

Deaggregation in Oil-in-Water Emulsions II

Influence of Alcohols and Dioctyl Sodium Sulfosuccinate

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The influence of *n*-butanol, *n*-hexanol, *n*-octanol, and dioctyl sodium sulfosuccinate (AOT) on the deaggregation of a 2 per cent hexadecane-in-water emulsion was studied. It was found that *n*-butanol did not significantly affect the deaggregation rate, *n*-hexanol depressed deaggregation rate at higher concentrations, and both *n*-octanol and AOT were capable of inhibiting deaggregation completely at relatively low concentrations. AOT was observed to depress the rate of deaggregation further than equivalent concentrations of NaCl shown previously. These observations were interpreted as further evidence that both reduction of zeta potential and film-film attractions are important to enhancing aggregation in disperse systems.

PREVIOUS INVESTIGATIONS on the deaggregation of hexadecane in water emulsions stabilized with dioctyl sodium sulfosuccinate¹ (AOT) have shown the phenomenon to be influenced by the presence of electrolytes (1). In the presence of electrolytes, the deaggregation rate is reduced, and the effect is attributed primarily to the reduction in zeta potential of the particles. However, there is some indication that micelle formation, induced by higher electrolyte concentrations, may also be of significance.

It has been found (2-4) that addition of long-chain alcohols to a colloidal system has the same qualitative effect as that of addition of electrolytes in decreasing the critical micelle concentration. The decrease in critical micelle concentration observed upon addition of alcohols was attributed to the formation of mixed micelles (2, 3). It has also been found (5, 6) that higher concentrations of surfactant in emulsion or suspension systems enhance the aggregation of the dispersed particles.

Therefore, it was of interest in these studies to examine the effect of aliphatic alcohols and the concentration of AOT on the deaggregation rate.

EXPERIMENTAL

Materials.—All the aliphatic alcohols used—*n*-butanol, *n*-hexanol, and *n*-octanol—were of laboratory reagent grade. The *n*-hexadecane used in this study (Eastman practical grade) was purified by redistillation under reduced pressure. AOT ("100% pure") was purified by the fractional precipitation

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¹ Marketed as Aerosol OT by the American Cyanamid Co., Pearl River, N. Y.

method (7).² The water used throughout was redistilled from permanganate in a glass apparatus.

Procedure.—A detailed procedure was reported in the first paper (1) which dealt with the effect of electrolyte concentration and temperature on the rate of deaggregation of particles in a hexadecane-water emulsion system. Stock emulsions of 40% by volume were prepared by the electrical dispersion method (8) and were aged for 1 week at 30° in a constant-temperature bath with gentle tumbling to permit equilibration to occur. The stock emulsions were diluted appropriately by the addition of water in order to obtain a 2% *n*-hexadecane emulsion containing 0.09% AOT. Various concentrations of aqueous solutions of alcohols and AOT were added to the system during the dilution of the stock emulsions. The alcohol concentrations were in the range of 5×10^{-4} to 8×10^{-3} *M*, while the concentration of AOT was in the range between 5×10^{-4} and 4×10^{-3} *M*. In each run, the various concentrations of emulsion were prepared by dilution of a single stock emulsion. Then the different samples of emulsion were placed in 15-ml. glass vials and put in the tumbling bath at 30° (26 r.p.m.) for 11-15 days. The volume in each vial was the same (13 ml.).

The number of determinations of each sample of the same concentration varied from three to four, except for those samples at higher concentration of additive where data were highly reproducible.

Particle size analyses were made at different intervals of time with the Coulter counter³ (5). The 50- μ aperture tube used throughout these studies was calibrated previously by using standard polystyrene particles⁴ in two different sizes—1.17- and 1.305- μ diameter.

RESULTS AND DISCUSSION

The previous paper (1) showed that the 2% *n*-hexadecane emulsion containing 0.09% AOT used as the standard emulsion deaggregated under tumbling conditions in a constant temperature bath (30°) and that the rate of deaggregation was constant for approximately 20 days. Deaggregation was indicated by the increase in peak population over the time of

² With a slight modification to the procedure by Higuchi, W. I., and Misra, J., private communication.

³ Coulter Industrial Sales, Chicago, Ill.

⁴ Kindly supplied by J. W. Vanderhoff, Dow Chemical Co., Midland, Mich.

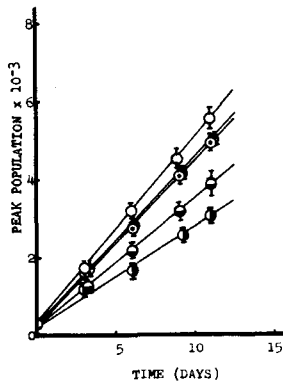


Fig. 1.—Influence of *n*-hexyl alcohol concentration on the rate of deaggregation in 2% *n*-hexadecane emulsion containing 0.09% AOT. Key: O, zero concentration (control); ●, 1×10^{-3} M; ○, 2×10^{-3} M; ●, 4×10^{-3} M; ○, 8×10^{-3} M.

the experiment. The peak population corresponds to the maximum in the particle size distribution of our system.

Deaggregation in the Presence of Alcohols.—To determine the effect of aliphatic alcohols on the deaggregation of this system, various concentrations of *n*-butanol, *n*-hexanol, and *n*-octanol were added to the standard emulsion. In the concentration range studied, 1×10^{-3} to 8×10^{-3} M, *n*-butyl alcohol had an insignificant effect on deaggregation. Some effect was observed with *n*-hexyl alcohol, indicated in Fig. 1, in which peak population of the emulsion system is plotted against time for the emulsions containing differing concentrations of *n*-hexyl alcohol. Figure 1 shows that *n*-hexyl alcohol has little or no effect on the rate of deaggregation at the low concentrations up to 2×10^{-3} M; but as the concentration is increased to 4×10^{-3} M, an inhibition of deaggregation becomes apparent. The effect is greater at a *n*-hexyl alcohol concentration of 8×10^{-3} M.

Figure 2 is a plot of peak population height against time for the system containing *n*-octyl alcohol at various concentrations (5×10^{-4} to 4×10^{-3} M). This figure illustrates the effect of *n*-octyl alcohol on the rate of deaggregation clearly. As the concentration of *n*-octyl alcohol is increased from 5×10^{-4} to 4×10^{-3} M, a successively increasing inhibition of deaggregation is observed to approach complete inhibition at the latter concentration. At concentrations of 3×10^{-3} and 4×10^{-3} M, *n*-octanol completely prevents deaggregation in the system.

Upon comparing the effects of the alcohols studied on the deaggregation phenomenon, it becomes apparent that the chain length of these compounds is important in their inhibitory activity. Further study is necessary to delineate more completely the relationship between molecular structure and enhancement of particle interactions in this or similar systems.

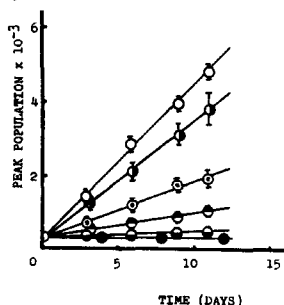


Fig. 2.—Influence of *n*-octyl alcohol concentration on the rate of deaggregation. Key: O, zero concentration (control); ●, 5×10^{-4} M; ○, 1×10^{-3} M; ●, 1.5×10^{-3} M; ○, 2×10^{-3} M; ●, 3 and 4×10^{-3} M.

In our previous studies (1), electrolytes were observed to inhibit the deaggregation of particles in this emulsion system, and the inhibition was attributed to either or both of two possible mechanisms. Very possibly in the presence of electrolytes there is a decrease in the zeta potential (9), caused by the increased concentration of counter ions in the diffuse double layer; thus, the repulsive forces would be reduced. It is also possible that film-film attractions (10) could be operative, especially in the critical micelle concentration region.

In this study, the authors have observed that aliphatic alcohols can inhibit the deaggregation of particles. This inhibition cannot be related to a drop in zeta potential because alcohols do not possess ionic groups in their structures. Thus, the inhibition observed must be due to some other mechanism; the most likely one is that of film-film attraction. That these alcohols are capable of interacting with the AOT present in the emulsion is indicated by their effect on the critical micelle concentration of AOT solutions. The concentration of *n*-octyl alcohol which will induce the formation of micelles in an 0.09% AOT solution was determined by both surface tension measurement of 0.09% AOT solutions containing concentrations of *n*-octyl alcohol

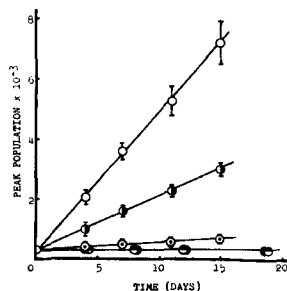


Fig. 3.—Influence of additional AOT concentration on the rate of deaggregation. Key: O, zero addition concentration (control); ●, 5×10^{-4} M; ○, 1×10^{-3} M; ●, 1.5×10^{-3} M; ○, 4×10^{-3} M.

above and below that necessary to induce micelle formation and by determining absorbance of a 1×10^{-4} M pinacyanol chloride solution in 0.09% AOT, to which *n*-octyl alcohol was added in increasing amounts. Surface tension measurements gave a value of 2.0×10^{-3} M, and the dye method gave a value of 2.1×10^{-3} M for the concentration of *n*-octanol which induces an 0.09% AOT solution to form micelles.

It is interesting that this concentration of *n*-octanol corresponds closely to the concentration of *n*-octyl alcohol in the emulsion system at which almost complete inhibition of deaggregation was observed. However, further study is necessary to establish whether a positive relationship exists between micelle formation in these systems and inhibition of deaggregation.

Effect of Dioctyl Sodium Sulfosuccinate on Deaggregation.—It was also of interest to investigate the effect of AOT on deaggregation. Figure 3 is a plot of peak population of the system containing additional AOT at various concentrations (5×10^{-4} to 4×10^{-4} M) versus the time. This figure shows that the effect of AOT on the rate of deaggregation is marked. Increasing the concentration of AOT decreased the slope of the line. Upon addition of 1×10^{-3} moles per liter AOT (approximately 3×10^{-3} M total concentration), almost complete inhibition of deaggregation was observed. An increase in the concentration of added AOT to

as high as $4 \times 10^{-3} M$ did not depress the slope to a negative value, an indication that the system in the initial state was aggregated fully under its experimental environment.

That a simple relationship between micelle formation and inhibition of deaggregation does not exist in these systems is apparent in these experiments. It has been reported previously (1) that the critical micelle concentration of the AOT fraction used in these studies is approximately $2.5 \times 10^{-3} M$. The system at a total AOT concentration of approximately $3 \times 10^{-3} M$ did show some slight deaggregation tendency. Complete inhibition was noted at a total AOT concentration of approximately $3.5 \times 10^{-3} M$ (addition of 1.5×10^{-3} moles per liter AOT to the standard emulsion). Slight deaggregation was also observed in an experiment in which an emulsion containing $6 \times 10^{-3} M$ AOT total concentration (deaggregation completely inhibited for 15 days) was diluted to a final concentration of 1% hexadecane and $3 \times 10^{-3} M$ AOT. Thus, in these systems some deaggregation was apparent, even at AOT concentrations apparently above the critical micelle concentration.

It is interesting to compare the results of the inhibition of deaggregation by AOT to those obtained with NaCl in the previous study (1). In a similar 2% hexadecane in water emulsion containing 0.09% AOT prepared in an identical manner, a concentration of approximately $3 \times 10^{-3} M$ NaCl was required to inhibit deaggregation completely. In AOT, a concentration between 1×10^{-3} and $1.5 \times 10^{-3} M$ additional AOT was effective in completely preventing deaggregation. If this inhibition is related to the decrease in the zeta potential only, it would

be expected that a concentration of $3 \times 10^{-3} M$ AOT would be necessary to inhibit the deaggregation, because AOT should make the same contribution to ionic strength as NaCl, assuming complete dissociation. However, the observed inhibition with AOT is greater, and the difference can be explained only by the existence of some other mechanism operating in the system, either alone or along with a drop in the zeta potential. It seems logical to relate this difference in the inhibition observed to a film-film attraction mechanism, perhaps rendered more effective by multilayer adsorption on the particle at higher AOT concentrations. The results also show that addition of 1×10^{-3} moles per liter AOT results in a greater depression of deaggregation rate than the addition of *n*-octyl alcohol at the same concentration. This would be in general agreement with the concept that increased molecular size of the film components enhances the attractive forces between particles which cause the aggregation.

REFERENCES

- (1) Lemberger, A. P., and Mourad, N., *THIS JOURNAL*, **54**, 229(1965).
- (2) Ooshika, Y., Wall, R., and Adams, G., *J. Colloid Sci.*, **13**, 441(1958).
- (3) Herzfeld, S. H., Corrin, M. L., and Harkins, W. D., *J. Phys. Chem.*, **54**, 271(1950).
- (4) Ralston, A. W., and Eggenberger, D. N., *J. Am. Chem. Soc.*, **70**, 983(1948).
- (5) Higuchi, W. I., Okada, R., and Lemberger, A. P., *THIS JOURNAL*, **51**, 683(1962).
- (6) Moore, A., and Lemberger, A. P., *ibid.*, **52**, 223(1963).
- (7) Williams, E. F., Woodberry, N. T., and Dixon, J. K., *J. Colloid Sci.*, **12**, 452(1957).
- (8) Nawab, M. A., and Mason, S. G., *ibid.*, **13**, 179(1958).
- (9) Greiner, L., and Vold, R. D., *J. Phys. Colloid Chem.*, **53**, 87(1949).
- (10) Cockbain, E. G., *Trans. Faraday Soc.*, **48**, 185(1952).

Phytochemical Investigation of *Acacia angustissima*

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Preliminary pharmacological screening of the ethanol and aqueous extracts of *Acacia angustissima* has revealed sedative and antitumor effects. A phytochemical study has established the presence in the defatted ethanol extract of the leucoanthocyanidin moiety, 7, 3', 4'-trihydroxyflavan-3,4 diol (I). The intractable amorphous ethanol extract, after partial solvent fractionation, was separated into seven fractions with a 100-stage countercurrent distribution. Three of these seven fractions contained the same leucoanthocyanidin unit (I). A fourth countercurrent fraction has been characterized tentatively as a flavone or flavonol compound. Further studies are being conducted to determine the extent of polymerization of the three fractions and to isolate the monomer leucoanthocyanidin.

ACACIA ANGUSTISSIMA (Mill) Kuntze, *Leguminosae*, grows as a small shrub or bush at elevations above 3000 ft. and can be found in Arizona, Texas, Missouri, southern Florida, and

South America. In the past, the *Acacia* genus has served as a source of condensed tannins for the tanning industry (1) and as a source of the dried gummy exudate used as a demulcent, emulsifying agent, and vehicle in the preparation of emulsions and troches.

Various concentrations of acacia were tested by Pollia (2) against sarcoma tumors in rats, but no inhibition of the tumor growth was observed. Antitumor properties have been demonstrated in the aqueous, chloroform-ethanol, and ethanol extracts of the pulverized leaves, stems, and

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