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Base-Catalyzed Hydrolysis of Acetylcholine Chloride in the Presence of Anionic and Nonionic Surfactants

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Base-catalyzed hydrolysis of acetylcholine chloride (Ach) in the presence of anionic and nonionic surfactants was investigated. The rate of hydrolysis was determined by measuring the rate of disappearance of Ach with a spectrophotometer. At a constant pH of 9.0, addition of heptaethylene glycol dodecyl ether (HED) at concentrations above the critical micelle concentration (cmc) results in a slight increase in the apparent rate, v, which is expressed by two reaction rates, v_f and v_m, in the aqueous and micellar phases, respectively. The increase in the apparent rate is due to increase in v_m. In the presence of sodium dodecyl sulfate (SDS), the apparent rate constant, k_{app}, is approximately constant at concentrations below the cmc, and k_{app} decreases above the cmc. In the presence of SDS-HED mixed micelles, k_{app} decreases with the mole fraction of SDS. In these cases, the decrease in the apparent rate is due to the decrease in both v_f and v_m. These two k_{app} decreases are due, respectively, to the decrease in Ach in the aqueous phase and the electrostatic repulsion between OH^- and DS^- at the micelle surface in conformity with the Gouy-Chapman theory.

Micellar catalysis has been a subject of interest in various fields: Hydrolysis of acetylcholine chloride (Ach) is widely studied in various kinetic model experiments for stability prediction,1) and also studied in relation to its chemical and enzymatic stability. Studies carried out since 1930 have established the dependence on pH and temperature,2–4,7) an salt effect,5) and the enthalpy6) of hydrolysis of Ach. Regarding the influence of surfactants on the hydrolysis of Ach, Nogami et al.8) have revealed a retardation of base-catalyzed hydrolysis of Ach by sodium dodecyl sulfate (SDS) micelle by comparing half lives of Ach in the absence and presence of SDS. However, any kinetic analysis on the hydrolysis in the presence of surfactant has not yet been carried out.

In this investigation, a kinetic study on the base-catalyzed hydrolysis of Ach has been carried out in the presence of anionic surfactant, nonionic surfactant, or mixed micelles of anionic and nonionic surfactants. The relation between the rate of hydrolysis and the surface potential of micelle also has been made clear by taking into account the accurate critical micelle concentration (cmc) of surfactant in the presence of Ach9) and the distribution of Ach between micellar and bulk phases.10)

Experimental

Materials. Acetylcholine chloride (Ach) from Nakarai Chemicals Co. was of guaranteed reagent grade and was used without further purification. Sodium dodecyl sulfate (SDS) supplied by Nihon Emulsion Co. was purified as described previously.9–12) Heptaethylene glycol dodecyl ether (HED) from Nikkol Chemicals was of guaranteed reagent grade and was used without further purification. SDS-HED mixed micelles were prepared as described previously.11)

Method of Measurement. The disappearance rate of Ach was followed at a constant pH and 25 °C by determining the concentration of Ach by the method described by Hestrin.13) This method is the same as that employed in our previous study10) and that used by Nogami et al.8)

Procedures. Ach was dissolved in 100 ml of aqueous surfactant solution, and the pH was adjusted by adding aqueous sodium hydroxide solution. The test solution was connected with a pH-stat equipped with a 1 ml glass syringe as described previously12) to keep the pH at a constant value during the reaction period, because acetic acid is produced by the hydrolysis of Ach.14,15) The test solution was collected in 0.1 ml portions at suitable intervals, and the concentration of Ach was determined spectrophotometrically.

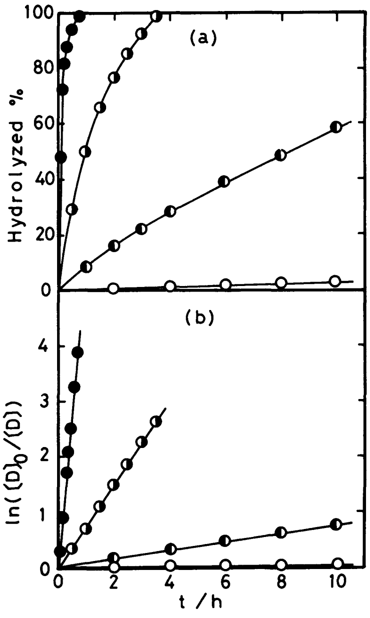

Fig. 1. Effect of pH on hydrolysis of Ach in the absence of surfactant.
(a) Hydrolyzed %, and (b) ln($[D]_0/[D]$) vs. time.
pH: ○ 7.60, ◐ 9.00, ◑ 9.94, ● 11.00.
Concentration of Ach $[D]_0$: 5 mmol dm^{-3}.

Results

Effect of pH on the Hydrolysis of Ach. The pH dependence of the hydrolysis of Ach in the absence of surfactant was examined. The percentage of Ach hydrolyzed is plotted against reaction time in Fig. 1(a).

The apparent rate constant for the hydrolysis, k_{app},

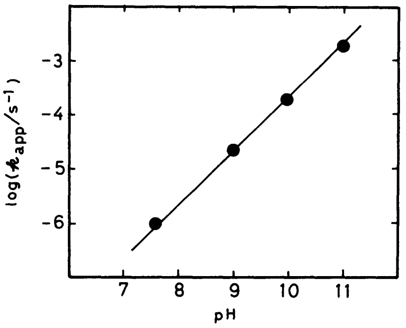

Fig. 2. Relation between pH and apparent rate constant for hydrolysis of Ach in the absence of surfactant.

is defined by

$$\ln\frac{[D]_0}{[D]} = k_{app}\cdot t, \tag{1}$$

where $[D]_0$ is the initial concentration of Ach and $[D]$ is the concentration of Ach at time t. Plots of $\ln([D]_0/[D])$ vs. t are shown in Fig. 1(b), indicating a good proportionality. The relation between pH and the rate constant obtained from the slope of Fig. 1(b) is shown in Fig. 2.

As can be seen in Fig. 2, the plot of log k_{app} vs. pH is linear with a slope of +1, which indicates that the hydrolysis reaction is of apparent first order in hydroxide ion concentration and can be expressed by

$$k_{app} = k_{2,app}[OH^-], \tag{2}$$

where $k_{2,app}$ is the rate constant of second order reaction.

The value of k_{app} at pH 9 and 25 °C, 2×10^{-5} s^{-1}, is well consistent with the value, 1.9×10^{-5} s^{-1}, obtained by Showen et al.,[4] which is in support of the validity of the results obtained here.

Effect of Surfactant on the Hydrolysis of Ach. Effect of surfactants on the hydrolysis of Ach were examined at pH 9.0. It has been found that the surfactants used here are relatively stable at pH 9.0,[12,16–21] so that the decomposition of surfactants can be ignored.

In order to determine the value of k_{app}, values of $\ln([D]_0/[D])$ are plotted against t according to Eq. 1 in Fig. 3 for several examples in the presence of surfactants. In Fig. 3, the percentage of Ach hydrolyzed also is given at the right-side ordinate. In all cases, good proportionality is observed. The hydrolysis of Ach in the presence of surfactants can, therefore, be expressed by the equation of apparent first order in hydroxide ion concentration.

Values of k_{app} obtained from Fig. 3 are shown in Fig. 4 against the total concentration of surfactants, C_t. In the presence of nonionic surfactants HED, k_{app} increases slightly above the cmc (0.066 mmol dm^{-3}).[11] In the presence of SDS, the dependence of k_{app} on surfactant concentration changes in three regions of surfactant concentration. In the presence of HED–SDS mixed micelle (X_{SDS}=0.4), the change in k_{app} is similar to that in the presence of SDS alone, but the first region cannot be found because the value of cmc is as low as 0.1 mmol dm^{-3} for the HED–SDS mixed system (X_{SDS}=0.4) in the presence of 5 mmol dm^{-3} Ach.

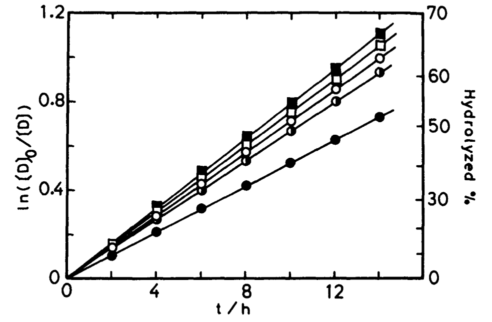

Fig. 3. Hydrolysis behavior of Ach in the presence of surfactants.
Hydrolyzed % vs. time, and $\ln([D]_0/[D])$ vs. time. Surfactants and concentrations: HED □: 2, ■: 20 mol dm^{-3} SDS ○: 2, ◑: 10, ●: 20 mmol dm^{-3}. pH: 9.0. $[D]_0$: 5 mmol dm^{-3}.

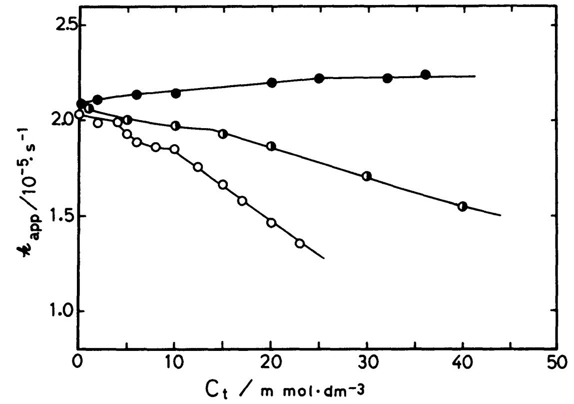

Fig. 4. Effect of surfactant on apparent rate constant for hydrolysis of Ach, k_{app}.
Surfactants: ●: HED alone, ○: SDS alone. ◑: HED–SDS mixed system (X_{SDS}=0.4). pH: 9.0. $[D]_0$: 5 mmol dm^{-3}.

The effect on k_{app} of the composition of HED–SDS mixed system at the total concentration of 20 mmol dm^{-3} is shown by symbols ◑ in Fig. 5(a). The value of k_{app} decreases with the mole fraction of SDS, X_{SDS}.

Effect of Concentrations of Ach and SDS on the Hydrolysis of Ach. To investigate the effect of SDS on the hydrolysis of Ach in a little more detail, experiments were carried out with Ach solutions of 3 and 10 mmol dm^{-3} as well as 5 mmol dm^{-3}, plots of k_{app} against the concentration of SDS being shown in Fig. 6. The first break points correspond to the cmc of SDS in the presence of Ach of each concentration: 4.9, 4.0, and 2.5 mmol dm^{-3} for Ach of 3, 5, and 10 mmol dm^{-3}, respectively.[9] The k_{app} varies with the SDS concentration differently in three regions: k_{app} is approximately constant below the cmc (first region);

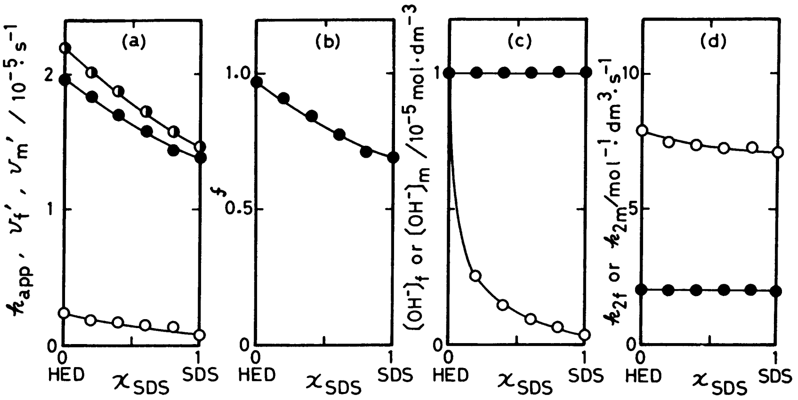

Fig. 5. Effect of HED–SDS mixed system on hydrolysis rate (a), Distribution ratio of Ach in the aqueous phase (b),[10] Concentration of OH^- (c) and Second order rate constant (d).
(a): ◑ k_{app}, ● v'_f, ○ v'_m. (b): $f=[D_f]/[D]$, $[D]=[D_f]+[D_m]$. (c): ● $[OH^-]_f$, ○ $[OH^-]_m$. (d): ● k_{2f}, ○ k_{2m}.
Total concentration of surfactants C_t: 20 mmol dm^{-3}. pH: 9.0. $[D]_0$: 5 mmol dm^{-3}.

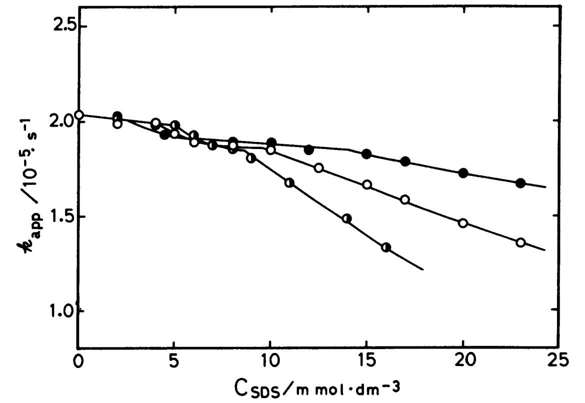

Fig. 6. Effect of SDS on k_{app} with varying concentration of Ach.
$[D]_0$: ◑ 3, ○ 5, ● 10 mmol dm^{-3}. pH: 9.0.

k_{app} decreases slowly above the cmc (second region); k_{app} decreases faster than in the second region (third region). The higher the concentration of Ach is, the longer the second region is and the slower the decrease in k_{app} in the third region is.

The decrease in k_{app} in the presence of SDS at concentrations above the cmc is considered to be due to the fact that the percentage of Ach included in the micellar phases increases with increasing concentration of the micelle[10] and that the approach of OH^- to Ach in the micellar phase is suppressed by the electrostatic repulsion of negatively charged micelle.

Discussion

Derivation of Rate Constants for the Hydrolysis of Ach in the Bulk and Micellar Phases. Ach is distributed between the micellar and bulk phases at surfactant concentrations above the cmc. The rate of the hydrolysis of Ach is, therefore, considered to be a sum of two terms:

$$-\frac{d[D]}{dt} = k_{app}[D] = v = v_f + v_m, \tag{3}$$

where v_f and v_m are the rates of hydrolysis in the aqueous and micellar phases, respectively.

Next, k_{app} can be represented as follows[22] by using the value of distribution ratio of Ach in the aqueous phase, f:[10]

$$k_{app} = k_f f + k_m(1-f), \tag{4}$$

where k_f and k_m are the first order rate constants of hydrolysis in the aqueous and micellar phases, respectively, and the value of $(1-f)$ means the distribution ratio of Ach in the micellar phase. Equation 4 is expressed from Eq. 2 as

$$k_{2,app}[OH^-] = k_{2,f}f[OH^-]_f + k_{2,m}(1-f)[OH^-]_m \tag{5}$$

where $k_{2,f}$ and $k_{2,m}$ are the second order rate constants of hydrolysis in the aqueous and micellar phases, respectively. When the total concentration of added surfactants, C_t, is lower than the cmc, namely C_t<cmc, Eq. 5 simply becomes

$$k_{2,app} = k_{2,f} \text{ (for } C_t < \text{cmc)}, \tag{6}$$

since $f=1$ and $[OH^-]_m=0$.

The rates of hydrolysis in the aqueous and micellar phases, v'_f and v'_m, are defined from Eq. 5 as

$$v'_f = k_{2,f}f[OH^-]_f, \tag{7}$$

$$v'_m = k_{2,m}(1-f)[OH^-]_m, \tag{8}$$

where v'_f and v'_m are related to v or k_{app} by

$$v'_f + v'_m = v' = \frac{v}{[D]} = k_{app}, \tag{9}$$

and $[\mathrm{OH^-}]_\mathrm{m}$ is expressed as

$$[\mathrm{OH^-}]_\mathrm{m} = [\mathrm{OH^-}]_\mathrm{f}\exp\left(-\frac{zF_\mathrm{A}\psi}{RT}\right), \tag{10}$$

where z is the valency of $\mathrm{OH^-}$, F_A is the Faraday constant, ψ is the surface potential at the micelle surface, T is the absolute temperature, and R is the gas constant. Therefore, $k_{2,\mathrm{m}}$ is calculated from Eq. 11 as

$$k_{2,\mathrm{m}} = \frac{(k_{2,\mathrm{app}}-k_{2,\mathrm{f}}f)}{(1-f)}\exp\left(\frac{zF_\mathrm{A}\psi}{RT}\right). \tag{11}$$

With the values $z=-1$, $F_\mathrm{A}=9.648\times10^4$ ($\mathrm{C\,mol^{-1}}$), $R=8.314$ ($\mathrm{J\ mol^{-1}\,K^{-1}}$), and $T=298.15\,\mathrm{K}$ at 25 °C, Eq. 11 is rewritten into

$$k_{2,\mathrm{m}} = \frac{(k_{2,\mathrm{app}}-k_{2,\mathrm{f}}f)}{(1-f)}\exp\left(0.03892\frac{\psi}{\mathrm{mV}}\right). \tag{12}$$

Effect of HED–SDS Mixed Micelles on the Hydrolysis of Ach. The apparent rate of the hydrolysis of Ach in the presence of HED–SDS mixed micelles decreases with the mole fraction of SDS, as shown by symbols ◑ in Fig. 5(a). The values of $k_{2,\mathrm{f}}$ are 2.02 $\mathrm{mol^{-1}\,dm^3\,s^{-1}}$ for the HED–SDS mixed system (X_SDS= 0—0.8) and 1.99 $\mathrm{mol^{-1}\,dm^3\,s^{-1}}$ for the single SDS system, as observed at the cmc (the first break point in Fig. 4) or estimated by Eqs. 2 and 6. Next, the values of $k_{2,\mathrm{m}}$ in the presence of HED–SDS are calculated from Eq. 12 by using the values of f and ψ obtained in our previous study.[10] (Here, the values of $k_{2,\mathrm{m}}$ and ψ are practically unknown quantities. In order to determine the value of $k_{2,\mathrm{m}}$, we use the value of ψ[10] obtained for the HED–SDS mixed micelle system under conditions that $C_\mathrm{t}=20\ \mathrm{mmol\,dm^{-3}}$ and $[\mathrm{D}]_0=5\ \mathrm{mmol\,dm^{-3}}$). The values of $k_{2,\mathrm{m}}$ thus obtained are shown in Fig. 5(d), together with $k_{2,\mathrm{f}}$, and the hydrolysis rates of Ach in the aqueous and micellar phases, v_f' and v_m' are calculated from Eqs. 7 and 8 and shown in Fig. 5(a), together with the data of k_app. In addition, the values of f[10] and $[\mathrm{OH^-}]_\mathrm{m}$ obtained from Eq. 10 are shown in Figs. 5(b) and (c), respectively.

The decrease in v_m' with the mole fraction of SDS (Fig. 5(a)) is due to the decrease in $[\mathrm{OH^-}]_\mathrm{m}$ as shown in Fig. 5(c). The electrostatic effect is well-known with micellar catalyses of various surfactants.[16,23–25]

Effect of Concentrations of Ach and SDS on the Hydrolysis of Ach. The second break point indicated in Fig. 6 may be considered to be the limit point where the number of negative charge supplied from SDS micelle is nearly equal to the number of positive charge supplied from Ach in the micelle (above the second break point, the number of negative charge> the number of positive charge). If the apparent distribution coefficient K' ($K'=([\mathrm{D_m}]/[\mathrm{S}])/[\mathrm{D_f}]$)[10] of Ach between the micellar and aqueous phases is independent of the concentration of surfactant, the number of negative charge {(negative charge)—(positive charge)} increases with increasing concentration of SDS. Values of K' obtained for sufficient concentrations of micelle were found to be independent of the concentration of surfactant at a given concentration of Ach.[10] However, values of K' for insufficient concentrations of micelle were not exactly constant. This leads to the result that the number of negative charge is nearly equal to the number of positive charge below the second break point. The limit point was calculated on the basis of the result[10] on the distribution of Ach between the micellar and aqueous phases obtained by ultrafiltration. The dissociation degree, α, of micellar SDS was assumed to be 0.11 for the calculation. This value of α is in agreement with the previously obtained value of 0.102.[11] The limit points were found to be the points where C_SDS are 8.6, 9.6, and 14.0 $\mathrm{mmol\,dm^{-3}}$ in the presence of Ach of 3, 5, and 10 $\mathrm{mmol\,dm^{-3}}$, respectively.

The third region is considered to be the region where the number of negative charge supplied by SDS micelle exceeds the number of positive charge supplied by Ach existing in the micelle. Therefore, the larger decrease in k_app in the third region is considered to be due to the larger decrease in v_m'. The decrease in v_m' in the third region can be explained in terms of the surface potential of the SDS micelle, ψ. For the hydrolysis rate of Ach in the third region shown in Fig. 6, the value of v_f' is calculated from Eq. 7 by using the values of f[10] and $k_{2,\mathrm{f}}$ observed at the cmc of SDS in the presence of Ach of various concentrations shown in Fig. 6. The values of v_m' for the hydrolysis of Ach in the third region shown in Fig. 6 are calculated from Eq. 9 by subtracting the value of v_f' from the value of k_app ($=v_\mathrm{f}'+v_\mathrm{m}'$) which is obtained experimentally. The decrease in v_m' is considered to be due to the decrease in $[\mathrm{OH^-}]_\mathrm{m}$ which depends on the surface potential of the micelle, although $k_{2,\mathrm{m}}$ of Eq. 8 is the same ($k_{2,\mathrm{m}}=7.023\ \mathrm{mol^{-1}\,dm^3\,s^{-1}}$ for SDS micelle, which is shown in Fig. 5(d)). The value of $[\mathrm{OH^-}]_\mathrm{m}$ is, therefore, calculated from Eq. 8 by using the values of $k_{2,\mathrm{m}}$ and f. The values of v_f', v_m', and $[\mathrm{OH^-}]_\mathrm{m}$ thus obtained are shown in Figs. 7(a) and (b). Furthermore, the values of surface potential of the SDS micelle, ψ, which have not been determined except the values of ψ[10] obtained for the HED–SDS mixed micelle system under conditions that $C_\mathrm{t}=20\ \mathrm{mmol\,dm^{-3}}$ and $[\mathrm{D}]=5\ \mathrm{mmol\,dm^{-3}}$, are calculated by

$$\frac{\psi}{\mathrm{mV}} = -\,59.16\log\frac{[\mathrm{OH^-}]_\mathrm{f}}{[\mathrm{OH^-}]_\mathrm{m}}, \tag{13}$$

where the values of $[\mathrm{OH^-}]_\mathrm{m}$ can be calculated from Eq. 8 by using the values of v_m', $k_{2,\mathrm{m}}$, and f which are obtained already. The values of ψ thus obtained are shown in Fig. 8.

The surface potential of the SDS micelle becomes more negative with increasing concentration of SDS.

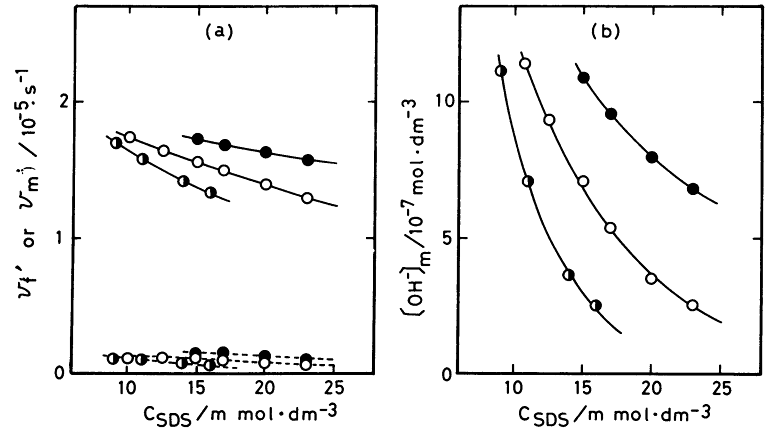

Fig. 7. Relation between (a) Hydrolysis rate and C_{SDS}, and (b) $[OH^-]_m$ and C_{SDS}.
(a) Solid line: v'_f, dotted line: v'_m. $[D]_0$: ◑ 3, ○ 5, ● 10 mmol dm^{-3}. These were calculated for the third region of relation between k_{app} and C_{SDS} shown in Fig. 6.

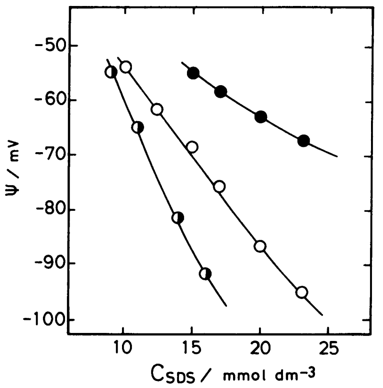

Fig. 8. Relation between surface potential of SDS micelle, ψ, and C_{SDS} in the presence of various concentrations of Ach $[D]_0$: ◑ 3, ○ 5, ● 10 mmol dm^{-3}. These were calculated for the third region in Fig. 6.

This is considered to be due to the decrease in the adsorbed amount of Ach per SDS micelle.

The change in the surface potential according to the increase in SDS concentration is largest when the concentration of Ach is as low as 3 mmol dm^{-3}.

According to the Gouy-Chapman theory, the relation between the surface potential, ψ, and the surface charge density, σ, is expressed as

$$\frac{\sigma}{C_{\mathrm{cm}^{-2}}} = -5.8673 \times 10^{-6} \sqrt{\sum_i \left[\left(\frac{C_i}{\mathrm{mol\ dm^{-3}}}\right)\left\{\exp\left(-\frac{z_i F_A \psi}{RT}\right) - 1\right\}\right]}, \tag{14}$$

where 78.54[26)] is used as the value of relative dielectric constant of water at 25 °C, C_i is the concentration of ion (Ach^+, Cl^-, Na^+, DS^-, OH^-, H^+), and z_i is the valency of ion. The value of σ obtained from ψ by using Eq. 14 is shown in Fig. 9. The value of σ becomes more negative with increasing concentration of SDS.

The relation between the surface charge density, $|\sigma|$,

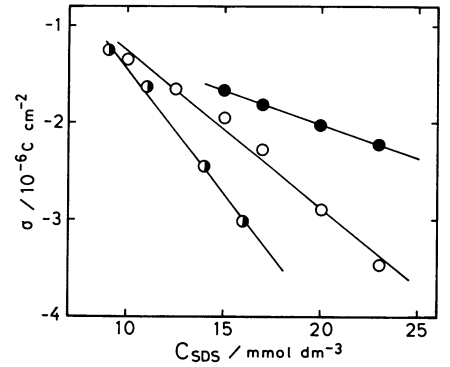

Fig. 9. Relation between surface charge of SDS micelle, σ, and C_{SDS} in the presence of various concentrations of Ach.
$[D]_0$: ◑ 3, ○ 5, ● 10 mmol dm^{-3}. These were calculated from the values of ψ shown in Fig. 8 by the Gouy-Chapman equation.

and the molar number of SDS per unit area, n, which contributes to the supply of the negative charge at the third region shown in Fig. 6, is expressed as

$$\frac{n}{\mathrm{mol\ cm^{-2}}} = \frac{|\sigma|}{F_A}. \tag{15}$$

The relation between ($|\sigma|/F_A$) and (ΔC_{SDS}−$[D_m]$) is shown in Fig. 10, where ΔC_{SDS} is the concentration of micellar dissociated SDS (that is, ΔC_{SDS}=(C_{SDS}−cmc)$\times\alpha$). In Fig. 10, the zero value of (ΔC_{SDS}−$[D_m]$) corresponds to the second break point in Fig. 6 (the value of ψ is −52.5 mV). The relation between ($|\sigma|/F_A$) and (ΔC_{SDS}−$[D_m]$) is found to be embodied by a straight line independent of the concentration of Ach. Therefore, regarding the third region for the k_{app}−C_{SDS} profile shown in Fig. 6, it is evident that σ is determined by the subtraction of concentrations of micellar dissociated SDS and Ach existing in the micellar phase, which is caused by the increase in the molar

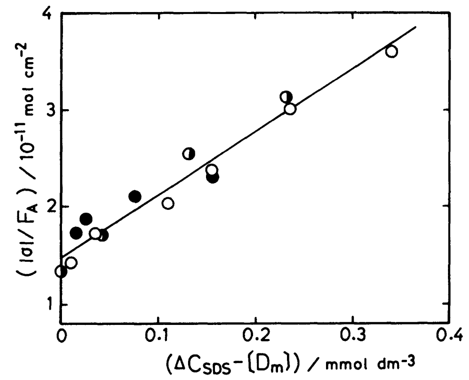

Fig. 10. Plot of ($|\sigma|/F_A$) vs. ($\Delta C_{SDS}-[D_m]$). $[D]_0$: ◑ 3, ○ 5, ● 10 mmol dm^{-3}. These were calculated from the values of σ shown in Fig. 9 and the results of distribution of Ach10) for the third region in Fig. 6.

number of SDS per unit area.

Conclusion

The apparent rate of the base-catalyzed hydrolysis of Ach is decreased by the presence of SDS micelle or HED-SDS mixed micelles because of the decrease in both the rates in the aqueous and micellar phases. It has been concluded that this result can be explained in terms of two parameters, the distribution of Ach between the aqueous and micellar phases and the electrostatic effect at the micelle surface: (1) The decrease in the rate in the aqueous phase is due to the decrease in Ach in the aqueous phase, and (2) the decrease in the rate in the micellar phase is due to the approach of OH^- to Ach in the micellar phase being suppressed by the electrostatic repulsion of negatively charged micelle, which is in conformity with the Gouy-Chapman theory.

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