with $NaHCO_3$ solution and H_2O, dried Na_2SO_4 and evaporated, giving 150 mg. of paste, which was solidified on standing. The solid was washed with Et_2O and the resulting yellow crystals (100 mg.) with fluorescence were recrystallized from MeOH and then from H_2O-Me_2CO, m.p. 200~202°. *Anal.* Calcd. for $C_{20}H_{18}O_6$: C, 67.79; H, 5.12. Found : C, 67.67; H, 5.20. IR : $\nu_{\max}^{\mathrm{CHCl_3}}$ cm^{-1} 1759 ($OCOCH_3$), 1629.

The diacetate was reconverted to (XIII) by alkaline hydrolysis with KOH solution in a quantitative yield.

6-Methoxynaphthacenequinone (XV)[13]——A suspension of 100 mg. of 6-hydroxynaphthacenequinone and 50 mg. of KOH in 10 ml. of MeOH was refluxed for 15 min. and evaporated to dryness in a reduced pressure. The residue was added to a mixture of 1 g. of anhyd. K_2CO_3, 10 g. of MeI and 20 ml. of dehyd. Me_2CO, and the mixture was refluxed for 13 hr. During the reaction, initial deep red-purple color of the reaction mixture turned to pale yellow. Inorganic salts were removed by filtration and the filtrate was evaporated. The resulting residue was crystallized from EtOH or Me_2CO to give 100 mg. of light yellow needles, m.p. 210~212° (Lit., 210°[13]). IR : $\nu_{\max}^{\mathrm{CHCl_3}}$ cm^{-1} 1672 (CO).

11-Methoxy-5(12*H*)-naphthacenone (XVI)——To a solution of 100 mg. of (XV) in 10 ml. of MeOH a solution of 1 g. of $Na_2S_2O_4$ and 0.5 g. of NaOH in 10 ml. of H_2O was added. The resulting dark green mixture was refluxed for 20 min., giving a pink-colored clear solution. The solution was poured into 20 ml. of H_2O and the colorless crystals deposited (62 mg.) were recrystallized from H_2O-Me_2CO, m.p. 175°. *Anal.* Calcd. for $C_{19}H_{14}O_2$: C, 83.20; H, 5.15. Found : C, 82.95; H, 5.01. IR : $\nu_{\max}^{\mathrm{CHCl_3}}$ cm^{-1} 1653 (CO).

Summary

1,3,11-Trimethoxynaphthacenequinone (V) and 1,3-dimethoxyanthraquinone (XIII) were prepared as shown in Charts 1 and 2. Reduction with sodium hydrogensulfite in alkaline medium converted the quinones, (V), (XIII) and 6-methoxynaphthacenequinone (XV), to 1,3,11-trimethoxy-5(12*H*)-naphthacenone (VI), 2,4-dimethoxyanthrone (XII) and 11-methoxy-5(12*H*)-naphthacenone (XVI), respectively.

(Received November 16, 1961)

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[13] Cf. G. Wolf : J. Am. Chem. Soc., **75**, 2673 (1953).

UDC 615.786[612.741.15]-011

162. **Tokuji Suzuki** : Studies on Decomposition and Stabilization of Drugs in Solution. XI.[*1] Chemical Kinetic Studies on Aqueous Solution of Succinylcholine Chloride. 3. Overall Velocity Constants for Succinylmonocholine Chloride Hydrolysis as a Function of pH.

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In the preceding paper,[1] velocity constants of hydrolysis of succinylcholine chloride (I) could be shown for over the whole pH range of 0.9 to 8.5 by using ion exchange chromatography method. From the literature,[2] the degradation of (I) in aqueous solution may be represented in the following manner.

[*1] This work constitutes a part of a series entitled "Studies on Decomposition and Stabilizaion of Drugs in Solution" by H. Nogami. Part X : This Bulletin, **10**, 912 (1962).

[*2] Hongo, Tokyo (鈴木徳治).

[1] T. Suzuki : This Bulletin, **10** 912 (1962).

[2] V.P. Whittaker : Experientia, **7**, 251 (1951).

$$\text{Succinylcholine chloride (I)} \xrightarrow{H_2O} \text{Succinylmonocholine chloride (II)} + \text{Choline chloride}$$

$$\text{Succinylmonocholine chloride (II)} \xrightarrow{H_2O} \text{Succinic acid} + \text{Choline chloride}$$

Succinylmonocholine chloride (II), the first hydrolytic product of (I), has similar curare-like action to that of (I), although considerably less potent. Thus, it is necessary to know the rate of degradation of (II), in order to show exactly the decreasing rate of physiological activity of (I) solution.

According to Frazer,$^{3)}$ (II) is hydrolyzed by human plasma pseudo-cholinesterase and a ratio of the rate of hydrolysis of (I) to that of (II) was 1.4. Phillips$^{4)}$ reported that the curare-like activity of (II) would not show up in the concentration in which (II) would appear as a result of the destruction of (I), but when (II) is given in high concentrations enough to be effective in producing neuromuscular blocking action, its action lasts longer than that of (I), presumably because of less rapid enzymatic breakdown in the body. According to a simple analysis of the alkaline hydrolysis of dicarboxylic ester, it might be expected that the negative charge formed by hydrolysis of the first carboxyl repels the attack by negative ion, such as hydroxyl ion. This may make the second carboxyl more difficult to be hydrolyzed than the first. However, Phillips stated that (II) was hydrolyzed about three times as fast as (I) under certain conditions (pH 3~4).

From the previous observation,$^{1)}$ it was confirmed that the degradation of (I) was a first-order reaction with respect to (I). The degradation of (II) was shown to be a first-order reaction as shown later. Thus, the expression for the hydrolysis of (I) may be written singly at a fixed pH as

$$\text{(I)} \xrightarrow{k_1} \text{(II)} \xrightarrow{k_2} \text{Succinic acid}$$

Where, k_1 and k_2 are the first-order rate constants. If A and B are the concentrations of (I) and (II) respectively at any time, the rates of disappearance of (I) and (II) are given at any instant by equations (1) and (2).

$$-\frac{dA}{dt}=k_1A \tag{1}$$

$$\frac{dB}{dt}=k_1A-k_2B \tag{2}$$

Solution of the differential equation gives equations (3) and (4).

$$A=A_0\exp(-k_1t) \tag{3}$$

$$B=\frac{A_0k_1}{k_2-k_1}\{\exp(-k_1t)-\exp(-k_2t)\} \tag{4}$$

Where, A_0 is initial concentration of (I). If k_1 and k_2 are known, it is possible to follow the breakdown or the formation of these substances at an optional time.

Experimental

Material——Succinylmonocholine chloride (II) was the same as reported previously.$^{5)}$

Buffer——The buffers used in this study are listed in Table I. The determination of pH value was carried out with Towa Denpa Kōgyō Co. pH meter Model HM-5 at each temperature.

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3) P.J. Frazer : Brit. J. Pharmacol., **9**, 429 (1954).
4) A.P. Phillips : J. Am. Chem. Soc., **75**, 4725 (1953).
5) H. Nogami, *et al.* : This Bulletin, **9**, 646 (1961).

Table 1. List of Buffers

Molarity of buffer components	pH 40°	pH 50°	pH 60°	pH 70°
0.288*M* H_2SO_4		0.40	0.50	0.58
0.150*M* HCl 0.050*M* KCl		0.90	0.92	0.93
0.070*M* HCl 0.130*M* KCl		1.20	1.22	1.26
0.027*M* HCl 0.173*M* KCl		1.66	1.68	1.70
0.010*M* HCl 0.190*M* KCl		2.07	2.08	2.09
0.601*M* AcOH		2.44	2.50	2.50
0.061*M* AcOH		3.03	3.04	3.01
0.459*M* AcOH 0.041*M* AcONa		3.60	3.60	3.54
0.406*M* AcOH 0.094*M* AcONa		4.00	3.98	4.03
0.328*M* AcOH 0.172*M* AcONa		4.40	4.41	4.42
0.168*M* AcOH 0.332*M* AcONa		4.98	4.96	5.00
0.058*M* AcOH 0.442*M* AcONa		5.57	5.58	5.57
0.026*M* AcOH 0.474*M* AcONa		5.90	5.90	5.92
0.153*M* KH_2PO_4 0.046*M* NaOH		6.38	6.39	6.40
0.126*M* KH_2PO_4 0.076*M* NaOH		7.00	6.97	6.96
0.200*M* KH_2PO_4 0.180*M* NaOH	7.82	7.80	7.71	
0.322*M* H_3BO_3 0.073*M* NaOH	8.32	8.30	8.21	
0.437*M* H_3BO_3 0.137*M* NaOH	8.59	8.50	8.41	

Kinetic procedure——A 35 mg. sample of (II) was dissolved in 50 cc. of the buffered solution to be be examined. Then, the solution was poured into 5 cc. ampules. These ampules were kept in a thermostatically controlled water bath adjusted to 40°, 50°, 60°, and 70°, accurate to a limit ±0.1°. The ampules were taken out periodically at given intervals, cooled immediately with ice water and an aliquot of 5 cc. was used for colorimetry. The colorimetry was same as reported previously.[1]

Results and Discussions

Consideration for Degradation of (II)

The amounts of (II) remaining in the buffered solutions at 50°, 60°, and 70° after the period of storage were determined colorimetrically. The linear relation of logarithm of concentration of (II) against time might be considered as of an apparent monomolecular reaction. The relation between logarithm of the velocity constant of decomposition and the pH value at each temperature is shown in Fig. 1. The rates of hydrolysis were not measured between pH 7.8 and 8.5 at 70°, but measured at 40°, because the decomposition is very fast at 70°.

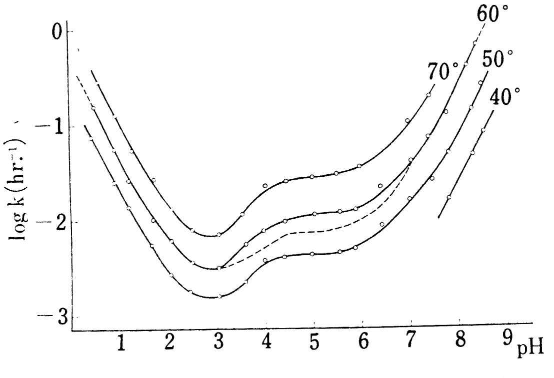

Fig. 1. Overall Velocity Constants for Succinylmonocholine Chloride as a Function of pH

—o—o— Experimental value
--------- Calculated value

Judging from these curves, it appears to be quite similar to the pH-rate curve obtained in the case of hydrolysis of aspirin.[6] The dissociation constant for (II), 1.41×10^{-4}, was calculated from the pH of a half neutralized solution of (II), containing 0.2*M*

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[6] L. J. Edwards: Trans. Farady Soc., **46**, 723 (1950).

sodium chloride at 60°, while that of aspirin at 17° is 2.72×10^{-4}. Thus, a ratio of the concentration of the undissociated acid form for carboxyl radical of (II) to that of the dissociated acid form in aqueous solution changes with variation of the pH value of (II) solution. According to Edwards,[6] the hydrolysis of aspirin may be expressed by six simultaneous reactions, i.e., the molecular and ionic form of aspirin are catalyzed by hydrogen ion, hydroxyl ion and water molecule respectively. The catalytic effect of each catalyst on the degradation of each of these species was different. On the assumption that the mechanism of the hydrolysis of (II) is same as that of aspirin, equation (5) is obtained.

$$\frac{-d\{(\mathrm{II})_{\mathrm{COOH}}+(\mathrm{II})_{\mathrm{COO^-}}\}}{dt}=(\mathrm{II})_{\mathrm{COOH}}\{k_{\mathrm{I}}(\mathrm{H^+})+k_{\mathrm{II}}(\mathrm{H_2O})+k_{\mathrm{III}}(\mathrm{OH^-})\}+(\mathrm{II})_{\mathrm{COO^-}}\{k_{\mathrm{IV}}(\mathrm{H^+})+k_{\mathrm{V}}(\mathrm{H_2O})+k_{\mathrm{VI}}(\mathrm{OH^-})\} \quad (5)$$

Where, $(\mathrm{II})_{\mathrm{COOH}}$ is the concentration of the undissociated acid form, $(\mathrm{II})_{\mathrm{COO^-}}$ the concentration of the dissociated acid form, and each bimolecular rate constant is expressed as k_{I}, k_{II}, k_{III}, k_{IV}, k_{V}, and k_{VI} respectively in the same way as Edwards. Integrated at a fixed pH value, the velocity constant of the apparent monomolecular reaction is given in equation (6) and is a function of the six second-order constants and the dissociation constant of (II). Where, K is the dissociation constant of (II).

$$k=\frac{k_{\mathrm{I}}(\mathrm{H^+})+k_{\mathrm{II}}(\mathrm{H_2O})+k_{\mathrm{III}}(\mathrm{OH^-})}{1+K/(\mathrm{H^+})}+\frac{k_{\mathrm{IV}}(\mathrm{H^+})+k_{\mathrm{V}}(\mathrm{H_2O})+k_{\mathrm{VI}}(\mathrm{OH^-})}{1+(\mathrm{H^+})/K} \quad (6)$$

The six second-order constants were assessed with the same procedure as in aspirin from the kinetic data and the dissociation constant obtained at 60°. In the pH-rate curve for (II), it will be noted that there are pH regions (pH 0.5~1.2, 7.8~8.5), where logarithm of the apparent rate constant is a linear function of pH value with a slope of −1 or +1. It may be considered that these correspond to the reaction between the undissociated acid form and hydrogen ion with a bimolecular velocity constant of 0.466 (L.·mol.$^{-1}$·hr.$^{-1}$), and between the dissociated acid form and hydroxyl ion with a bimolecular velocity constant of 26,300 (L.·mol.$^{-1}$·hr.$^{-1}$). The value of k_{I} and k_{VI} obtained at each temperature are given in Table II.

TABLE II. Bimolecular Rate Constants, k_{I} and k_{VI} at Each Temperature

Temp.	k_{I} (L.·mol.$^{-1}$·hr.$^{-1}$)	Temp.	k_{VI} (L.·mol.$^{-1}$·hr.$^{-1}$)
70°	1.06	60°	2.63×10^{4}
60°	4.66×10^{-1}	50°	1.38×10^{4}
50°	1.98×10^{-1}	40°	6.61×10^{3}

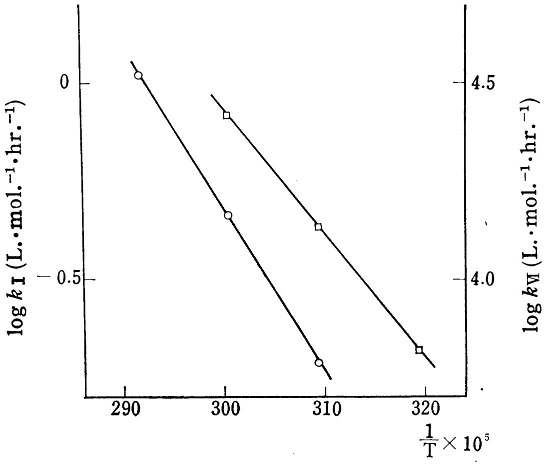


Fig. 2. Arrhenius Plots of k_{I} and k_{VI}

—○—○— k_{I}
—□—□— k_{VI}

Fig. 2 shows the relation between log k_{I} and the reciprocal of absolute temperature, and between log k_{VI} and the reciprocal of absolute temperature.
The Arrhenius equations for these reactions obtained therefrom are as follows :

$$k_{\mathrm{I}} = 5.09 \times 10^{11} \exp\left(\frac{-18,332}{RT}\right) \qquad (7)$$

$$k_{\mathrm{VI}} = 6.53 \times 10^{13} \exp\left(\frac{-14,299}{RT}\right) \qquad (8)$$

The k_w values necessary for the calculation of concentration of hydroxyl ion were calculated similarly as reported previously.[1]

Edwards[6] showed the absence of general acid-base catalysis in the hydrolysis of aspirin at pH 4.7 in aqueous solution by varying the concentration of acetic acid-acetate buffers from 0.005*M* to 0.333*M* and observing no significant increase of the hydrolysis rate. In the hydrolysis of (II), the changes of velocity constants due to a buffer concentration were recognized slightly as shown in Table III.

From the velocity constant at the horizontal portion of the pH-rate curve at 60° in Fig 1., it was recognized that k_{V}, the bimolecular rate constant of the reaction between the dissociated acid form and water molecule, was 1.33×10^{-4} (L.·mol.$^{-1}$·hr.$^{-1}$) and k_{III}, the bimolecular rate constant of the reaction between undissociated form and hydroxyl ion, was the value without any appreciable effect on the observed value of *k*. In this calculation, the value extrapolated to zero buffer concentration at 60°, 0.74×10^{-2} (hr.$^{-1}$) was used as the value of the horizontal portion. From the velocity constant at pH 2.90 (minimum velocity at 60°), it was recognized that k_{IV}, the bimolecular rate constant of the reaction between the dissociated acid form and hydrogen ion, was 14.9 (L.·mol.$^{-1}$·hr.$^{-1}$) and k_{II}, the bimolecular rate constant of the reaction between undissociated acid form and water molecule, was the value without any appreciable effect on the observed value of *k*. Thus, the rate of reaction over the whole pH range at 60° may be practically represented by equation (9).

TABLE III. Velocity Constant Changes due to Buffer Concentrations at 60°

Buffer	k (hr.$^{-1}$)
pH 5.53 $\mu=0.4$	
0.400*M* AcONa 0.056*M* AcOH	1.14×10^{-2}
pH 5.50 $\mu=0.2$	
0.200*M* AcONa 0.028*M* AcOH	1.06×10^{-2}
pH 5.53 $\mu=0.4$	
0.200*M* AcONa 0.028*M* AcOH 0.200*M* NaCl	0.98×10^{-2}

$$k = \frac{0.466\,(\mathrm{H}^+)}{1 + \dfrac{1.41 \times 10^{-4}}{(\mathrm{H}^+)}} + \frac{14.9\,(\mathrm{H}^+) + 1.33 \times 10^{-4}(\mathrm{H_2O}) + 26,300\,(\mathrm{OH}^-)}{1 + \dfrac{(\mathrm{H}^+)}{1.41 \times 10^{-4}}} \qquad (9)$$

The dotted curve shown in Fig. 1 was obtained by calculating equation (9) and the complete curve was obtained by the experimental values. A good agreement was obtained between the calculated curve and the experimental curve in pH value less than 3 and more than 7. The difference between the calculated curve and experimental value may be considered to be due to the effect of the buffer components and the ionic strength of the medium.

The velocity constants of the reaction which account for the minimum rate constant at pH 2.90 are as follows :

$$k=\frac{k_{\mathrm{I}}(\mathrm{H}^+)^2+k_{\mathrm{IV}}K(\mathrm{H}^+)+k_{\mathrm{V}}K(\mathrm{H_2O})}{(\mathrm{H}^+)+K} \tag{10}$$

On differentiating with respect to hydrogen ion concentration and equating to zero, equation (11) is obtained.

$$(\mathrm{H}^+)=K\left\{\left(1+\frac{k_{\mathrm{V}}(\mathrm{H_2O})}{k_{\mathrm{I}}K}-\frac{k_{\mathrm{IV}}}{k_{\mathrm{I}}}\right)^{1/2}-1\right\} \tag{11}$$

Substituting the bimolecular constants obtained above into equaion (11), pH 2.95 was obtained and this value is in good agreement with the experimental value, pH 2.90, at 60°.

Consideration for the Consecutive Reaction in Degradation of (I)

According to equation (4), the amount of (II) in any time can be calculated, when k_1, k_2, and initial concentration of (I) are known. On separating (I) from a partially hydrolyzed (I) solution, the amounts of (II) in various intervals have been already obtained in the procedure in the previous report.[1] Thus, the accordance between calculated values and experimental values was studied using the rate constants, k_1 and k_2, at various pH. Typical results are shown in Figs. 3 and 4. The result in Fig. 3 is an example in the region, where the hydrolysis was mainly catalyzed by hydrogen ion. The result in Fig. 4 is an example in the region, where the hydrolysis was mainly catalyzed by hydroxyl ion. From these figures, it was confirmed that the decomposition of (I) was a consecutive reaction with two first-order reactions.

Fig. 3.
pH 0.93, 70°
$k_1=1.74\times10^{-1}(\mathrm{hr.}^{-1})$ $k_2=1.28\times10^{-1}(\mathrm{hr.}^{-1})$

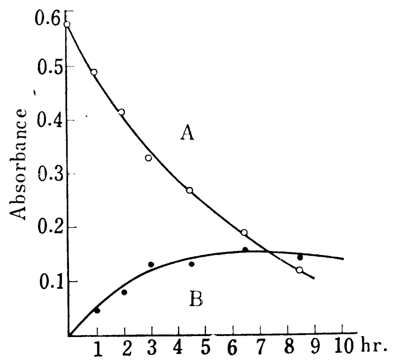
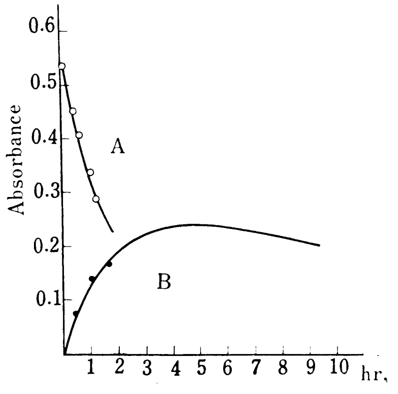


Fig. 4.
pH 7.40, 60°
$k_1=4.98\times10^{-1}(\mathrm{hr.}^{-1})$ $k_2=7.04\times10^{-2}(\mathrm{hr}^{-1})$



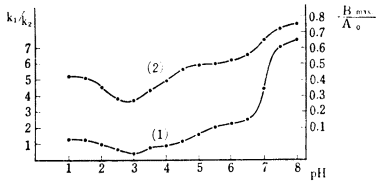
Curves Calculated from the Constants Determined for the Hydrolysis of Succinylmonocholine Chloride

$$\mathrm{A}=A_0\exp(-k_1t)$$

$$\mathrm{B}=A_0\frac{k_1}{k_1-k_2}\{\exp(-k_2t)-\exp(-k_1t)\}$$

Experimental Value :
- ○ Succinylcholine Chloride
- ● Succinylmonocholine Chloride

The ratio, $\frac{k_1}{k_2}$, was shown at various pH values by the curve (I) in Fig. 5.



(1) : The Ratio, $\frac{k_1}{k_2}$, of the Velocity Constant of Snccinylcholine Chloride to That of Succinylmonocholine Chloride against pH.

(2) : The Molar Ratio, $\frac{B_{\max}}{A_0}$, of the Maximum Amount of Succinylmonocholine Chloride produced to the Initial Amount of Succinylcholine Chloride against pH.

Fig. 5. The Ratio of k_1 to k_2 against pH and the Molar Ratio of the Maximum Amount of Succinylmonocholine Chloride produced to the Initial Amount of Succinylcholine Chloride against pH

The rate constants, k_1 and k_2, between pH 1.0 and pH 5.5 in this figure were obtained in the same buffered solution respectively, but the rate constants of (I) above pH 6.0 were observed in unbuffered aqueous solution, maintaining at the initial pH value. The curve (2) expressed the molar ratio of the maximum amount of (II) produced to the initial amount of (I) against pH. From this figure, it may be said that both of these ratios increase with the rise of pH in the region above pH 3.

The author wishes to express thanks and appreciation to Prof. Dr. H. Nogami of the University of Tokyo, for his guidance and encouragement throughout this study, and to Dr. S. Awazu for the discussions and also Mr. S. Fujimori for his technical assistance.

Summary

1) The degradative reaction of succinylmonocholine chloride, the first hydrolytic product of succinylcholine chloride, was investigated over the whole pH range of 0.5 to 8.5 from the standpoint of chemical kinetics. The degradation is an apparent monomolecular reaction at a fixed pH value.

2) The decomposition may be accounted for by the six rate constants of bimolecular reaction and the dissociation constant of succinylmonocholine chloride. The rate(hr.$^{-1}$) of reaction at 60° can be practically represented by

$$k=\frac{0.466(\mathrm{H}^+)}{1+\dfrac{1.41\times10^{-4}}{(\mathrm{H}^+)}}+\frac{14.9(\mathrm{H}^+)+1.33\times10^{-4}(\mathrm{H_2O})+26{,}300(\mathrm{OH}^-)}{1+\dfrac{(\mathrm{H}^+)}{1.41\times10^{-4}}}$$

The relatively slight variation in k caused by buffer effect as compared with succinylcholine chloride was recognized.

3) The hydrolysis of succinylcholine chloride in aqueous solution at a fixed pH value was fitted to a kinetic expression for a consecutive reaction with two first-order reactions. A good agreement was obtained between calculated and experimental values.

(Received August 22, 1961)