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Adsorption of D-, L-, and DL-Phenylalanines at the Mercury-Aqueous Solution Interface

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The adsorption of D-, L-, and DL-phenylalanines at the mercury-aqueous sodium fluoride solution interface has been studied by measuring differential capacities, potentials of zero charge, pzc, and surface tensions at the pzc. The racemate of phenylalanine showed higher surface activity than the enantiomers. This difference has been interpreted in terms of the intermolecular interactions, orientation and re-orientation of adsorbed molecules at the interface.

In a previous paper[1] we described the adsorption properties of a series of aliphatic α-amino acids at the mercury-aqueous solution interface with reference to two factors, *i.e.*, the contribution of the zwitterion group and that of the hydrocarbon group. This paper reports adsorption behavior of D-, L-, and DL-phenylalanines at the mercury-aqueous solution interface. Our major interest in this study is to investigate a possible difference in the adsorption properties between either of the enantiomers and the racemate. Although in liquid state and in solution a racemate is usually considered to have very similar physicochemical properties to those of the pure enantiomers,[2] the difference between them may become appreciable in the processes involving interfacial phenomena, since the contribution of molecular interaction between chiral adsorbate molecules at the interface is generally much more significant owing to higher concentration and the preferential orientation of adsorbed molecules.

Inesi *et al.*[3] reported on the difference between two adsorption properties of the racemate and the enantiomer at the mercury-aqueous solution interface. They found that DL-dibenzoyltartaric acid is more surface active than the enantiomers. They interpreted this difference by the analogy of surface sate to crystal state. In this paper we describe the difference between the adsorption properties of DL-phenylalanine and D-, or L-phenylalanine at the mercury-aqueous solution interface, interpretation being given in terms of the intermolecular interaction, orientation and re-orientation of adsorbed molecules.

Experimental

The differential capacities of the interface were measured as functions of the phenylalanine concentration and electrode potential with the a.c. bridge method.[1] The bridge was adjusted to balance at the moment when a mercury drop spontaneously fell from the dropping mercury electrode. The drop time was measured as follows. The sudden unbalanced signal from the bridge due to the detachment of the drop from the capillary tip was transformed by a lock-in amplifier into an electric pulse, which was then fed to an electronic counter for pulse interval measurement. Drop times of the capillary used were between 13 and 15 s at the potential of zero charge, pzc, in 0.5 mol l^{-1} sodium fluoride. The a.c. signal superimposed to the electrode was of 1 kHz and 5 mV peak to peak.

The potentials of zero charge were determined by a streaming mercury method.[4] The surface tensions at the pzc were measured by the drop time method. Glass tubes, outer diam 4 mm and inner diam 0.1 mm, were drawn to get fine capillaries having *ca.* 0.5 mm and 0.025 mm outer and inner diameters, respectively. The capillaries were dewetted with a silicone coating reagent (Fuji Systems, Japan) and cut to expose clean orifice and give a suitable drop time. Typical characteristics of the capillaries thus prepared were $t=14$ s and $m=0.3$ mg s^{-1} when the height of mercury was 110 cm and at the pzc in 0.5 mol l^{-1} sodium fluoride. The drop times recorded 30 times for each solution were reproducible within $\pm 0.05\%$ as relative standard deviation. The drop times tended to become shorter over several sets of measurements probably due to deterioration of the capillary glass wall by chemical attack of sodium fluoride solution.[5] Thus, it was necessary to calibrate the capillary before and after each measurement. For the calibration we used 0.05 mol l^{-1} sodium sulfate for which the surface tension of mercury is 426.2 mN m^{-1} at the pzc.[6] For conversion of drop time into surface tension we used the approximate relation $\ln(\gamma/\gamma_r)=K\ln(t/t_r)$, where γ and γ_r are surface tensions for the test and a reference solutions, respectively, proposed by Verdier *et al.*[7] Parameter K is a function of the height of mercury, inner diameter of the capillary and surface tension. However, it can be taken as a constant over a fairly wide range of surface tension when the height of mercury is sufficiently large.[7] For a particular capillary of inner diameter 0.002 cm, the value of K was calculated to be 0.976 over a 390—427 mN m^{-1} range of surface tension when the height of mercury was 110 cm. The average standard deviation of the surface tensions we obtained was 0.15 mN m^{-1}.

A test solution was deaerated by bubbling nitrogen gas through the solution for 30 min before measurement and was kept under nitrogen atmosphere during the course of measurement. The electrode potentials were measured with respect to a saturated calomel electrode, SCE. All the measurements were at 25 ± 0.2 °C.

Reagent grade D-, L-, and DL-phenylalanines(Nakarai Chem. Co., Japan) were recrystallized twice from triple distilled water. Recrystallized D-, and L-phenylalanines showed 95 and 93% optical purity, respectively, in optical rotation measurements. Sodium fluoride, a standard reagent for quantitative analysis of 99.95% purity (Hashimoto Chem. Co., Japan), was used without further purification. An ultra pure grade sodium sulfate (E. Merk, Germany) was used for a standard solution in surface tension measurements.

Results

The potentials of zero charge and the surface tensions at the pzc for seventeen concentrations of L-phenylalanine (0.003—0.120 mol l^{-1}), twelve concentrations of D-phenylalanine (0.010—0.120 mol l^{-1}), and twelve

TABLE 1. POTENTIALS OF ZERO CHARGE, E_{pzc}, AND SURFACE TENSIONS OF MERCURY AT THE PZC, γ_{pzc}, IN CONTACT WITH 0.5 mol l^{-1} AQUEOUS SODIUM FLUORIDE SOLUTION CONTAINING D-, L-, OR DL-PHENYLALANINE AT 25 °C

c/mol l^{-1}	$(-E_{pzc}$ *vs.* SCE)/V			γ_{pzc}/mN m^{-1}		
	D	L	DL	D	L	DL
0.003	—	0.458	—	—	419.1	—
0.004	—	0.460	—	—	417.7	—
0.005	—	0.462	0.459	—	417.0	419.6
0.006	—	0.464	0.463	—	416.2	416.3
0.008	—	0.464	0.466	—	414.8	414.7
0.010	0.469	0.467	0.465	413.7	413.7	413.7
0.012	0.467	0.466	0.467	412.5	412.6	412.7
0.015	0.469	0.467	0.464	411.3	411.6	411.5
0.020	0.467	0.467	0.463	409.8	409.8	410.0
0.025	0.467	0.467	0.464	408.5	408.6	408.7
0.030	0.466	0.465	0.463	407.3	407.7	407.5
0.040	0.462	0.462	0.460	405.7	405.8	405.4
0.050	0.459	0.457	0.456	404.1	404.1	403.8
0.060	0.454	0.452	0.453	402.9	402.8	402.5
0.080	0.447	0.444	—	400.7	400.9	—
0.100	0.441	0.437	—	399.2	399.2	—
0.120	0.432	0.432	—	397.6	397.7	—

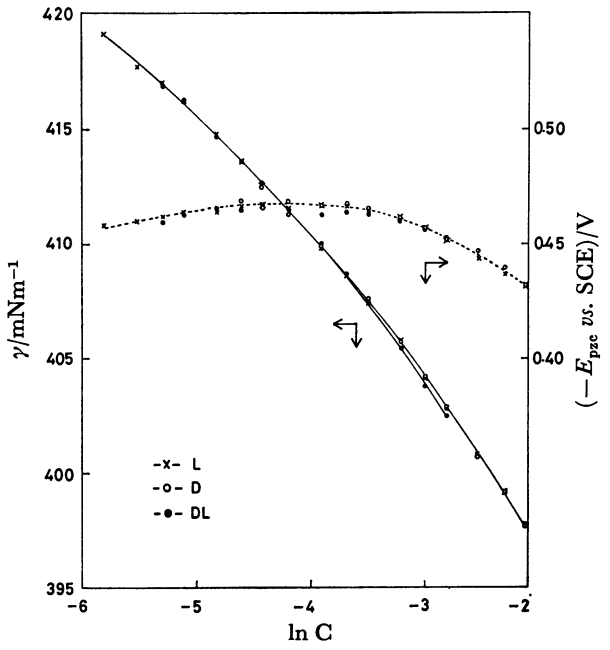

Fig. 1. Potentials of zero charge and surface tensions at the pzc for L-(×), D-(○), and DL-(●) phenylalanines as a function of the logarithm of the phenylalanine concentration in the adsorption on mercury from aqueous 0.5 mol l^{-1} sodium fluoride solutions.

concentrations of DL-phenylalanine (0.005—0.060 mol l^{-1}) in 0.500 mol l^{-1} sodium fluoride are given in Table 1. Due to the limited solubility no data were available beyond 0.06 mol l^{-1} for DL-phenylalanine. Surface tension is plotted in Fig. 1 as a function of the logarithm of phenylalanine concentration. The curves for D-, and L-phenylalanines show excellent agreement over the whole concentration range. At concentration lower than 0.030 mol l^{-1} the curve for DL-phenylalanine is indistinguishable from the curve for both enantiomers. However, with increasing concentration of phenylalanine, it starts to split away from the curve for the enantiomers and lies below them in higher concentration region. This indicates that the racemate is more surface active than the enantiomers.

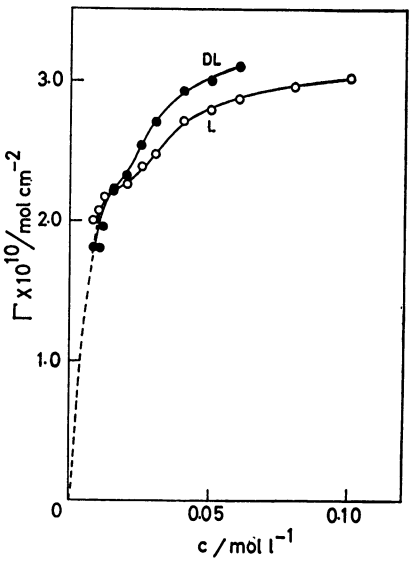

Fig. 2. Adsorption isotherms for L-(○) and DL-(●) phenylalanines at the potential of zero charge.

The surface excesses of L- and DL-phenylalanines, which may be regarded as the actual amount of adsorbed phenylalanines under the present experimental conditions,[8] are plotted in Fig. 2 as a function of the molarity of phenylalanines. As expected, the surface excesses of the racemate are greater than those of the enantiomer. Both these isomers have a kink around 0.02 mol l^{-1} which is characteristic of the change of orientation of adsorbed species at the interface.[9] We have calculated these relative surface excesses from the curves in Fig. 1 using the electrocapillary equation for ideally polarized interfaces.[10] A moving second order least square method[11] with five data points as one set was employed in the numerical differentiation of the curves. Constancy of the activity coefficients of phenylalanines was assumed over the experimental concentration range. The activity of sodium fluoride was also assumed to remain unchanged when the concentration of phenylalanine was varied, though no information is available for the present ternary system to confirm these assumptions. The possible error due to these assumptions[12] is of minor importance in our case since our primary concern here is the difference in the surface excesses between D- or L-phenylalanine and DL-phenylalanine rather than their absolute magnitude.

No significant difference was detected in the potentials of zero charge between the enantiomer and the racemate (Table 1). The potentials of zero charge are plotted against the logarithm of the phenylalanine molarity

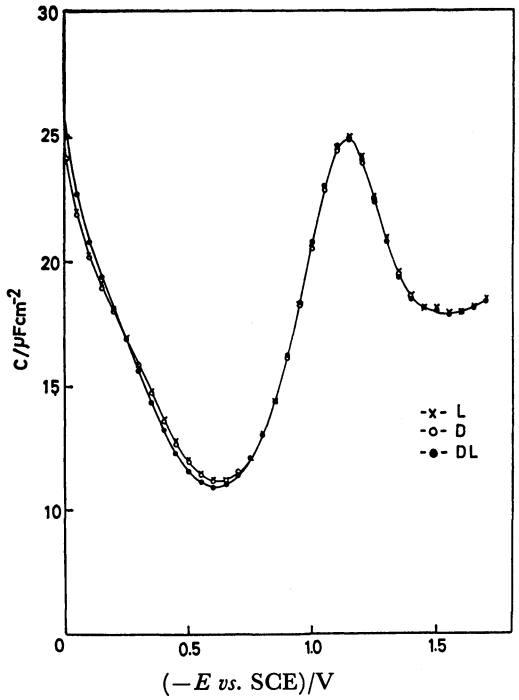

Fig. 3 Comparison of the differential capacity curves for L-(×), D-(○), and DL-(●) phenylalanines in aqueous solution of 0.5 mol l^{-1} sodium fluoride containing 0.05 mol l^{-1} phenylalanine at 25 °C.

in Fig. 1, in which the slopes of the curves change sign with increase in phenylalanine concentration. This suggests the re-orientation of adsorbed phenylalanine molecules with the increase of the surface coverage, as has often been observed in the adsorption of aromatic compounds at electrified interfaces.[13,14]

Differential capacities were measured as functions of the electrode potential and the phenylalanine concentration covering the same range as the surface tension measurements. Comparison is made of the differential capacity curves obtained for D-, L-, and DL-phenylalanines at 0.05 mol l^{-1} in Fig. 3. The curves for D- and L-phenylalanines agree with each other throughout the potential range studied. On the other hand, the curve for the racemate shows appreciable deviation from those curves and lies below them around the pzc. This also indicates the stronger adsorption of the racemate around the pzc than the enantiomers.

In order to estimate the surface excess of phenylalanines at different electrical states of the interface other than the pzc, the differential capacity curves for L- and DL-phenylalanines were twice integrated numerically with respect to the electrode potential. The potentials of zero charge and surface tensitions at the pzc (Table 1) were used as the two independent integration constants. From the electrocapillary curves thus obtained the surface excesses of phenylalanines were calculated as a function of the electrode potential at each phenylalanine concentration. The surface excess *vs.* electrode potential curves for L- and DL-phenylalanines are compared in Fig. 4 at 0.04 mol l^{-1}. The difference between the surface excesses of the racemate and the enantiomer exists not only at the pzc

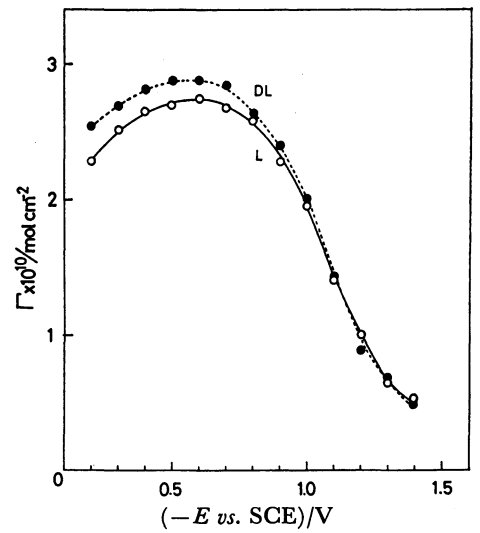

Fig. 4. Comparison of surface excesses of L-phenylalanine with those of DL-phenylalanine at 0.04 mol l^{-1} at various electrode potentials.

but throughout the positive branch, whereas it vanishes at the negative extreme of the potential.

Discussion

Figures 2 and 4 indicate that the racemate of phenylalanine has the higher surface activity than its enantiomers at the mercury-aqueous solution interface in the sense that DL-phenylalanine gives greater surface excess than L- or D-phenylalanine at a given bulk concentration and electrode potential except the cathodic extreme. In Fig. 2 the two isotherms significantly deviate from each other at increased bulk concentrations of phenylalanine, while they merge together with increasing concentration of phenylalanine giving the same limiting slope of the adsorption isotherms at infinite dilution. Similar behavior of adsorption isotherms has been reported by Inesi *et al.*[3] for the adsorption of DL- and D-(and L-)dibenzoyltartaric acids from aqueous solution onto the mercury electrode.

The difference in surface activity should be interpreted in terms of the difference between the adsorption free energy of the racemate and the enantiomers. If we choose the infinite dilution as a reference state of the chemical potentials of phenylalanines in both the adsorption phase and solution phase, the standard adsorption free energy of the racemate should be the same as that of the enantiomers. The difference in surface activity is then ascribed to the difference in the non-ideality due to the intermolecular interactions between the adsorbed phenylalanine molecules. The difference in the interaction energy in the solution phase between racemates and the corresponding enantiomers has been studied theoretically[2,15] and experimentally.[16–18] These works show that the difference in solution phase is negligible in dilute solutions as in our case. Therefore, the observed difference in the adsorbability of phenylalanines should arise from the different molecular interaction in the adsorption phase where the surface concentration of phenylalanine is much

higher than in the solution phase. For example, the surface excess of 3×10^{-10} mol cm^{-2} corresponds to the surface coverage of 0.71 if the maximum surface concentration is assumed to be 4.2×10^{-10} mol cm^{-2}, a value based on CPK model[19] of phenylalanine adsorbed with its aromatic ring plane oriented perpendicular to the mercury surface.

At the lower surface coverage and at the potential around the pzc phenylalanine probably adsorbs with its aromatic ring plane parallel to the electrode surface, as commonly observed in the adsorption of simple aromatic compounds on mercury.[13] This orientation brings the pzc to more negative potential (see Fig. 1) due to the partial charge transfer of π-electron of the aromatic ring to the electrode,[20] while the zwitterion group of phenylalanine may be oriented with its dipole axis parallel to the electrode surface. With increasing surface coverage phenylalanine molecules adsorb with their aromatic ring plane oriented perpendicular to the electrode surface, so that the closer packing of the adsorbed molecules is attained. Since the vertical orientation is unfavorable to the interaction between the π-electron and the electrode, the pzc then shifts to the positive direction[20] with increasing surface excess at the higher surface coverage (Fig. 1).

As an isotherm in which the re-orientation is taken into account, Parry and Parsons[13] and Damaskin[21] have proposed a couple of isotherms assuming that the adsorbate can take two different orientations at the interface;

$$B_1c = \frac{\theta_1}{n_1(1-\theta_1-\theta_2)^{n_1}}\exp(-2n_1a_{11}\theta_1 - 2n_1a_{12}\theta_2)$$

$$B_2c = \frac{\theta_2}{n_2(1-\theta_1-\theta_2)^{n_1}}\exp(-2n_2a_{22}\theta_2 - 2n_2a_{21}\theta_1),$$

where B_i is the adsorption coefficient, θ_i the surface coverage, n_i the ratio of the area occupied by an adsorbate molecule to that of a water molecule and a_{ij} the parameter of intermolecular interactions between adsorbed molecules; indices 1 and 2 are referred to the verical and flat orientations of adsorbed molecules. These isotherms have seven adjustable parameters apart from two maximum surface concentrations, $\Gamma_{m(1)}$ for vertically oriented species and $\Gamma_{m(2)}$ for flatly oriented one and it seems impractical in our case to determine these parameters by fitting experimental data to the theoretical isotherms. Instead, we shall show by invoking these isotherms that the basic features of the experimental adsorption isotherms can be reproduced by choosing probable values for the parameters. Because of the specific interaction between the aromatic π-electron and the mercury surface, the adsorption coefficient of the flatly oriented phenylalanine should be larger than that of the vertically oriented one.[20] We chose the values $B_1=20$ and $B_2=50$ l mol^{-1} and estimated that $n_2/n_1=1.4$ ($\Gamma_{m(1)}=4.3\times10^{-10}$ and $\Gamma_{m(2)}=3.1\times10^{-10}$ mol cm^{-2}) on the basis of the CPK model. The intermolecular interaction would be stronger for the vertical position than the flat position because of the hydrophobic interaction between the adjacent aromatic rings.[20] Hence, we assumed $a_{22}=0.2$ and calculated the isotherms with $a_{11}=1.5$, 1.3, and 1.1 for curves 1, 2, 3 in Fig. 5, in which we further assumed that $n_1=1$ and $a_{12}=a_{21}=0$. The relative surface excesses are plotted against concentration in Fig. 5, from which it is seen that the features of the isotherms are very sensitive to the choice of the interaction parameters. A comparison of Fig. 5 with the experimental isotherms in Fig. 2 suggests that the attractive interaction between the adsorbed phenylalanine molecules is larger for the racemate than for the enantiomers.

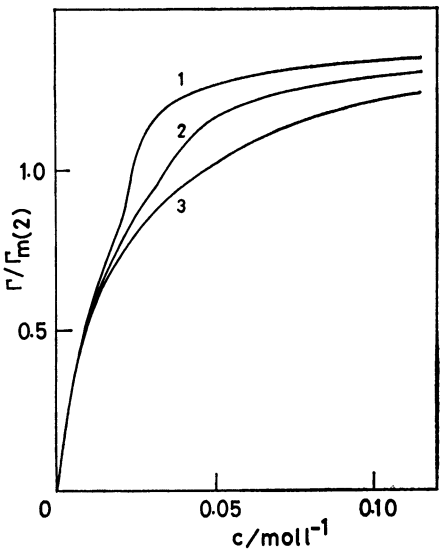

Fig. 5. Calculated curves for reduced adsorption isotherms according to equations in the text with $a_{22}=0.2$, $a_{12}=a_{21}=0$, $n_1=1.0$, $n_2=1.4$, $B_1=20$, $B_2=50$, for $a_{11}=$ 1.5 (1), 1.3 (2), and 1.1 (3).

There are two possible modes in the vertical orientation; one with the axis of the zwitterion group oriented parallel to the electrode and the other with its dipole axis oriented vertical. The former orientation mode should be predominant around the pzc, whereas the latter with the positive end of the dipole facing toward the electrode surface should prevail in the extremely negative branch. In this orientation the dipole would exert repulsive interaction between adsorbed molecules, leading to a smaller value of a_{11} parameter which would give rise to monotonic, Langmuir-type isotherms without kink (curve 3, in Fig. 5). This actually corresponds to the fact that the difference between the surface excess of DL- and D-(or L-) phenylalanines disappears in the cathodic branch away from the pzc (Fig. 4) and also the fact[22] that the adsorption isotherms for L-phenylalanine in the negative extreme have no kink and are well described by Langmuir isotherms which lack the interaction parameter.

References

1) T. Kakiuchi and M. Senda, *J. Electroanal. Chem. Interfacial Electrochem.*, **88**, 219 (1978).
2) H. Mauser. *Chem. Ber.*, **90**, 299, 307 (1957).
3) A. Inesi, F. Rallo, and L. Rampazzo, *Trans. Faraday Soc.*, **64**, 3340 (1968).
4) D. C. Grahame, R. P. Larsen, and M. A. Poth, *J. Am. Chem. Soc.*, **71**, 2978 (1948).
5) A. De Battisti, A. Amadelli, and S. Trasatti, *J. Colloid*

Interface Sci., **63**, 61 (1978).
6) C. A. Smolders and E. M. Duyvis, *Recl. Trav. Chim. Pays-Bas*, **80**, 635 (1961).
7) E. Verdier, R. Grand, and P. Vanel, *J. Chim. Phys.*, **66**, 376 (1969).
8) B. B. Damaskin, O. A. Petrii, and V. V. Batrakov, "Adsorption of Organic Compound on Electrodes," Plenum Press, New York (1971), Chap. 1.
9) B. B. Damaskin, *Eletrokhimiya*, **13**, 816 (1977).
10) D. M. Mohilner, "Electroanalytical Chemistry," ed by A. Bard, Marcel Dekker, New York (1966), Vol. 1, p. 241.
11) P. R. Mohilner and D. M. Mohilner, "Computers in Chemistry and Instrumentation," ed by J. S. Mattson, H. B. Mark, Jr., and H. C. MacDonald, Jr., Marcel Dekker, New York (1972), Vol. 2, p. 3.
12) D. M. Mohilner and H. Nakadomari, *J. Electroanal. Chem. Interfacial Electrochem.*, **65**, 843 (1975).
13) J. M. Parry and R. Parsons, *J. Electrochem. Soc.*, **113**, 992 (1966).
14) K. G. Baikerikar and R. S. Hansen, *J. Colloid Interface Sci.*, **61**, 239 (1977).
15) K. Amaya, *Bull. Chem. Soc. Jpn.*, **34**, 1689 (1961).
16) C. J. McGinn, *J. Phys. Chem.*, **65**, 1896 (1961).
17) S. Takagi, R. Fujishiro, and K. Amaya, *J. Chem. Soc., Chem. Commun.*, **1968**, 480.
18) A. Horeau, *Tetrahedron Lett.*, **1969**, 3121.
19) R. A. Harie, "Molecules in Three Dimensions," Am. Soc. of Biol. Chem., Bethesda, Maryland.
20) Ref. 7, Chap. 3.
21) B. B. Damaskin, *Electrokhimiya*, **5**, 346 (1969).
22) T. Kakiuchi, Thesis, Kyoto University, (1978).