82
0.3496	0.1732	136.78	0.2500	0.0827	137.09
0.3496	0.2314	88.59	0.1999	0.0993	83.92
0.3496	0.1148	180.86	0.1999	0.1324	54.17
0.3003	0.1488	119.23	0.1999	0.0661	112.16
0.3003	0.1988	78.67			
15% DMF					
0.4001	0.2032	150.34	0.3005	0.1519	133.68
0.4001	0.2700	115.57	0.3005	0.2021	92.36
0.4001	0.1364	230.52	0.3005	0.1023	182.61
0.3500	0.1765	151.40	0.2502	0.1263	113.36
0.3500	0.2349	104.31	0.2502	0.1683	79.51
0.3500	0.1192	208.53	0.2502	0.0851	157.53
25 %DMF					
0.3500	0.1754	176.83	0.2497	0.1254	142.59
0.3500	0.2339	126.62	0.2497	0.1671	99.25
0.3500	0.1180	252.86	0.2497	0.0843	194.75
0.2988	0.1501	162.45	0.2000	0.1003	115.41
0.2988	0.2000	111.70	0.2000	0.1340	82.77
0.2988	0.1008	224.86	0.2000	0.0676	161.16
35% DMF					
0.2999	0.1506	209.39	0.2004	0.1001	150.08
0.2999	0.2005	143.50	0.2004	0.1339	103.97
0.2999	0.1006	291.77	0.2004	0.0671	210.68
0.2503	0.1249	182.23	0.1501	0.0750	118.63
0.2503	0.1674	122.88	0.1501	0.1002	82.69
0 2503	0.0839	247.80	0.1501	0.0505	164.26
45% DMF					
0.1501	0.0752	149.75	0.1501	0.1003	106.83
0.1501	0.0504	209.09	0.1021	0.0512	111.22
0.1021	0.0683	78.09	0.1021	0.0344	145.11
0.0797	0.0399	91.79	0.0797	0.0533	63.02
0.0797	0.0269	124.78			

Table 3
Enthalpies of dilution of *l*-valine in mixtures of water and DMF at 298.15 K

m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})	m_i (mol kg^{-1})	m_f (mol kg^{-1})	$\Delta_{dil} H_m$ (J mol^{-1})
5% DMF					
0.3504	0.1711	−166.51	0.2501	0.0817	−153.69
0.3504	0.2296	−108.83	0.1997	0.0979	−90.72
0.3504	0.1127	−211.58	0.1997	0.1316	−61.30
0.2995	0.1469	−137.08	0.1997	0.0652	−121.47
0.2995	0.1967	−93.05	0.1495	0.0737	−67.35
0.2995	0.0976	−182.31	0.1495	0.0988	−45.66
0.2501	0.1229	−113.68	0.1495	0.0490	−89.16
0.2501	0.1645	−77.63			
15% DMF					
0.2994	0.1467	−147.38	0.2001	0.0655	−127.47
0.2994	0.1968	−98.64	0.1501	0.0740	−72.37
0.2994	0.0972	−194.98	0.1501	0.0993	−49.05
0.2495	0.1227	−120.12	0.1501	0.0494	−95.34
0.2495	0.1644	−81.60	0.1001	0.0496	−48.61
0.2495	0.0813	−161.68	0.1001	0.0662	−32.53
0.2001	0.0986	−96.41	0.1001	0.0328	−64.23
0.2001	0.1318	−65.49			
25% DMF					
0.2486	0.1226	−118.85	0.1806	0.0595	−111.47
0.2486	0.1641	−80.08	0.1499	0.0742	−69.66
0.2486	0.0814	−155.97	0.1499	0.0992	−46.91
0.1999	0.0988	−96.39	0.1499	0.0494	−90.29
0.1999	0.1321	−64.23	0.1002	0.0496	−46.15
0.1999	0.0657	−125.18	0.1002	0.0664	−30.79
0.1806	0.0891	−86.23	0.1002	0.0331	−58.26
0.1806	0.1194	−57.36			
35% DMF					
0.1600	0.0792	−77.27	0.1202	0.0397	−62.03
0.1600	0.1059	−51.00	0.0999	0.0496	−39.46
0.1600	0.0528	−98.57	0.0999	0.0662	−25.85
0.1401	0.0695	−64.14	0.0999	0.0330	−49.26
0.1401	0.0927	−42.08	0.0811	0.0403	−29.26
0.1401	0.0462	−83.00	0.0811	0.0539	−21.07
0.1202	0.0596	−47.42	0.0811	0.0268	−38.54
0.1202	0.0798	−31.39			
45% DMF					
0.0997	0.0496	−27.97	0.0997	0.0660	−20.67
0.0793	0.0395	−21.03	0.0793	0.0449	−16.91
0.0793	0.0527	−14.26	0.0793	0.0341	−21.98
0.0793	0.0263	−25.46	0.0793	0.0475	−15.29

DMF content in mixed solvents. The h_2 values of glycine and *l*-serine are negative and they decrease with increasing DMF content in mixed solvents, while h_2 values of *l*-valine are positive on which the influence of DMF is not obvious. The enthalpy of interaction between amino acid molecules is very solvent dependent. For zwitterionic amino acids in solution, the configuration likely to contribute most to the pairwise interaction is that in which the molecular association is side-on with the positively charged amino group and its associated cosphere of one amino acid interacting with the solvated negatively charged carboxyl group of the second molecule [14,15]. This solvated dipolar interaction is very depended on the surrounding solvent property. DMF molecules interact mainly with positively charged centers in solutions because of the steric effects of the two methyl groups in DMF molecules, influencing the pairwise interactions of amino acids. For glycine, which has no side-chain, the strong dipolar interactions are dominate in the pairwise interaction of glycine and are expected to result in a negative contribution to h_2 [15,16]. With increasing DMF content in mixed solvents, negatively charged centers of DMF molecules interact more strongly with positively charged centers of glycine molecules, contributing more bigger to h_2 values, which makes h_2 values decrease.

In the pairwise association of *l*-serine and *l*-valine, interactions involving the side-chains will be significant. For *l*-serine with side-chain functional group –OH that can participate in hydrogen bonding on pairwise interaction, the contribution to h_2 should be negative [16,17]. Hydrogen bonding attractions arise from contributions stemming from hydroxyl group–hydroxyl group, and hydroxyl group–zwitterionic head group interactions. Formation of hydrogen bonds is very dependent on the surrounding solvent. The hydrogen bonding interactions between *l*-serine molecules increase rapidly with increasing DMF content in mixed solvents. Hydroxyl groups assist in the formation of hydrogen bonds in interior protein molecules, which makes protein structure stabilized.

For *l*-valine with apolar side-chain $–C(CH_3)_2$, positive contributions to h_2 arise from both the interactions between hydrated hydrocarbon chains [15,18] and those between the hydrated side-chains and the ionic groups [16,19]. The positive values of h_2 for *l*-valine with alkyl side-chain suggest that interactions involving the side-chains dominate over the zwitterion–zwitterion interactions. Interactions involving the side-chains and zwitterion–zwitterion interactions contribute to h_2 differently with increasing DMF content in mixed solvents, resulting in little change of h_2 of *l*-valine. The value of h_2 is maximum at 15% of DMF in the "rich" water region, and it is minimum at 35% of DMF in the region of rich DMF. Repulsing interactions arising from apolar alkyl group side-chains have little change in mixtures of water and DMF, but they increase with increasing alkyl groups [5]. Alkyl groups assist in the highly extension of peptide chains, making protein structure stabilized.

Acknowledgements

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References

[1] T.H. Lilley, R.H. Wood, J. Chem. Soc., Faraday Trans. I 76 (1980) 901.

[2] B.Y. Okamoto, R.H. Wood, P.T. Thompson, J. Chem. Soc., Faraday Trans. I 74 (1978) 1990.

[3] M. Sahayan, G.R. Hedwig, J. Chem. Thermodyn. 26 (1994) 361.

[4] R.S. Humphrey, G.R. Hedwig, I.D. Watson, G.N. Malcolm, J. Chem. Thermodyn. 12 (1980) 595.

[5] M.A. Gallardo, T.H. Lilley, H. Linsdell, S. Otin, Thermochim. Acta 223 (1993) 41.

[6] J.M. Sturtevant, J. Am. Chem. Soc. 62 (1940) 1879.

[7] F.T. Gucker, H.B. Pichard, W.L. Ford, J. Am. Chem. Soc. 62 (1940) 2698.

[8] T.H. Lilley, in: M.N. Jones (Ed.), Biochemical Thermodynamics, 2nd ed., Chapter 1, Elsevier, Amsterdam, 1988.

[9] J.A. Riddick, W.B. Bunger, Techniques of Chemistry, Vol. II, Organic Solvents, 3rd ed., Wiley, 1970, p. 446, 836.

[10] J.E. Desnoyers, G. Perron, L. Avedikian, J.P. Morel, J. Solut. Chem. 5 (9) (1976) 631.

[11] H.L. Friedman, J. Solut. Chem. 1 (1972) 387, 413

[12] M. Bloemendal, G. Somsen, J. Solut. Chem. 12 (1983) 83.

[13] W.G. McMillan Jr., J.E. Mayer, J. Chem. Phys. 13 (1945) 276.

[14] T.H. Lilley, in: G.C. Barrett (Ed.), Chemistry and Biochemistry of the Amino Acids, Chapter 21, Chapman & Hall, London, 1985.

[15] T.F. Wegrzyn, I.D. Watson, G.R. Hedwig, J. Solut. Chem. 13 (1984) 233.

[16] G.M. Blackburn, T.H. Lilley, E. Walmsley, J. Chem. Soc., Faraday Trans. I 78 (1982) 1641.

[17] F. Franks, M. Pedley, D.S. Reid, J. Chem. Soc., Faraday Trans. I 72 (1976) 359.

[18] G. Barone, G. Castronuovo, P. Del Vecchio, V. Elia, S. Puzziello, J. Solut. Chem. 18 (1989) 1105.

[19] J. Fernandez, T.H. Lilley, J. Chem. Soc., Faraday Trans. 88 (1992) 2503.