Gel Filtration Study of the Effect of Urea on the Dissolution State of Sodium Dodecyl Sulfate

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Using gel columns of Sephadex G 50 Fine and CPG gel, gel filtration studies were made of the sodium dodecyl sulfate (SDS)–urea system. The results demonstrated that both the CMC and the elution volume of SDS were increased by the addition of urea. The increase of the elution volume indicates the decrease in micellar size, and this coupled with the increase of CMC indicates the increased difficulty of micelle formation by the addition of urea. The direct interaction between SDS and urea was also examined by the gel filtration method and such interaction was confirmed to be absent both below and above the CMC of SDS. Furthermore, the micellar weight of SDS was estimated by the gel filtration method and values of 23000 and 11000 were obtained for SDS solutions containing 0 and 3 mol dm^{-1} urea, respectively.

A number of studies have been reported of the effect of urea on the micelle formation of surfactants.[1–6] The addition of urea increases the critical micelle concentration (CMC) of surfactants. This effect has been explained in terms of the decrease of entropy of micellization due to the breakdown of the structure of water or the weakening of the hydrophobic bonding between the hydrophobic chains of surfactant molecules by the addition of urea.[4] Other explanations have also been presented.[1,2,5]

A similar effect has been discussed for the urea–protein system.[1,7,8] Here, the interaction between the protein and urea is suggested in addition to the interaction between water and urea.[8] Furthermore, urea–polyethlene glycol[9] and urea–fatty acid interactions[10] have been reported. In these studies, conductometric,[1,5] spectroscopic,[2] surface tension,[3] and membrane-potential[5] measurements have been employed.

In the present paper. the gel filtration method which enables a simple and direct measurement[11] has been adopted to study the effect of urea upon the aqueous solution of the ionic surfactant, sodium dodecyl sulfate (SDS). The discussion has been made upon both the water–urea and the SDS–urea interactions.

Experimental

Materials. SDS was prepared according to the Dreger method.[12]. It was purified by recrystalliztion from ethanol and washed with ethyl ether. Purity was confirmed by the absence of a minimum in the surface tension-concentration curve. The commercial SDS product from Nippon Surfactant Chemicals Co. was similarly purified and used. Commercial urea of a guaranteed grade was recrystallized from 95% ethanol. Substances used as a molecular weight standards were reagent grade sodium chloride, blue dextran (Pharmacia product), Pluronics F 68 and F 108 (Asahi Denka products), poly(vinyl alcohol) (PVA) (Nippon Gōsei Kōgyō product), and poly(sodium ethylenesulfonate) (PES) (commercial product). Gel used for packing columns were Sephadex G 50 Fine (Uppsala Pharmacia product) and CPG-10 (Electro-Nucleonic Inc. product). Distilled water was used after degassing by boiling.

Apparatus and Measurement. The apparatus used for the gel filtration was the same as reported in a previous paper.[11] Gel columns used were of 1.2 cm inner diameter and 32 and 80 cm in height for Sephadex G 50 Fine and CPG-10, respectively.

Measurement was made by charging 20 to 35 cm^3 of the sample solution onto the top of the gel column and the solution eluted at constant flow rates of 20 and 30 cm^3/h for Sephadex gel and CPG-10 gel, respectively. In the case of the SDS gel filtration, elution was made using water. For the SDS–urea gel filtration, the gel column was previously filled with urea solution of the same concentration as that of the sample solution. The sample solution was then charged and the solution eluted with urea solution of the same concentration. The eluted solutions were detected either by a conductometer or a differential refractometer (Mitsumi LDC Refractomonitor) and the elution curves obtained on a recorder.

All experiments were conducted at 30±1 °C.

Results and Discussion

Sephadex–Urea System. CMC vs. Urea Concentration: The elution curves of conductance *vs.* flow volume are shown in Fig. 1a and 1b which are similar to those of SDS below and above the CMC in the absence of urea, respectively.[11] Here, κ_T and κ_S represent conductances of the total solution and single ions, respectively and V_f, V_m, and V_s denote elution volumes of the elution front, SDS micelles and single ions, respectively. From these data, the CMC's of the SDS–urea system were obtained for 0, 1.5, and 3 mol dm^{-3} urea concentrations by plotting κ_T and κ_S against the concentration of SDS, as shown for instance in Fig. 2 for the SDS–urea system (3 mol dm^{-3} urea). Figure 3 shows the CMC *vs.* urea concentration curve. The CMC's are seen to increase with increasing concentration of urea. Figure 3 also gives the values calculated from the conductometric data of Emerson and Holtzer.[13]

Urea–SDS Interaction: The urea–SDS interaction has been explained indirectly as the breakdown of the structure of water by urea. However, urea has been reported to interact directly with protein,[8] surfactant,[9] and fatty acid monolayers,[10] and since there is no confirmation of the absence of such a direct interaction between urea and SDS, it was necessary to establish this point. For this purpose, the gel filtration measurement offers a simple and direct method.[14]

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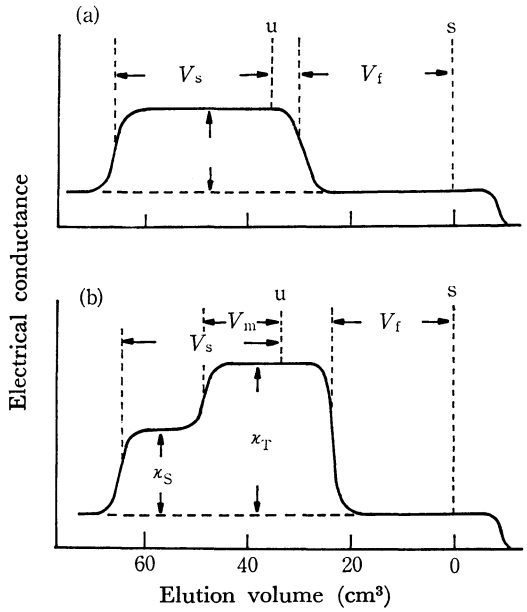

Fig. 1. Gel filtration of aq urea-SDS solution.
(a), (b); Elution curves below and above CMC, respectively. S, U; Points of urea and urea-SDS solution charge, respectively.

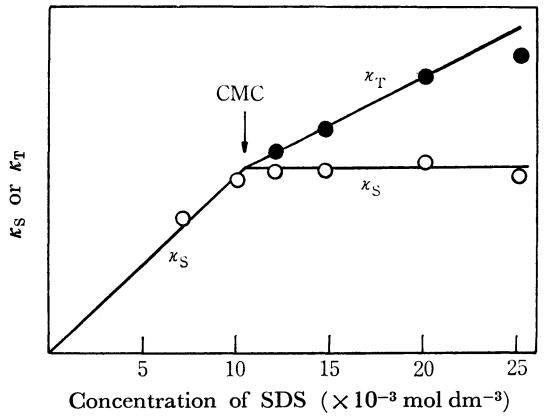

Fig. 2. Conductance of eluent *vs.* concentration of SDS plot.
Gel; Sephadex G 50 Fine, urea; 3 mol dm^{-3}, temperature; 30 °C, CMC; 10.5×10^{-3} mol dm^{-3}

Table 1. Gel filtration of SDS through Sephadex G 50

	Concn of SDS 10^{-3} mol dm^{-3}	Elution volume (cm³) Front V_f	Tail V_m	Tail V_s
Urea 0 mol dm^{-3}	6.7	35.0		35.0
	9.9	31.5	11.7	35.8
	12.0	29.1	11.8	35.5
	14.7	25.1	11.3	35.0
	20.5	22.1	13.6	34.7
Urea 3 mol dm^{-3}	7.1	33.4		33.6
	9.9	33.0		34.2
	12.0	30.1	11.0	33.8
	14.7	27.4	11.3	33.7
	20.0	24.3	14.3	33.8
	25.0	23.0	15.8	34.0

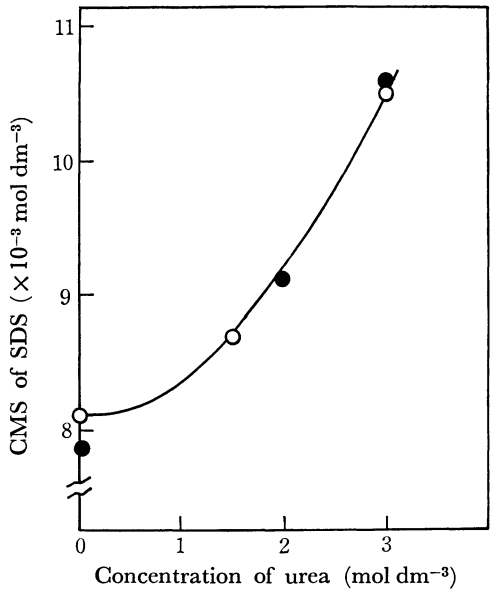

Fig. 3. CMC of SDS *vs.* concentration of urea plot.
○; Present experiment, ●; data of Emerson and Holtzer, temperature; 30 °C.

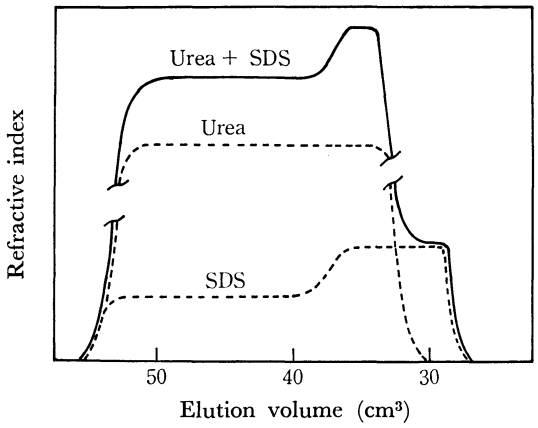

Fig. 4. Elution curve of aq urea-SDS solution.
Gel; Sephadex G 50 Fine, temperature; 30 °C.

Table 1 shows elution volumes V_f, V_m, and V_s of the urea–SDS system containing 0 and 3 mol dm^{-3} urea. Assuming that the urea–SDS complex is formed in the solution in equilibrium with single dodecyl sufate ions (DS^-) and urea, the complex will be eluted faster than DS^- or urea and thus V becomes smaller than V_s below the CMC as in the case of poly(vinyl acetate)–SDS complex formation.[14] However as seen in Table 1, V_f and V_s are equal to each other in the urea–SDS for a 3 mol dm^{-3} urea concentration below the CMC. Therefore the possibility of complex formation is excluded below the CMC. We also confirmed that the interaction of urea with SDS above the CMC or the solubilization was negligible, since the elution curve of the urea–SDS mixture above the CMC was obtained as a simple addition of the elution curves for the separate components as shown in Fig. 4. This differs from the complicated elution curve exhibited in the case of the solubilization of 1-dodecanol by SDS.[15] In Fig. 4, the broken lines show the elution curves of urea and

Table 2. Gel filtration of standard substances through Sephadex G 50

Substance	Urea concn (mol dm^{-3})	
	0	3
Pluronic F68 ($\overline{M}$=8350)	12.0	11.5
Pluronic F108 ($\overline{M}$=15500)	11.3	9.7
Blue dextran	10.2	9.2
NaCl	32.1	32.9

SDS above the CMC and the solid line denotes the elution curve of the urea–SDS system which is obtained by the addition of the ordinates of the former two curves. A similar absence of interaction has also been reported between urea and nonionic surfactant.[16]

V_m of SDS in Urea Solution: As seen from Table 1, the values of V_m show a complicated change with concentration of SDS and urea. Also the V_s values are not equal to each other for the systems of 0 and 3 mol dm^{-3} urea solutions. These facts may be attributed to the change of Sephadex gel matric produced by the addition ures and SDS. This has been confirmed by the change in the elution volumes of the substances of known molecular weight such as NaCl, Pluronics, and dextran by the addition of urea as shown in Table 2. Therefore, to estimate the micellar weight of SDS, it becomes necessary to measure the elution volume of the urea–SDS, system using a gel which is free from such a change. For this purpose CPG-10 gel was chosen.

CPG–Urea–SDS System. Preparation of CPG Gel Column: The CPG gel was superior to the Sephadex gel since the gel matrix was not affected by various solvents, but it showed less difference between V_m and V_s of SDS. A gel column more than twice as long as the Sephadex gel column was necessary to obtain a comparable separation efficiency.

The CPG gel was purified first by washing with distilled water and decantation. Then it was left free in concentrated nitric acid overnight, washed with distilled water, with a water ethanol mixture and finally with pure ethanol. The purified CPG was packed in a glass column mentioned already and care was taken to pack the column uniformly. The packed column was further washed with concentrated aqeous solutions of SDS and ethanol. The completeness of the washing was confirmed by absence of a peak in the elution curve of water obtained by refractometry.

Micellar Weight of SDS: Elution volumes of SDS, NaCl and blue dextran were measured in the absence and presence (3 mol dm^{-3}) of urea. In the latter case, the gel column was previously filled with 3 mol dm^{-3} urea and the same solution used for the elution of the sample solution. Table 3 and 4 show the results. It may be seen in the tables that the elutions volume of SDS single ions (V_s), NaCl and blue dextran are not affected by the addition of 3 mol dm^{-3} urea. Comparison of these results with Table 2 confirms that the CPG gel matrix is unaffected by the urea addition. Further, as seen in Table 3, the values of V_m of SDS are independent of SDS concentration

Table 3. Gel filtration of SDS through CPG-10

	Concn of SDS 10^{-3} mol dm^{-3}	Elution volume (cm^3)		
		Front V_f	Tail	
			V_m	V_s
Urea 0 mol dm^{-3}	18.0	45.0	39.1	50.0
	19.9	44.1	39.2	49.9
	24.7	42.9	39.2	49.5
Urea 3 mol dm^{-3}	18.0	46.5	40.8	50.1
	20.1	45.9	40.6	49.5
	25.2	44.2	40.6	50.2

Table 4. Elution volume of reference substances

Substance	Urea concn (mol dm^{-3})	
	0	3
Blue dextran	38.8	38.8
NaCl	47.1	47.2
PVS	39.5	39.5

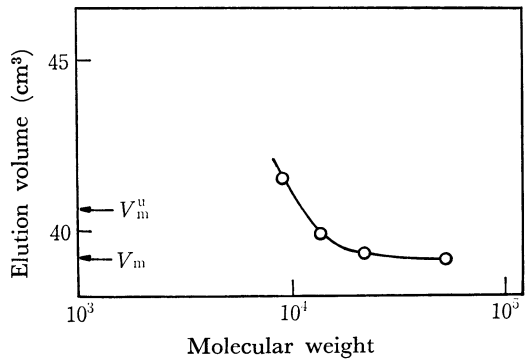

Fig. 5. Elution volume *vs.* molecular weight plot. Gel; CPG gel, V_m, V_m^u; Elution volumes of SDS micelles in water and in aq urea solution, respectively

both for the 0 and 3 mol dm^{-3} urea systems and the value of the 3 mol dm^{-3} urea solution is larger than that of the solution without urea. This evidently shows that the addition of urea decreases the size of the micelle independent of the SDS concentration.

To estimate the micellar weight or SDS, it was necessary to secure a series of substances of known molecular weight, which were used to construct a calibration curve of elution volume *vs.* molecular weight. A series of PVA of mean degrees of polymerization, 200, 300, 500, and 1200 were used as standard substances. The elution volume of PVA without urea is uncertain because of a marked tailing, whereas the tailing is reduced in the presence of 3 mol dm^{-3} urea and a distinct and identical elution volume is obtained for the front and the tail of PVA solution. This may indicate the absence of both CPG– and urea–PVA interactions and the elution volume was considered to be directly related to the molecular weight of PVA. Figure 5 shows the molecular weight *vs.* elution volume plot for 3 mol dm^{-3} urea solution, which was used as a calibration curve for the estimation of the micellar weight. It should be noted that the

range covered by this calibration curve is not satisfactory for the molecular weight determination, however it may be sufficient for the estimation of molecular weight as a whole. Thus, from this curve and the elution volumes of SDS micelles listed in Table 3, the micellar weights of 23000 and 11000 were obtained for SDS in 0 and 3 mol dm^{-3} urea solutions. The former value is comparable to the value of 25000 formerly reported.[17] Further, the micellar weight was confirmed to decrease by the addition of urea, which together with the increase of the CMC mentioned already may indicate the difficulty of micelle formation produced by the additon of urea. Contraty to this result, a slight increase of micellar aggregation number by the addition of urea has been reported for a nonionic surfactant.[18]

The above estimation of the micellar weight assumes a nearly equal weight ratio of hydration between PVA and SDS in the state of solution and the amount of hydration of SDS being independent of the addition of urea. The first point may be confirmed from the coincidence of unhydrated molecular weight *vs.* elution volume plots between PVA and SDS as estimated from the diagram of a previous paper.[17] The second point may be confirmed by comparison of the elution volumes of PES of nearly the same size (elution volume) as those of SDS micelles in 0 and 3 mol dm^{-3} urea solutions. This has been discussed in a previous paper.[19] From the data shown in Table 4, it may be seen that the elution volumes of PES measured in 0 and 3 mol dm^{-3} urea solution are equal to each other. From this we assumed that the amount of yhdration of PES and thereore SDS was not affected by the addition of urea.

References

1) W. Brunning and A. Holtzer, *J. Am. Chem. Soc.*, **83**, 4865 (1961).
2) P. Mukerjee and A. Ray, *J. Phys. Chem.*, **67**, 190 (1963).
3) M. J. Schick, *J. Phys. Chem.*, **68**, 3585 (1964).
4) J. A. Rupley, *J. Phys. Chem.*, **68**, 2002 (1964).
5) J. Piercy, M. N. Jones, and G. Ibbotson, *J. Colloid Interface Sci.*, **37**, 165 (1971).
6) W. U. Malik and S. P. Verma, *Kolloid Z. Z. Polym.*, **233**, 985 (1969).
7) I. M. Klotz and K. Shikama, *Arch. Biochem. Biophys.*, **123**, 551 (1968).
8) J. A. Gordon and W. P. Jencks, *Biochemistry*, **2**, 47 (1963).
9) G. G. Hammes and P. R. Schimmel, *J. Am. Chem. Soc.*, **89**, 442 (1967).
10) D. F. Sears, *J. Colloid Interface Sci.*, **29**, 288 (1969).
11) H. Suzuki and T. Sasaki, *Bull. Chem. Soc. Jpn.*, **44**, 2630 (1971).
12) E. E. Dreger, G. I. Keim, G. D. Miles, L. Schedlovsky, and J. Ross, *Ind. Eng. Chem.*, **36**, 610 (1944).
13) M. F. Emerson and A. Holtzer, *J. Phys. Chem.*, **71**, 3320 (1967).
14) T. Sasaki, K. Tanaka, and H. Suzuki, *Proc. 6th. Int. Congr. Surface Active Substnaces*, **2**, 849 (1972).
15) H. Suzuki, *Bull. Chem. Soc. Jpn.*, **49**, 381 (1976).
16) K. Tajima, *Bull. Chem. Soc. Jpn.*, **49**, 3403 (1976).
17) H. Suzuki, *Bull. Chem. Soc. Jpn.*, **49**, 375 (1976).
18) M. J. Schick, S. M. Atlas, and F. R. Erich, *J. Phys. Chem.*, **66**, 1326 (1962).
19) H. Suzuki, *Bull. Chem. Soc. Jpn.*, **49**, 1470 (1976).