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CALORIMETRIC STUDY OF SOME α-AMINO ACIDS IN WATER AT 25°C

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ABSTRACT

A calorimetric study of six α-amino acids is presented. These form three couples which belong to three different classes of α-amino acids.

Different values of the ionization enthalpy can be related to the structure variation of the R substituent group.

A sharp difference was found between hydrophobic and hydrophilic R groups.

INTRODUCTION

Proton ionization from amino acids has been widely investigated for several years with quite diverse results.

The α-amino acids are very important compounds which are also called "standard" amino acids. They differ from one another in the structure of the side-chains, called R groups.

The α-amino acids can be classified by these R groups:

(a) apolar or hydrophobic groups;
(b) polar groups without charge;
(c) polar groups with negative charge;
(d) polar groups with positive charge.

Many of these compounds [1–6] have been studied from the thermodynamic point of view (ΔG^0, ΔH^0, ΔS^0) by means of potentiometric and calorimetric measurements.

The aim of this work is to study six α-amino acids calorimetrically. These amino acids form three couples which belong to three of the four classes presented above. Therefore, the different values of the ionization enthalpy can be related to small structure variations.

The α-amino acids are: *d*,*l*-valine (Val), L(+)-leucine (Leu), *d*,*l*-phenylalanine (Phe), L(−)-tyrosine (Tyr), *d*,*l*-aspartic acid (Asp), L(+)-glutamic acid (Glu).

EXPERIMENTAL AND PROCEDURE

The compounds (all Carlo-Erba RPE chemicals, except Phe which was from Merck) were used without purification. The compounds were weighed and handled in a nitrogen-filled dry-box.

The calorimetric apparatus has been previously described [7–9].

For Val, Leu, Phe and Tyr the ionization process can be represented as

$$NH_3^+RCOOH_{(aq)} \rightarrow NH_3^+RCOO^-_{(aq)} + H^+_{(aq)} \qquad (1)$$

The molar enthalpy of ionization, $\Delta\overline{H}_1$, of NH_3^+RCOOH was obtained by measuring the molar enthalpy of solution, $\Delta\overline{H}_2$, of crystalline NH_3^+RCOOH, in water at pH 5.98

$$NH_3^+RCOO^-_{(cr)} \rightarrow NH_3^+RCOO^-_{(aq)} \qquad (2)$$

and by measuring the molar enthalpy of protonation, $\Delta\overline{H}_3$, of the same compound in water at pH 0.0.

$$NH_3^+RCOO^-_{(cr)} + H^+_{(aq)} \rightarrow NH_3^+RCOOH_{(aq)} \qquad (3)$$

The molar enthalpy, $\Delta\overline{H}_1$, of process (1) can be obtained by subtracting the two values $\Delta\overline{H}_2$ and $\Delta\overline{H}_3$.

In processes (2) and (3), concentrations ranging from 10^{-3} to 10^{-4} m were used. Therefore, the $\Delta\overline{H}_1$ values can be considered at infinite dilution [7–9]. These values refer to the ionization process of one mole of NH_3^+RCOOH, dissolved at infinite dilution in 1000 g of water, yielding one mole of $NH_3^+RCOO^-$ molecules and one mole of protons solvated in the same amount of water.

It is noteworthy that: (a) at pH 0.0 only the NH_3^+RCOOH form exists; (b) pH 5.98 is the mean of the isoelectric pH values of the compounds, it can therefore be assumed that in this solution the $NH_3^+RCOO^-$ form is predominant.

In this way, it is possible to calculate the first ionization enthalpy.

TABLE 1

Enthalpy values (kcal mol^{-1}) of processes (1), (2) and (3) for some α-amino acids in water at 25°C

	ΔH_1^0	ΔH_2^0	ΔH_3^0
d,l-Valine	1.28	1.11	0.17
L(+)-Leucine	0.70	0.34	0.36
d,l-Phenylalanine	1.96	2.96	−1.00
d,l-Aspartic acid	6.16	5.36	0.80
L(−)-Tyrosine	4.77	5.06	−0.29
L(+)-Glutamic acid	5.91	5.74	0.17

For the two acids, reactions (2) and (3) must be written as

$$\mathrm{COOHRNH_3^+COO^-_{(cr)}} \rightarrow \mathrm{COO^-RNH_3^+COO^-_{(aq)}}$$

and

$$\mathrm{COOHRNH_3^+COO^-_{(cr)}} + \mathrm{H^+} \rightarrow \mathrm{COOHRNH_3^+COOH_{(aq)}}$$

Therefore, it is well known [10] that at pH 6 the 2-carboxyl is also completely dissociated, so that for the acids the $\Delta \bar{H}_1$ values of dissociation of both the carboxyl groups are reported.

The enthalpy values of solution, protonation and ionization of the α-amino acids are reported in Table 1.

DISCUSSION

From the ionization values of Table 1 it can be inferred that for the first couple (Val-Leu) the difference between 0.17 and 0.36 can be related to the additional CH_3 group which diminishes both the electron-withdrawing and the field effects of the NH_3^+ group. These effects, as is well known, tend to dissociate the proton of the 1-carboxyl group.

In the second couple (Phe-Tyr) the difference in the ionization values is related to the effect of the OH group (σ-acceptor and π-donor).

The σ-effect tends to increase the electron-withdrawing effect of the NH_3^+ group on the carboxyl, while the π-electron releasing resonance effect decreases it.

The last couple considered is formed by Asp and Glu. For these compounds the enthalpy measured is related to the dissociation of both the carboxyl groups. It is noteworthy that the total enthalpy is smaller for the dissociation of the glutamic acid than for that of the adipic acid. This trend is also found in the literature [1–4] for the ΔH_1^0 values of the first and second dissociations of the two acids. These values were calculated by means of potentiometric measurements.

Our calorimetric and literature potentiometric measurements agree within 27% in the former and within 54% in the latter case. For valine and leucine the agreement is within 47 and 85%, respectively. For the two acids, both the calorimetric and potentiometric values lead one to assume that the larger distance of the 2-carboxyl from the NH_3^+ group favours the first and second dissociations.

It can also be noted that the solution and protonation terms vary in a different way in the three couples. In the Phe-Tyr couple particularly, a large difference between the two solution values appears. This can be explained by the medium structure-breaking effect displayed by the OH group which forms hydrogen bonds with water. Thus a sharp difference is found between the hydrophobic and the hydrophilic R groups.

Our experimental evidence also supports the following remarks.

While the α-amino acids in acid solution can be represented as NH_3^+RCOOH, in solutions approaching pH 7 the principal species are neutral and may either be in the NH_2RCOOH form or the zwiterion form, $NH_3^+RCOO^-$. Therefore, in eqn. (3) at pH 0, only one form (NH_3^+RCOOH) is present, while in eqn. (2) this is not true.

By means of the dissociation constants [10] it has been possible to calculate the isoelectric pH values of the various compounds. The pH of eqn. (2) is very close to the isoelectric pH values of some of our compounds. It could therefore be hypothesized that for these compounds $NH_3^+RCOO^-$ is the predominant form.

Indeed, the value which is in closest agreement with those found in the literature belongs to leucine, the isoelectic pH of which is equal to that of eqn. (2).

REFERENCES

1 P.K. Smith, A.C. Taylor and E.R.B. Smith, J. Biol. Chem., 122 (1937) 109.
2 D.H. Everett and K.F.K. Wynne-Jones, Trans. Faraday Soc., 1390 (1939) 35.
3 E.R.B. Smith and P.K. Smith, J. Biol. Chem., 146 (1942) 187.
4 J. Llopis and D. Ordonez, J. Electroanal. Chem., 5 (1963) 129.
5 K.P. Anderson, K.O. Greenhalgh and R.M. Izatt, Inorg. Chem., 5 (1966) 2106.
6 E.J. King, J. Am. Chem. Soc., 79 (1957) 6151.
7 F. Rodante and M.G. Bonicelli, Thermochim. Acta, 66 (1983) 225.
8 F. Rodante and M.G. Bonicelli, Thermochim. Acta, 71 (1983) 187.
9 F. Rodante and M.G. Bonicelli, Thermochim. Acta, 74 (1984) 207.
10 A.L. Lehninger, Biochimica, 2nd end., Nicola Zanichelli, Bologna, 1979, p. 69.