

Volumetric and solvation behaviour of amino acids in some aqueous binaries: Part III—Glycine, DL-alanine and DL- α -aminobutyric acid in aqueous solutions of non-electrolytes at 298.15 K

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Received 23 May 1989; revised and accepted 31 October 1989

Apparent molal volumes, apparent molal adiabatic compressibilities and solvation numbers of glycine, DL-alanine and DL- α -amino-*n*-butyric acid have been determined at 298.15 K in aqueous solutions containing upto 50% (by wt) of ethylene glycol, propylene glycol and glycerol and also in aqueous dimethyl sulphoxide. The volumes and compressibilities of transfer and the solvation numbers at various compositions of the mixtures have also been obtained and interpreted in terms of the nature of the given binary systems and the hydration of amino acids.

In continuation of our studies^{1,2} on the solvation behaviour of amino acids in various aqueous binary mixtures, we present here the apparent molal volumes, apparent molal adiabatic compressibilities and hydration numbers of glycine, DL-alanine and DL- α -aminobutyric acid in some aqueous non-electrolyte solutions. Aqueous non-electrolyte media are suitable for the study of weak non-bonding interactions existing in biological systems³⁻⁷. The cosolvents, viz., ethylene glycol (EG), propylene glycol (PG), glycerol (GL), are all hydrophilic in nature^{8,9} while dimethyl sulphoxide (DMSO) is somewhat hydrophobic in character¹⁰⁻¹³. Although amino acids have been studied by different authors in various media¹⁴⁻¹⁷, there are only a few studies on volumetric properties in non-electrolyte media¹⁸⁻²⁰. Interestingly enough, EG, PG and GL seem to interact similarly with water, but behave differently with respect to protein stabilisation/destabilisation in solution⁹⁻²¹. As such, it will be interesting to see how these cosolvents influence the hydration of amino acids, which are protein components. Dimethyl sulphoxide (DMSO) shows a contrasting behaviour with respect to water, and hence it will be interesting to compare the results of hydrophilic solvents and somewhat hydrophobic DMSO.

Materials and Methods

Glycine and DL-alanine were of chromatogra-

phic grade (BDH, England) and DL- α -amino-*n*-butyric acid was obtained from Sigma Chemicals, USA. These were dried in a vacuum desiccator for 8 days before use. Ethylene glycol (Merck), propylene glycol (Merck), glycerol (GR, Merck) and dimethyl sulphoxide (LR, BDM) were purified by standard methods.

The apparent molal volumes (ϕ_v), apparent molal adiabatic compressibilities (ϕ_k) and their limiting values and the solvation numbers of the amino acids at various compositions of the binary mixtures were obtained at 298.15K by methods described in our previous publication¹. The concentrations of the amino acids were varied from 0.2 to 0.9 mol kg⁻¹.

Results and Discussion

The ϕ_v^0 and ϕ_k^0 values of the amino acids in various binary mixtures are presented in Table 1. The transfer values of these parameters from water to the various binary mixtures are presented along with the solvation numbers in Table 2.

In solutions of amino acids like glycine (Gly), DL-alanine (Ala) and DL- α -aminobutyric acid (Aba) interactions with the solvent will be mostly at the NH₃⁺ and COO⁻ ends, i.e. their solvation will be mostly controlled by charge-dipole forces²². As such, the change, if any, in the solvation of the amino acids when they are transferred from water to the aquo-organic media, will be due to changes

Table 1—Limiting apparent molal volumes (ϕ_v^0) and adiabatic compressibilities (ϕ_k^0) of amino acids in different binary mixtures at 25°C

Mol fr. of cosolvent	ϕ_v^0 (ml mol ⁻¹)			$\phi_k^0 \times 10^4$ (ml mol ⁻¹ bar ⁻¹)		
	Gly†	Ala	Aba	Gly†	Ala	Aba
Water + ethylene glycol						
0.000	43.2	60.5	75.4	-25.9	-23.9	-26.0
0.0312	43.7	60.6 (0.6) ^a	75.3 (0.5)	-20.7	-18.9 (2.2)	-20.4 (1.8)
0.0676	44.6	60.9 (0.5)	75.5 (0.5)	-16.5	-14.2 (1.2)	-15.9 (1.2)
0.1106	45.2	61.3 (0.6)	75.9 (0.5)	-12.8	-10.3 (1.8)	-11.0 (1.4)
0.1620	45.8	61.9 (0.4)	76.5 (0.3)	-9.1	-6.7 (0.8)	-6.0 (0.6)
0.2248	—	62.5 (0.4)	77.2 (0.4)	—	-3.3 (0.7)	-1.0 (1.2)
Water + propylene glycol						
0.0256	43.6 (1.0) ^a	60.4 (0.6)	75.0 (0.6)	-20.1 (1.7)	-18.3 (1.1)	-19.6 (1.5)
0.558	44.3 (0.8)	60.8 (0.5)	75.1 (0.7)	-14.8 (1.5)	-12.6 (0.7)	-13.3 (1.6)
0.0920	45.2 (0.8)	61.3 (0.6)	75.8 (0.5)	-9.6 (1.0)	-8.2 (1.3)	-7.1 (2.5)
0.1362	46.1 (0.7)	62.1 (0.6)	76.7 (0.5)	-6.5 (1.7)	-3.6 (1.8)	-0.5 (1.8)
0.1913	47.0 (0.7)	63.0 (0.8)	77.9 (0.6)	-3.6 (0.5)	+1.7 (0.2)	+5.7 (-0.6)
Water + glycerol						
0.0212	43.8 (0.9) ^a	60.9 (0.5)	75.7 (0.5)	-21.4 (1.2)	-19.7 (1.1)	-20.4 (1.0)
0.0466	44.5 (0.9)	61.3 (0.5)	76.1 (0.5)	-17.9 (1.1)	-15.1 (0.8)	-15.3 (1.3)
0.0773	45.2 (0.8)	61.9 (0.5)	76.6 (0.5)	-13.5 (1.4)	-11.3 (1.2)	-10.5 (0.9)
0.1153	45.9 (0.9)	62.4 (0.5)	77.1 (0.5)	-9.8 (1.5)	-7.6 (1.3)	-5.8 (0.8)
0.1635	46.6 (0.9)	63.1 (0.5)	77.8 (0.6)	-6.2 (1.0)	-3.6 (2.3)	-1.4 (1.5)
Water + DMSO						
0.0249	43.5 (0.8)	60.3 (0.6)	75.0 (0.5)	-20.9 (2.1)	-18.9 (1.7)	-19.8 (1.0)
0.0544	43.9 (1.0)	60.4 (0.6)	74.9 (0.4)	-15.1 (1.7)	-13.3 (0.7)	-13.7 (0.3)
0.0898	44.6 (0.7)	60.6 (0.8)	74.9 (0.7)	-10.1 (1.1)	-8.7 (1.1)	-8.8 (1.3)
0.1330	45.5 (0.7)	61.4 (0.5)	75.7 (0.4)	-6.8 (2.0)	-4.0 (0.8)	-4.3 (1.0)

Gly = Glycine; Ala = DL-alanine; Aba = DL- α -amino-*n*-butyric acid.

(a) Slope values given in the parentheses.

†Data for water + EG system calculated using data from ref. 31.

in such forces as we go from water to the binary solvent.

Water-glycol (EG and PG) and water-glycerol (GL) systems

From pure water to water-EG mixtures containing 50% by weight of EG, the dielectric constant of the binary mixtures changes from 78.5 to 63.2 at 25°C (ref. 23), and the water structure breaks down considerably²⁴. Because of the reduction in

intermolecular interaction in the medium with increase in mole fraction of EG, the cavity creation effect increases resulting in a gradual increase in positive contribution to ϕ_v^0 of the amino acid. The hydration of amino acids will also go on decreasing, and since this hydration is mostly electrostrictive, it will also have a positive contribution to volume. However, the cavity effect increases with increase in the size of the amino acid, whereas the electrostrictive/solvation effect remains almost

Table 2—Limiting apparent molal volume of transfer ($\phi_{v, tr}^0$), adiabatic compressibility of transfer ($\phi_{k, tr}^0$) of amino acids from water to various binary mixtures and hydration numbers (n_h) in these media at 25°C

Mol fr. of cosolvent	$\phi_{v, tr}^0$ (ml mol ⁻¹)			$\phi_{k, tr}^0 \times 10^4$ (ml mol ⁻¹ bar ⁻¹)			n_h		
	Gly	Ala	Aba	Gly	Ala	Aba	Gly	Ala	Aba
Water + ethylene glycol									
0.0312	0.5	0.1	-0.1	5.2	5.0	5.6	2.7	2.5	2.7
0.0676	1.4	0.4	0.1	9.4	9.7	10.1	2.3	2.0	2.2
0.1105	2.0	0.8	0.5	13.1	13.6	15.0	1.9	1.6	1.6
0.1620	2.6	1.5	1.2	16.7	17.1	19.9	1.4	1.1	0.9
0.2248	—	2.0	1.9	—	20.5	24.9	—	0.6	0.2
Water + propylene glycol									
0.0256	0.4	0.0	-0.3	5.8	5.6	6.3	2.7	2.4	2.6
0.0558	1.1	0.3	-0.2	11.0	11.3	12.7	2.1	1.8	1.9
0.0920	2.0	0.9	0.5	16.2	15.7	18.9	1.5	1.3	1.1
0.1362	2.9	1.6	1.4	19.4	20.2	25.5	1.0	0.6	0.1
0.1913	3.7	2.6	2.5	22.2	25.6	31.7	0.6	-0.3	-0.9
Water + glycerol									
0.0212	0.6	0.4	0.4	4.5	4.2	5.5	2.8	2.6	2.7
0.0466	1.3	0.9	0.7	8.0	8.8	10.7	2.6	2.2	2.2
0.0773	2.0	1.5	1.2	12.3	12.6	15.4	2.1	1.8	1.6
0.1153	2.6	2.0	1.7	16.0	16.3	20.2	1.7	1.3	1.0
0.1635	3.4	2.7	2.5	19.6	20.3	24.6	1.1	0.7	0.2
Water + DMSO									
0.0249	0.3	-0.1	-0.3	5.0	5.0	6.1	2.8	2.5	2.6
0.0544	0.7	0.0	-0.4	10.7	10.6	12.3	2.2	1.9	1.9
0.0898	1.4	0.1	-0.4	15.7	15.2	17.2	1.5	1.3	1.3
0.1330	2.3	1.0	0.3	19.0	19.9	21.7	1.1	0.6	0.7
0.1872	3.2	1.8	1.2	23.3	22.2	29.4	0.4	0.3	-0.5

same among the present set of amino acids. Of course, for DL-alanine (Ala) and DL- α -aminobutyric acid (Aba) there will be some contribution from hydrophobic hydration, which will have a negative contribution to volume — being more negative for the more hydrophobic molecule²⁵. As such, the combined effect of all these will result in a volume of transfer in the order: $\phi_{v, tr}^0$ (Gly) > $\phi_{v, tr}^0$ (Ala) > $\phi_{v, tr}^0$ (Aba). However, the order in the variation of ϕ_k^0 and n_h is expected to be $\phi_{k, tr}^0$ (Aba) > $\phi_{k, tr}^0$ (Ala) > $\phi_{k, tr}^0$ (Gly) and $n_{h, tr}$ (Aba) > $n_{h, tr}$ (Ala) > $n_{h, tr}$ (Gly), because of the contribution from hydrophobic hydrations in the case of DL- α -aminobutyric acid (Aba). This has actually been

observed. It would be worthwhile to mention here that the nature of variation of above two functions is similar to that in other water + cosolvent systems studied. With increase in the mole fraction of EG, ϕ_v^0 and ϕ_k^0 should increase and n_h should decrease. This trend of variation in the above three functions is actually observed as can be seen from Tables 1 & 2.

The trends in variation in the above three functions in water + PG and water + GL systems are grossly similar to that in the water + EG system (vide Tables 1 and 2); and the small variations can be traced to the increased hydrophobicity in PG and increased hydrophilicity in GL.

Water-DMSO system

DMSO is known to enhance the three dimensional structure of water¹⁰ and as such at the initial addition of DMSO to water, cavity creation factor for dissolution of amino acid will be minimised, resulting in negative volumes of transfer. As the DMSO concentration increases, the cage structures can no longer be sustained and will break down. Under the circumstances the DMSO-water system behaves like the water + glycol or water + glycerol system, so that after a particular concentration of DMSO, the volumes of transfer ($\phi_{v, tr}^0$) of the amino acids would increase, which is actually observed. The $\phi_{k, tr}^0$ and n_h values however show a more regular variation, indicating that the amino acids get more and more dehydrated with increase in DMSO concentration (see Table 2).

Comparison of different systems

It appears from the data in Tables 1 and 2 that while $\phi_{v, tr}^0$ for glycine increases with increase in cosolvent content of the medium, those of alanine and aminobutyric acid have some negative values at certain mole fraction of cosolvent. This indicates that the sign and magnitude of $\phi_{v, tr}^0$ for these amino acids in these systems depend on the nature of the amino acid and on the nature as well as concentration of the cosolvent. Thus, the volume of transfer decreases with increase in the hydrophobic nature of the cosolvent or the hydrophobic nature of amino acids.

A comparison of $\phi_{v, tr}^0$ of amino acids in presence of electrolytes¹ (KCl) at 0.8 *m* with those in non-electrolytes also at 0.8 *m* (obtained by interpolation) indicates that $\phi_{v, tr}^0$ of amino acids are much greater in electrolyte solutions than in non-electrolyte solutions (Table 3). This appears to suggest that zwitterion-ion interactions are much stronger than zwitterion-nonelectrolyte interactions.

The positive values of $\phi_{k, tr}^0$ (Table 2) indicates that ϕ_k^0 of the amino acid solution is determined by the combined effects of (i) reduction²⁷ or loosening of electrostricted hydration zone around zwitterionic ends and (ii) destruction of hydrophobic hydration around the carbon skeleton^{27,28}, in the presence of cosolvent. Since both these effects have positive contributions to ϕ_k^0 , amino acids with larger carbon skeletons are likely to exhibit more positive values of $\phi_{k, tr}^0$. Again, among the cosolvents, the observed large values of $\phi_{k, tr}^0$ of the amino acids in the presence of PG, may be due to its effectiveness in reducing electrostriction around the zwitterionic ends through its hydrophilic centre and destruction of hydrophobic hy-

Table 3—Limiting apparent molal volume of transfer ($\phi_{v, tr}^0$) of amino acids from water to 0.8*m* electrolyte (KCl) and 0.8*m* non-electrolyte solutions at 25°C

Salt/ cosolvent	Molality of the salt/cosolvent (mol kg ⁻¹)	$\phi_{v, tr}^0$ (ml mol ⁻¹)		
		Gly	Ala	Aba
KCl ^a	0.8	1.25	1.42	1.27
EG	0.8	0.25	0.05	-0.05
PG	0.8	0.20	0.0	-0.20
GL	0.8	0.40	0.30	0.25
DMSO	0.8	0.15	-0.10	-0.25

(a) Taken from ref. 1.

dration zone of amino acid via interaction with hydration sphere of its hydrophobic moieties. Apart from these interactions (electrostriction or hydrophobic), the contribution of modified water structure by the solutes (amino acids) and cosolvents should be considered for the detailed analysis of $\phi_{k, tr}^0$.

The limiting apparent molal compressibilities (ϕ_k^0) have been used to compute hydration numbers of amino acids in these media. It is to be noted that hydration numbers of amino acids decrease (as also in the present study) in all the aqueous binaries, except in cases where selective solvation is possible²⁹. The reasons for the decreases in hydration number may be attributed to the removal of water molecules from the hydration zones of the amino acids because of interactions with the cosolvent.

It is very difficult at this stage to assess the relative contributions of various factors stated above and of any other possible phenomenon. However, by changing the interacting species (EG, PG, GL, DMSO) the variation in the hydration numbers of amino acids is not found to be that appreciable. This may be due to the fact that hydration at the zwitterionic ends is the major factor contributing to the hydration of these amino acids, and further that, it is hardly disturbed by the interactions with the non-electrolytes chosen in this study. Similar conclusions were also drawn earlier from volumetric studies of amino acids in aqueous urea^{22,30}.

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