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# Studies on the Stability of Ascorbic Acid. V.[1] Critical Micelle Concentration of Ascorbyl Monofatty Acid Esters[2]

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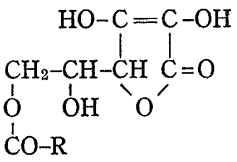
Solubilization of *p*-dimethylaminoazobenzene, change in color of pinacyanol iodide, and surface tension were measured to determine the critical micelle concentrations (CMC) of ascorbyl monofatty acid esters (AMFE).

The CMC is decreased by increasing the length of the fatty acid chain of AMFE. Relationship between log CMC of laurate, myristate, and palmitate which was determined by surface tension method and the numbers of carbon atoms in alkyl groups of these compounds may be represented by the expression, log CMC $= -0.584 - 0.187\ N$, where CMC is expressed in molarity, $N$ is number of carbon atoms in the chain.

Ascorbyl monofatty acid esters (AMFE) have the following general structure, and the existence of one hydrophilic endiol group and one lipophilic hydrocarbon group in the same molecule causes them to have the surface activity. However, the report on the surface activity of AMFE has not yet been seen. Accordingly, it seems worthwhile to attempt the surface-activity investigation of the AMFE in aqueous solution, and to study the relationship between the stability of the ascorbic acid derivatives and their surface activities.



R: alkyl group

The determinations of the critical micelle concentration (CMC) of AMFE were carried out by measurements of solubilization of *p*-dimethylaminoazobenzene (DMAB),[4] change in color of pinacyanol iodide,[5] and surface tension.[6]

## Experimental

**Material**——AMFE (laurate, myristate, and palmitate) were synthesized by the method of Swern, *et al.*[7] Melting points[8] and results of elementary analyses of these compounds are shown in Table I.

Other chemicals used were analytical reagent grade. The water used in this study was redistilled water. The AMFE solutions were prepared by dissolving AMFE in phosphate buffer solution freshly before each experiment. The buffer solution used throughout the study was consisted of a mixture of 0.4 M $Na_2HPO_4$ and 0.4 M $NaH_2PO_4$ and its pH 7.00 was adjusted with the aid of Hitachi-Horiba model F-5 pH meter.

**Determination of CMC**——1) Solubilization of DMAB: Twenty ml of various concentrations AMFE and about 60 mg of DMAB were placed in an ampule, and the air in the ampule was replaced by nitrogen.

1) Part IV: T. Tukamoto, S. Ozeki, and N. Kobayashi, *Ann. Rept. Pharm. Nagoya City Univ.*, **17**, 24 (1969).
2) This work was presented at Meeting of Tokai Branch, Pharmaceutical Society of Japan, Nagoya, June 1970.
3) Location: *Tanabe-dori, Mizuho-ku, Nagoya.*
4) I.M. Kolthoff and W. Stricks, *J. Phys. & Colloid Chem.*, **52**, 915 (1948).
5) M.L. Corrin, H.B. Klevens, and W.D. Harkins, *J. Chem. Phys.*, **14**, 480 (1946); M.L. Corrin and W.D. Harkins, *J. Am. Chem. Soc.*, **69**, 679 (1947); S.H. Herzfeedld, *J. Phys. Chem.*, **56**, 953, 959 (1952); P. Mukerjee and K.J. Mysels, *J. Am. Chem. Soc.*, **77**, 2937 (1955).
6) W.D. Harkins and F.E. Brown, *J. Am. Chem. Soc.*, **41**, 499 (1919).
7) D. Swern, A.J. Stirton, J. Turer, and P.A. Wells, *Oil & Soap*, **20**, 224 (1943) [*C.A.*, **38**, 501 (1944)].
8) Melting points were uncorrected.

Table I. Melting Points and Elementary Analyses of AMFE

| | mp (C°) | Analysis (%) Calcd. C | Calcd. H | Found C | Found H |
|---|---|---|---|---|---|
| Ascorbyl monolaurate | 102—103 | 60.32 | 8.44 | 60.01 | 8.89 |
| Ascorbyl monomyristate | 110—112 | 62.15 | 8.87 | 62.24 | 9.08 |
| Ascorbyl monopalmitate | 113—115 | 63.74 | 9.24 | 63.50 | 9.61 |

These ampules were sealed and shaken in a thermostatically controlled water bath maintained at 50°±1° until equilibrium was reached. The necessary lapse for the above equilibrium was at least sixteen up to twentyfour hours for dilute solutions and three up to five days for concentrated solutions of AMFE. The excess DMAB was removed by suction filtration and absolute ethanol was added. The solubilized DMAB was spectrophotometrically measured at 419 mμ. The spectrophotometer used in above experiments was a Hitachi–Perkin Elmer 139 spectrophotometer.

2) Change in Color of Pinacyanol Iodide: a) Visual method. A solution of AMFE in $1.5\times10^{-5}$ M pinacyanol iodide solution was made up at a concentration well above the CMC of the AMFE and diluted with $1.5\times10^{-5}$ M dye solution using a buret until the initial blue color had changed to the definite blue purple. The same end-point was used in all subsequent titrations. Computation of CMC was made by means of the equation,

$$\text{CMC} = V_a M_a / V_t$$

where $V_a$ and $M_a$ are the initial volume and molarity of the AMFE solution, respectively and $V_t$ is the total volume at the end-point. b) Spectrophotometric method. From the absorption spectrums of AMFE solutions of ten different molarities, the average positions of the band maxima were established at 490 mμ ($\gamma$), 570 mμ ($\beta$), and 615 mμ ($\alpha$). The values of log $I_0/I$ as a function of AMFE concentration were then plotted for each band.

3) Measurement of Surface Tension: Surface tension of AMFE solutions was performed by the drop weight method which was developed by Harkins and Brown,[6] and the corrections given by them were employed in the calculations.

## Result and Discussion

### 1. Solubilization of DMAB

The sudden increase of solubility of DMAB by the addition of AMFE was observed in their aqueous solution as shown in Fig. 1. The CMC of laurate was 1.40 mM, myristate 0.41 mM, and palmitate 0.12 mM, respectively. AMFE seems to be not oxidized to dehydro derivatives, because the air in the ampules has been replaced by nitrogen to supress oxidative degradation of AMFE as above-mentioned. However, hydrolysis of the ester bond of AMFE cannot be protected by the procedure, therefore, it may be seemed that the test solutions would contain hydrolytic products of AMFE.

### 2. Change in Color of Pinacyanol Iodide

Color change of pinacyanol iodide by the addition of AMFE was observed in their aqueous solutions. The values for the CMC of the AMFE determined by visual means are shown in column B of Table II. The CMC of laurate was 1.52 mM, myristate 0.59 mM, and palmitate 0.23 mM, respectively.

The experimental results which were obtained by the spectrophotometric method are shown in Fig. 2—4. It is apparent from these figures that the $\alpha$ and $\beta$ bands increase rapidly their intensity and the $\gamma$ band decreased when the AMFE concentration is increased beyond the CMC. The CMC of AMFE was determined by the mid-point of the rapid drop or rise. The CMC of laurate was 1.38 mM, myristate 0.48 mM, and palmitate 0.17 mM, respectively.

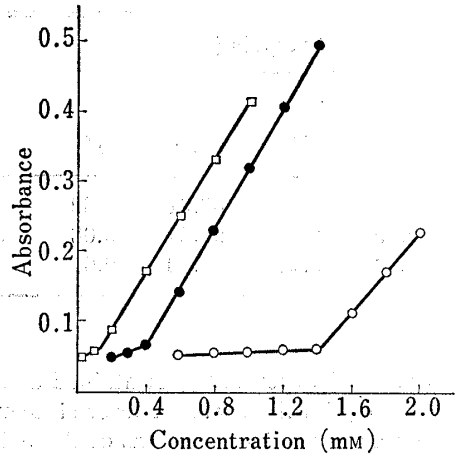



Fig. 1. Solubilization of DMAB in Various Concentrations of AMFE at 50°

—○—○—: laurate
—●—●—: myristate
—□—□—: palmitate

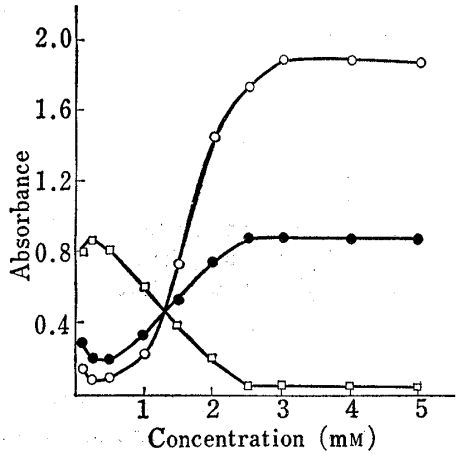



Fig. 2. The Absorbance at the Band Maximum of Pinacyanol in the Presence of Ascorbyl Monolaurate; Pinacyanol Concn. $1.5 \times 10^{-5}$ M

—○—○—: α band, 615 mμ
—●—●—: β band, 570 mμ
—□—□—: γ band, 490 mμ

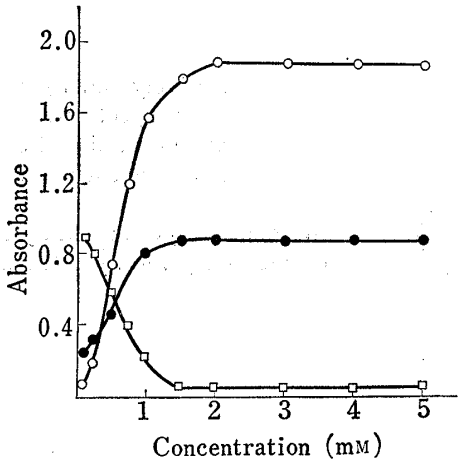



Fig. 3. The Absorbance at the Band Maximum of Pinacyanol in the Presence of Ascorbyl Monomyristate; Pinacyanol Concn. $1.5 \times 10^{-5}$ M

—○—○—: α band, 615 mμ
—●—●—: β band, 570 mμ
—□—□—: γ band, 490 mμ

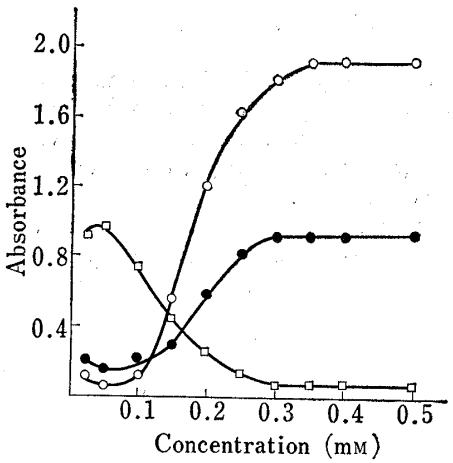



Fig. 4. The Absorbance at the Band Maximum of Pinacyanol in the Presence of Ascorbyl Monopalmitate; Pinacyanol Concn. $1.5 \times 10^{-5}$ M

—○—○—: α band, 615 mμ
—●—●—: β band, 570 mμ
—□—□—: γ band, 490 mμ

### 3. Surface Tension Method

The CMC was determined by the inflexion point of the surface tension *vs.* concentration curve. Relationship between surface tension and AMFE concentration (log C) are shown in Fig. 5. The CMC of laurate was 1.46 mM, myristate 0.62 mM, and palmitate 0.26 mM, respectively.

The above results are summarized in Table II.

A CMC has been shown to be satisfied by an equation which may be simplified and written in the following manner.[9]

9) H.B. Klevens, *J. Am. Oil Chemist's Soc.*, **30**, 74 (1953).

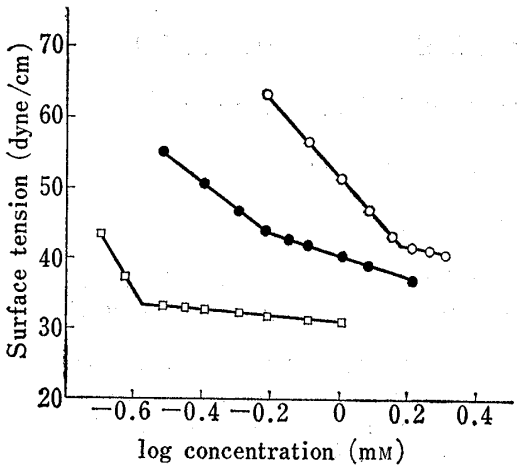



Fig. 5. The Surface Tension *vs.* Logarithm Concentration Curves of AMFE

—○—○—: laurate at 20°
—●—●—: myristate at 20°
—□—□—: palmitate at 50°

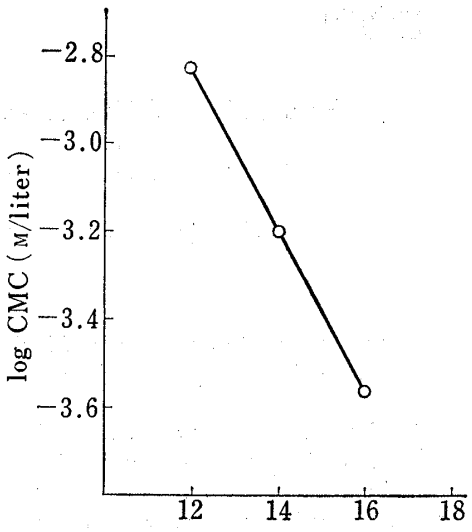



Fig. 6. Variation of CMC of AMFE with Number of Carbon Atoms of Fatty Acid Residues

Table II. Critical Micelle Concentration of AMFE (mM)

| | Method[a] | | | |
|---|---|---|---|---|
| | A | B | C | D |
| Ascorbyl monolaurate | 1.40 | 1.52 | 1.38 | 1.46 |
| Ascorbyl monomyristate | 0.41 | 0.59 | 0.48 | 0.62 |
| Ascorbyl monopalmitate | 0.12 | 0.23 | 0.17 | 0.26 |

*a*) A, solubilization of DMAB; B, visual method; C, spectrophotometric method; D, surface tension

$$\log \mathrm{CMC} = A - BN \qquad (1)$$

where CMC is in molar units, $N$ is the number of carbon atoms in the chain, $B$ is an empirical constant, and $A$ is a constant for the particular temperature and homologous series. It was assumed that equation (1) might hold good for AMFE. Linear relationship between log CMC of laurate, myristate, and palmitate which has been determined by the surface tension method and the number of carbon atoms in alkyl groups of these compounds is shown in Fig. 6.

Computation of the parameters in equation (1) from the CMC values gives the following specific equation for the AMFE,

$$\log \mathrm{CMC} = -0.584 - 0.187N \qquad (2)$$

and it is also in agreement with values determined by other methods.

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