

Availability of Drugs in the Presence of Surface-Active Agents I

Critical Micelle Concentrations of Some Oxyethylene Oxypropylene Polymers

By WITOLD SASKI and S. G. SHAH*

The critical micelle concentrations (CMC's) of the three nonionic surface-active agents—oxyethylene oxypropylene polymers known as Pluronic L 62, Pluronic L 64, and Pluronic F 68—were determined by the surface-tension method and two spectral absorption methods—one employing iodine and the other benzopurpurine 4B. The CMC's for these agents were found to be 2.40 Gm./dl., 2.20 Gm./dl., and 0.1 Gm./dl., respectively.

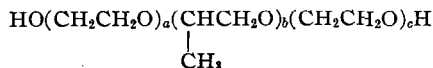
SURFACE-ACTIVE agents have become increasingly important in pharmaceutical technology because of their well-known property for increasing the solubility of water-insoluble organic substances. The applicability of surfactants in this regard has been demonstrated with oil-soluble vitamins (1, 2), estrogens (3), steroids (4), antibacterial agents (5, 6), and fungicides (7). Several studies have been made to elucidate the effect solubilization has on drug availability. The drugs studied included phenols (5, 6), iodine (8), parabens (7, 9–12), and estrogens (13). The mechanism of micellar solubilization is complex and requires more than one physical mode of incorporation of the solubilize in the micelle. The nature of both the solubilizers and the solubilizes studied are too varied to fit any one pattern, and the inclusion of additives, such as salts, produces different influences on the solubilization of different compounds (14). The biological and medicinal activity of the active drug is affected by drug-surfactant interactions profoundly. The effects differ with the characteristics of the drug, the surfactant, and the relative concentrations of each. It has been pointed out (15) that (a) below the limiting association concentration (LAC), the surfactants have little influence on the drug activity; (b) above the LAC, but below the critical micelle concentration (CMC), ionic surfactants interact with the drug to cause a potentiation; and (c) above the CMC, the drug activity is decreased dramatically, and

the activity, if any, is related to the per cent saturation of the solution.

To pursue a meaningful work on availability of drugs in the presence of surface-active agents, it is necessary to determine the CMC's of the surfactants contemplated as possible solubilizers or otherwise useful adjuncts in pharmaceutical formulations. The authors selected some non-ionic oxyethylene oxypropylene polymers for the CMC determination with the further objective of relating the biologic activity of selected drugs to the concentration of the surfactant present.

DISCUSSION

Oxyethylene oxypropylene polymers are prepared by condensing ethylene oxide with a hydrophobic moiety formed by the condensation of propylene oxide with propylene glycol (16) and may be represented by the formula



The molecular weight of either the hydrophobic or hydrophilic portion can be varied in small increments over a wide range. Therefore, it is possible to prepare a product to meet any requirements of molecular weight or hydrophilic-lipophilic balance. Three members of the series of the polymers were selected for the study: (a) an oxyethylene oxypropylene polymer containing 20% of polyoxyethylene (hydrophilic moiety) in total molecule of about 2500 mol. wt., hereafter referred to as Pluronic L 62¹ (liquid); (b) an oxyethylene oxypropylene polymer containing 40% of polyoxyethylene in total molecule of about 2900 mol. wt., hereafter referred to as Pluronic L 64¹ (liquid); and (c) an oxyethylene oxypropylene polymer containing 80% of polyoxyethylene in total molecule of about 8700 mol. wt., hereafter referred to as Pluronic F 68¹ (flakes).

No attempt at purification was made; the work was carried out with heterogeneous mixtures which

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¹ Supplied by Wyandotte Chemicals Corp., Wyandotte, Mich.

result on oxyethylation. For this reason, it was necessary to report the CMC's in grams per deciliter. Favorable reports on toxicological and dermatological properties of the pluronics (16) encouraged the study of their use as potential solubilizers and adjuncts in pharmaceutical technology.

EXPERIMENTAL

Apparatus and Materials

A du Nouÿ 70545 tensiometer with a platinum-iridium ring of 5.998 cm. circumference (Central Scientific Co.) and R/r equal to 54.5 was used for determinations of the CMC's of the pluronics by the surface-tension method. A Bausch and Lomb spectronic 20 spectrophotometer was employed for the CMC determinations by the iodine and benzopurpurin 4B methods. A magnetic stirrer, volumetric glassware, and screw-cap test tubes were used. The chemicals used were: iodine crystals, reagent grade (Allied Chemical and Dye Corp.) and benzopurpurin 4B, biological stain grade (Matheson Coleman and Bell).

Methods

Surface-Tension Method.—Solutions and dilutions of the pluronics were made by the standard procedures. Each of the beakers containing an appropriate dilution was gently stirred by a magnetic stirrer, then covered with aluminum foil and kept in a constant temperature oven at 25° for 12 hours. During the measurements and throughout the experiments, the temperature of the room was kept at 24–25°. Three successive measurements were taken for each of the concentrations and the average calculated and usual corrections applied (17). The corrected surface tension was plotted against the logarithm of the concentration in grams per deciliter.

Iodine Method.—A method developed by Ross and Olivier (18), based on the color change of iodine that takes place when nonionic micelles are present in an iodine solution, was used. A wavelength of 360 $m\mu$ for the maximum absorption of a colored iodine-micelle complex was utilized. A concentrated aqueous solution of iodine was made and subsequently diluted with distilled water to transmit 80% of the light transmitted by the pure solvent (stock solution A) and kept overnight in a constant temperature oven at 25°. A solution of the surfactant of a known concentration was made then using the stock solution A as the vehicle (stock solution B). By diluting solution B with solution A, a series of concentrations covering the anticipated range of concentrations of the polymer was obtained, each with the same concentration of iodine. All spectrophotometer readings were made with solution A as the standard for 100% transmission. The measurements were taken immediately or within 1 hour of the preparation of the final dilution. The per cent transmittance was plotted against the logarithm of the concentration of the surfactant.

Benzopurpurin 4B Method.—A method suggested by Becher (19) was used. Benzopurpurin 4B was made up as a 0.005% solution in distilled water. Addition of a nonionic surface-active agent to an aqueous solution of benzopurpurin 4B causes the color to change from deep red to a reddish-orange.

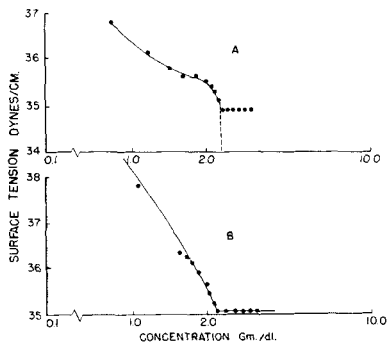


Fig. 1.—Graphs of surface tension against log surfactant concentration plotted to determine the CMC's of oxyethylene oxypropylene polymers. Key: A, Pluronic L 62; B, Pluronic L 64.

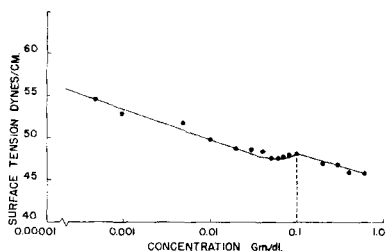


Fig. 2.—Graph of surface tension against log surfactant concentration plotted to determine the CMC of oxyethylene oxypropylene polymer, Pluronic F 68.

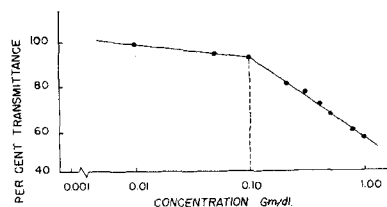


Fig. 3.—Graph of per cent transmittance at 360 $m\mu$, against log surfactant concentration plotted to determine the CMC of oxyethylene oxypropylene polymer Pluronic F 68 by the iodine method.

As the author pointed out, examination of the total absorption spectrum shows that a change in the spectrum is too small to be suitable for a measure of CMC. However, if the spectrum of the solution of surface-active agent is examined at 530 $m\mu$, using the aqueous dye solution as the reference liquid instead of water, this differential absorption spectrum shows pronounced differences as a function of the concentration of the surfactant.

RESULTS AND DISCUSSION

The surface tension of aqueous solutions of surface-active agents decreases rapidly until the CMC is reached, then remains constant above the CMC. Accordingly, the CMC can be determined from the inflections of the surface tension against the logarithm of the concentration curves. The method is satisfactorily applicable to pure nonionic surfactants. If the sample contains impurities, however,

the inflections in the surface tension *versus* log concentration curves become ambiguous and show minima (20). Figure 1 shows the results for the Pluronics L 62 and L 64. The surface tension decreases to 34.95 dynes/cm. at the concentration of 2.40 Gm./dl. and 35.03 dynes/cm. at the concentration of 2.20 Gm./dl., respectively, and is lowered no further with the increase in polymer concentrations. The values represent clear-cut cases of the respective CMC's, although they might appear rather high. This is not the case with Pluronic F 68, reported in Fig. 2, where the curve obtained is some-

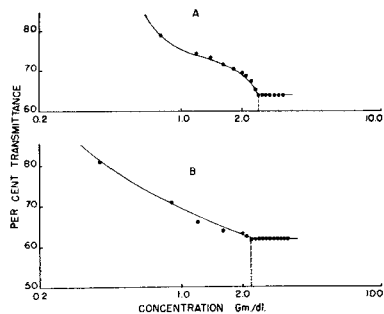


Fig. 4.—Graphs of per cent transmittance at 360 $m\mu$, against log surfactant concentration plotted to determine the CMC's of oxyethylene oxypropylene polymers by the iodine method. Key: A, Pluronic L 62; B, Pluronic L 64.

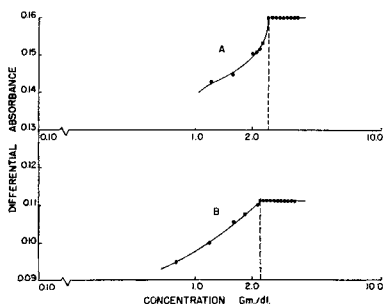


Fig. 5.—Graphs of differential absorbance of 0.005% solutions of benzopurpurin 4B at 530 $m\mu$ as a function of the concentration of surfactant plotted to determine the CMC's of oxyethylene oxypropylene polymers. Key: A, Pluronic L 62; B, Pluronic L 64.

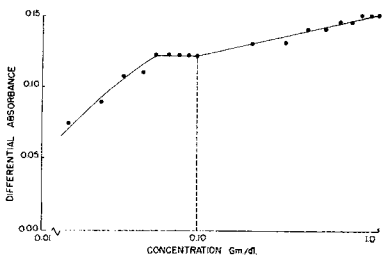


Fig. 6.—Graph of differential absorbance of 0.005% solutions of benzopurpurin 4B at 530 $m\mu$, as a function of the concentration of surfactant plotted to determine the CMC of oxyethylene oxypropylene polymer, Pluronic F 68.

what ambiguous. At 0.05 Gm./dl., a minimum inflection is observed, followed by an increase reaching its maximum at 0.1 Gm./dl. Since the minimum inflection could probably represent an impurity in the surfactant, the point corresponding to 0.1 Gm./dl. concentration was taken as the CMC value.

In the iodine method, all readings were taken within 1 hour of the preparation of the final dilution since, when more than 1 hour elapsed, the per cent transmittance was affected considerably due to gradual fading of color. The results for Pluronic F 68 are reported in Fig. 3, which shows an intersection of the two straight lines obtained upon plotting the per cent transmittance at 360 $m\mu$ against logarithm of concentration of the polymer. The plot follows the pattern depicted by Ross and Olivier. The experiment was carried out three times with freshly prepared stock solutions and dilutions and turned out to be reproducible, giving each time 0.1 Gm./dl. as the CMC value.

Also three series of reproducible experiments were performed with Pluronic L 62 and L 64. The results are presented in Fig. 4. The curves obtained for these two polymers were rather different, compared with Fig. 3. At the concentrations above the CMC, the presence of surface-active agent in solutions of iodine caused no decrease in the intensity of the light transmitted, and the resulting per cent transmittance curve was a straight line parallel to the surfactant concentration axis. The intersection of the two lines was taken as the CMC. Ross and Olivier included Pluronic L 62 in the list of surfactants, the CMC of which was determined by the method they developed. The value of 2.40 Gm./dl. obtained in this work coincides with the value they reported.

The most recent spectral dye method involving benzopurpurin 4B was used as an additional check on previous findings. Three series of reproducible experiments with each of the three polymers were carried out. The results for Pluronics L 62 and L 64 are presented in Fig. 5, where the differential absorbance is plotted against the logarithm of the per cent concentration. Above the concentration of 2.40 Gm./dl. and 2.20 Gm./dl., respectively, no further increase in differential absorbance was observed, and the curves obtained were straight lines parallel to the concentration axis. The resulting intersections presented clear-cut cases of the respective CMC's. In the curve obtained with Pluronic F 68 shown in Fig. 6, a plateau resulted between 0.05 Gm./dl. and 0.1 Gm./dl., followed by a linear increase with the increase of the concentration of the surfactant. The intersection of the two straight lines was taken as the CMC value, although admittedly, the beginning of the plateau corresponding to 0.06 Gm./dl. could also be so considered.

SUMMARY

The CMC's of the three nonionic surface-active agents—oxyethylene oxypropylene polymers known as Pluronics L 62, L 64, and F 68—were determined by three methods—surface tension, iodine, and benzopurpurin 4B. The data obtained were in agreement. The CMC's for Pluronic L 64 and F 68 are reported. The CMC for Pluronic L 62 coincided with that reported by the other workers.

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Cholesterol Particle Growth and Dissolution Rates in Aqueous Media

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As part of the program on the transport, deposition, and dissolution of cholesterol in aqueous media, the growth and dissolution rates and the nucleation behavior of this compound in saline have been investigated. The Coulter counter was used in these studies to follow the particle size distribution changes of cholesterol suspensions with time. A comparison of the results with theory showed that the growth and dissolution rates were close to being diffusion controlled, despite the low rates (10 to 20 hours to grow to about 2- μ diameter or to dissolve a 2- μ diameter particle). Experiments at the higher supersaturation levels showed that a metastable phase preferentially nucleates, grows out rapidly, then redissolves as the more stable phase nucleates more slowly and grows out.

THE TRANSPORT, deposition, and dissolution mechanisms of cholesterol and cholesterol esters are of major importance to the eventual understanding of the diseased and the normal states of man (1, 2). A search of the literature indicates, however, that there has been relatively little quantitative work done on the physical chemistry of the kinetic aspects of these processes. It would appear that significant advances on the problems related to diseases and conditions characterized by excess cholesterol and lipid deposition can be assisted materially by such studies.

To initiate such studies, the growth of cholesterol particles in their supersaturated solutions and the dissolution of these particles in their undersaturated solutions are being investigated. The present report describes the results of the study of cholesterol behavior in their super-

saturated and undersaturated saline solutions with particular emphasis on methodology. Other studies are now in progress on the application of present methods to evaluate the effects of additives on the growth and dissolution rates.

EXPERIMENTAL

General Considerations.—Because the solubility of cholesterol in water is extremely low,¹ it was expected that both the growth rates and the dissolution rates would be low in water, even under the ideal diffusion-controlled situations. Therefore, it was decided to employ the Coulter counter because it had been shown recently (4, 5) that this instrument was suitable for growth and dissolution rate studies of sparingly soluble materials in their suspension or emulsion states.

To eliminate the possible importance of artifact effects in the growth studies, it was decided to try several methods for preparing the supersaturated solutions. Comparable results would then establish the general behavior more firmly.

Procedures for Growth Studies.—The sample of cholesterol monohydrate characterized in connec-

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¹ About 2.5×10^{-8} Gm. ml.⁻¹. (See Reference 3.)