Influence of HLB Values of Surfactants on Ephedrine Release from Emulsified Liquid Systems

W. G. WAGGONER* and J. H. FINCHER

Abstract □ This study treated primarily the release characteristics of ephedrine from liquid emulsion systems as they were affected by the hydrophilic–lipophilic nature of the emulsifier present. Polysorbate 60 and sorbitan monostearate were varied by weight percent to produce emulsifiers having different hydrophilic–lipophilic balance (HLB) values. The experiments utilized a dialysis technique and consisted of seven controls plus the study of the emulsified systems throughout an HLB range of 4.7–14.9. The data indicated that the HLB value of the surfactants affected the release rate of ephedrine from an emulsified system, with the largest release rates occurring in the upper HLB range. The release of ephedrine from the oil phase was shown to be the rate-limiting step in its release from the emulsion. Surfactants did not impair the release of ephedrine from an oil system (control), whereas an inhibitory effect was noted in the water system (control). The data also indicated than an interfacial barrier was present in the emulsion and that ephedrine was released at two different rates from each of the various systems; the initial rate was greater than the second.

Keyphrases □ HLB values, surfactants—influence on ephedrine release characteristics from emulsified liquid systems, dialysis technique □ Surfactants, HLB values—effect on ephedrine release rates, oil and water emulsified systems □ Ephedrine release rates—effect of surfactant HLB values, oil and water emulsified systems, dialysis technique □ Hydrophilic–lipophilic balance—effect of polysorbate 60 and sorbitan monostearate values on ephedrine release rates in emulsified systems

The fact that emulsions are used as vehicles for medicinal agents is well established (1), and many investigators have studied the release of various drugs from liquid and semisolid systems (2–6). During most of these studies, the composition of the emulsion was considered to be an important factor affecting drug release. One important component known to influence drug release is the surfactant, and the hydrophilic–lipophilic balance (HLB) is considered to be one of the most important physical properties affecting release of drugs from vehicles (7–9). The results of most studies involving HLB and its effects have been elusive and inconclusive. Micellar solutions (10) and interfacial barriers (11) have been considered also as important factors influencing drug release.

The objective of this research was to determine the influence of the HLB value of the surfactant on the release rate of ephedrine from emulsified, liquid systems.

EXPERIMENTAL

The emulsifiers were used as received from the manufacturer since they were felt to be representative of those used in industry or in a pharmacy. An HLB range of about 5–15 was chosen, and the emulsifiers were picked accordingly. Polysorbate 60[1] with an HLB of 14.9 and sorbitan monostearate[2] with an HLB of 4.7 were considered satisfactory, since the only difference between the two molecules was the weight percent of polyoxyethylene groupings. This procedure allowed evaluation of emulsions and their parameters at HLB values of 4.7, 6.0, 7.0, . . . , 14.0, and 14.9. The drug, ephedrine[3], was selected since it was known to be soluble in both oil and water and could be assayed easily using spectrophotometric methods.

To ensure uniformity, all experimentation was performed on a weight basis with the drug supplying 1%, the surfactants 5%, and the paraffin oil[4] and water each 47%.

Phase I: Preliminary Procedures—*Preparation of Drug and Surfactants*—The moisture content of each surfactant was determined by using moisture balance[5]. The hydrous ephedrine was dried in a desiccator over concentrated sulfuric acid for a minimum of 72 hr. to remove the water of hydration.

Determination of HLB Values—The concentration of the surfactant required to obtain the desired HLB value was calculated by simple allegation, and then the correction factor was applied for the moisture contained in the surfactants. The average moisture was 3 and 2.5% for the polysorbate 60 and the sorbitan monostearate, respectively.

Assay Procedures—Aqueous solutions of ephedrine were assayed using a spectrophotometer[6] at 257 nm.

Dialysis Procedure—Two dialysis cells[7] (flowthrough type), each with a capacity of 10 ml., were used.

Standard dialysis membranes[8], having an average pore diameter of 4.8 nm., were soaked in distilled water at least 24 hr. for hydration purposes prior to use for dialysis. A thin coating of stopcock grease was applied to the inner walls of the cells before fitting the membrane to ensure there was no leakage from the compartments.

In the initial preparation, after the membrane was placed in position, the cells were weighed and exactly 10 g. of solution was introduced into each cell, thereby eliminating such factors as entrapment of air and viscosity. The weight of the sink was 100 g.; since the sink was water, the volume approximated 100 ml.

Two cells were arranged in series and immersed in a constant-temperature bath maintained at 37.5° by a power unit[9] and heater circulator. Connected to the inlet side of the dialysis cells by latex rubber tubing was a pump[10], which was used to maintain a constant flow rate of 60 ml./min. through the dialysis cells. This flow rate was maintained throughout all steps of the experimentation.

A rubber tube from the outlet side of the dialysis cells emptied into a 500-ml. round-bottom flask, which was used as the reservoir sink. This was also immersed in the constant-temperature bath and was used for the purpose of collecting samples. The samples were collected at definite time intervals, their UV absorbance was determined, and they were immediately returned to the sink, thereby negating the use of correction and/or dilution factors due to the loss of drug or volume of liquid. Connections were made to the inlet side of the pump from the reservoir, thereby completing a full circle and allowing the flow of liquid.

Phase II: Control Study—*Surfactant Dialysis from Water to Aqueous Sink*—To determine whether the surfactants would permeate the membrane, a separate surfactant–water solution or dispersion was prepared with each surfactant by introducing 5 g. of the surfactant into 95 g. of water, heating to 65° with stirring, and then cooling to 40°. Each surfactant–water solution or dispersion was dialyzed twice according to the previously described procedure.

[1] Tween 60, Atlas Chemical Corp., Wilmington, Del.
[2] Arlacel 60, Atlas Chemical Corp., Wilmington, Del.
[3] Ephedrine NF (hydrous), Merck and Co., Inc., Rahway, N. J.
[4] Paraffin oil USP, Fisher Scientific Co., Fair Lawn, N. J.
[5] Ohaus Scale Corp., Union, N. J.
[6] Model DB-G, Beckman Instruments, Inc., Fullerton, Calif.
[7] The Chemical Rubber Co., Cleveland, Ohio.
[8] Oxford Laboratories, San Mateo, Calif.
[9] Thermonitor, E. H. Sargent & Co., Cleveland, Ohio.
[10] Zero-Max Kinetic Clamp, Sigmamotor, Inc., Middleport, N. Y.

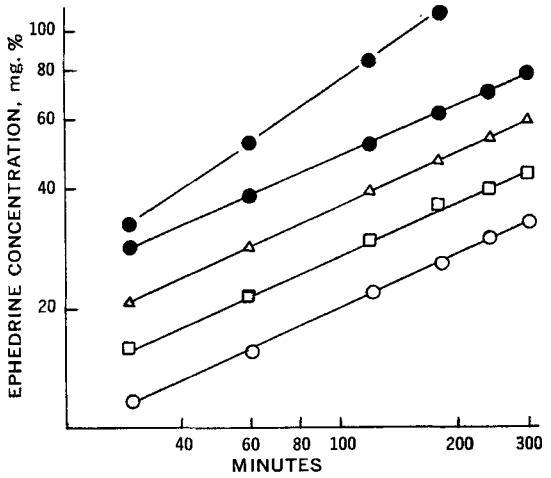

Figure 1—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 4.7. Each point on the figure represents the mean of two separate dialyses. Key: ●, ephedrine + water; △, ephedrine + water + surfactant; ●, ephedrine + oil; □, ephedrine + oil + surfactant; and ○, ephedrine emulsion.*

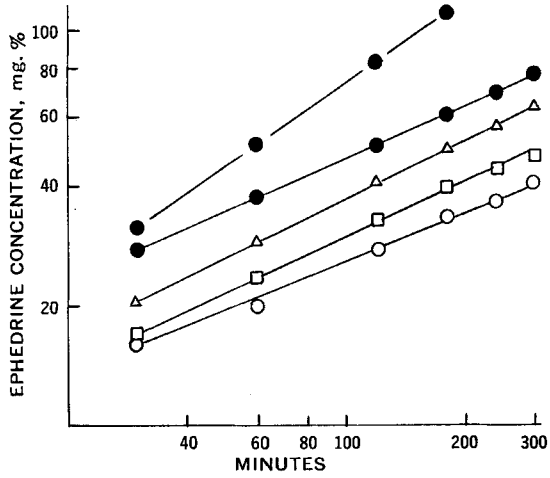

Figure 3—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 7.0. Each point on the figure represents the mean of two separate dialyses. Key: ●, ephedrine + water; △, ephedrine + water + surfactant; ●, ephedrine + oil; □, ephedrine + oil + surfactant; and ○, ephedrine emulsion.*

The surface tension of the sink was checked at the beginning of the run and at intervals throughout a total dialysis time of 13–17 hr. Surface-tension measurements were performed using a tensiometer[11]. Since the surface tension of the sink did not change significantly during the extended dialyses, it was concluded that sorbitan ester[12] and polysorbate 20 did not dialyze.

Phase III: Control Study—*Dialysis of Ephedrine from Water to Aqueous Sink*—One gram of ephedrine was dissolved in 99 g. of water, heated to 65° with stirring, and then cooled to 40°. Dialysis, sampling, and assay were performed according to previously described procedures.

Phase IV: Control Study—*Dialysis of Ephedrine from Oil to Aqueous Sink*—One gram of ephedrine was dissolved in 99 g. of oil, heated to 65° with stirring, and then cooled to 40°. The dialysis procedure was performed and analyzed as in Phase III.

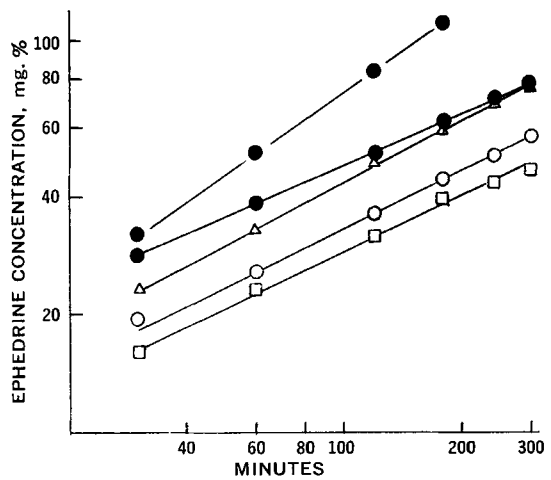

Figure 2—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 6.0. Each point on the figure represents the mean of two separate dialyses. Key: ●, ephedrine + water; △, ephedrine + water + surfactant; ●, ephedrine + oil; □, ephedrine + oil + surfactant; and ○, ephedrine emulsion.*

[11] Cenco DuNouy, Central Scientific Co., Chicago, Ill.
[12] Span 20.

Phase V: Control Study—*Dialysis of Ephedrine from Surfactant–Water Systems at Each HLB*—For each HLB value, each surfactant was weighed in the proper ratio to effect the desired HLB value. The water and drug were weighed and added in sufficient amounts to bring the total weight of the mixture to 100 g. (5 g. surfactant–1 g. drug–94 g. water). This mixture was heated to 65° with stirring on a hot plate-magnetic stirrer and cooled to 40°. The sorbitan monostearate was not soluble but dispersed readily with stirring. Dialysis, sampling, and assay were performed as previously described.

Phase VI: Control Study—*Dialysis of Ephedrine from Surfactant–Oil Systems at Each HLB*—For each HLB value, the surfactant, drug, and oil were combined in a weight ratio of 5:1:94 in an analogous manner to Phase V. The mixture was heated with stirring to 60° and cooled to 40°. Stirring was necessary to disperse the insoluble polysorbate 60. Dialysis, sampling, and assay were performed as previously described.

Phase VII: Control Study—*Distribution Coefficients at Each HLB Value*—The distribution apparatus was assembled using a 150-ml. beaker with a 7.62-cm. (3-in.) piece of glass tubing (8 mm. in diameter) glued to the inside wall. This tubing extended to within 1.27 cm. (0.5 in.) of the bottom. The apparatus permitted sampling

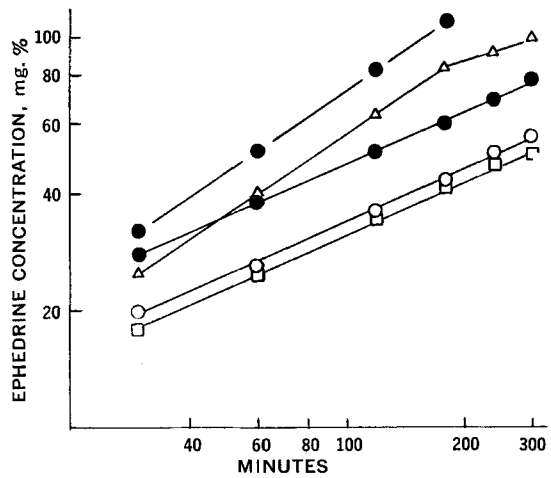

Figure 4—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 8.0. Each point on the figure represents the mean of two separate dialyses. Key: ●, ephedrine + water; △, ephedrine + water + surfactant; ●, ephedrine + oil; □, ephedrine + oil + surfactant; and ○, ephedrine emulsion.*

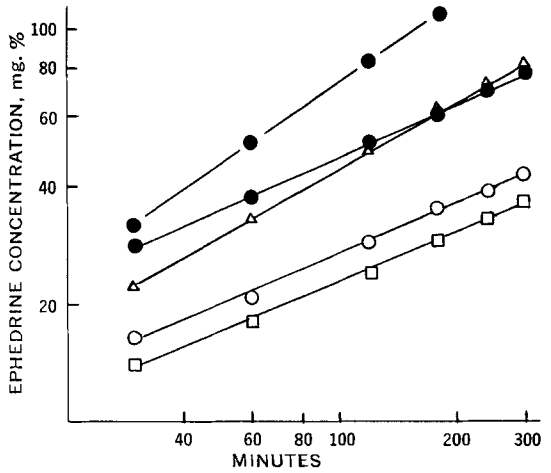

Figure 5—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 9.0. Each point on the figure represents the mean of two separate dialyses. Key:* ●, *ephedrine + water;* △, *ephedrine + water + surfactant;* ●, *ephedrine + oil;* □, *ephedrine + oil + surfactant; and* ○, *ephedrine emulsion.*

of the aqueous phase (lower phase) without disturbance to the liquid interface and allowed the interfacial area to remain constant in all studies.

The water and oil phases for the determination of the distribution coefficients were prepared exactly as those for emulsions. The water phase (water–surfactant–drug) was poured into the distribution apparatus; then the oil phase (oil–surfactant) was carefully placed on the surface of the water phase. The beaker was gently placed in a constant-temperature bath without disturbing the interface between the two liquids. At definite intervals, samples were drawn from the water phase and analyzed for the drug content. The sample was collected about 6 mm. (0.25 in.) from the bottom of the beaker in every case, because this was felt to give a more representative sample than from the tube.

To determine the amount of ephedrine in the oil phase, a 1-g. sample of the oil was drawn, diluted with 20 ml. of 0.1 *N* HCl, and shaken gently for 30 min. On acidifying the oil, the ephedrine should become completely water soluble due to the formation of ephedrine hydrochloride. After shaking, the sample was allowed to stand for 10 min. so the oil would separate. A small amount of emulsification occurred during this phase. The sample to be analyzed was drawn from the lower portion of the liquid (acid solution) and

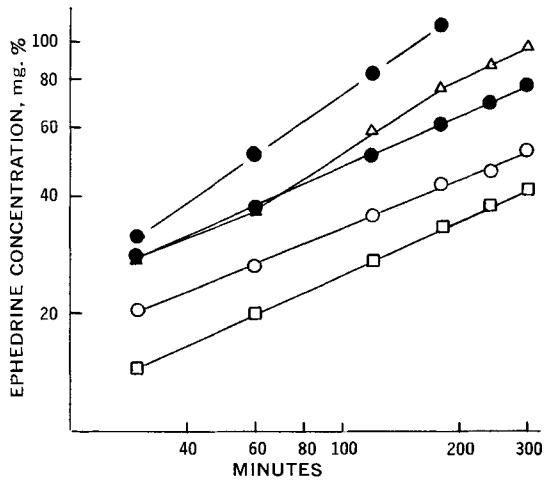

Figure 6—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 10.0. Each point on the figure represents the mean of two separate dialyses. Key:* ●, *ephedrine + water;* △, *ephedrine + water + surfactant;* ●, *ephedrine + oil;* □, *ephedrine + oil + surfactant; and* ○, *ephedrine emulsion.*

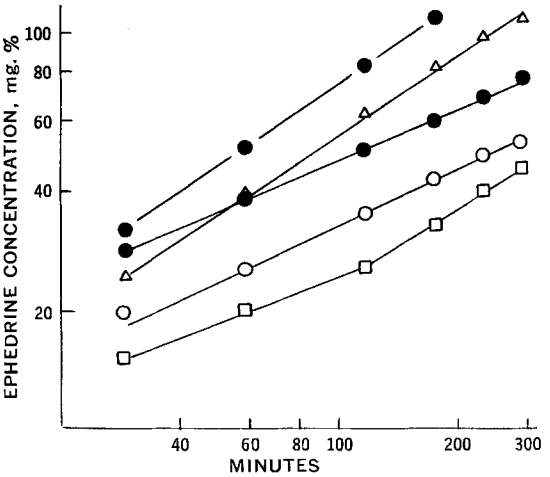

Figure 7—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid sytems in which the emulsifier had an HLB value of 11.0. Each point on the figure represents the mean of two separate dialyses. Key:* ●, *ephedrine + water;* △, *ephedrine + water + surfactant;* ●, *ephedrine + oil;* □, *ephedrine + oil + surfactant; and* ○, *ephedrine emulsion.*

passed through a filter system[13] utilizing a filter of 0.8-μ pore diameter. The sample was then analyzed spectrophotometrically at 257 nm. For this assay procedure, a new standard curve was prepared using standards of ephedrine in 0.1 *N* HCl.

An attempt was made to sample the interface between the oil and water using a syringe. The needle of the syringe was inserted through the oil phase (top layer), and samples were drawn at the interface of the two liquids. Much spontaneous emulsification had occurred at the time of sampling, and it was impossible to determine the exact position of the interface. The layer of spontaneous emulsification varied from about 1 to 8 mm., becoming increasingly greater as the HLB value was lowered. Analysis procedures were the same as reported for the oil samples.

Phase VIII: Emulsion Study—The surfactants, previously calculated for the desired HLB value, were weighed in separate 100-ml. beakers. One gram of ephedrine, dissolved in 47 g. of water, was added to the water-soluble surfactant and heated with stirring to 65°. Forty-seven grams of oil was added to the beaker with the oil-soluble surfactant and heated with stirring to 60°. The water phase was poured into a previously warmed glass container of a blender.

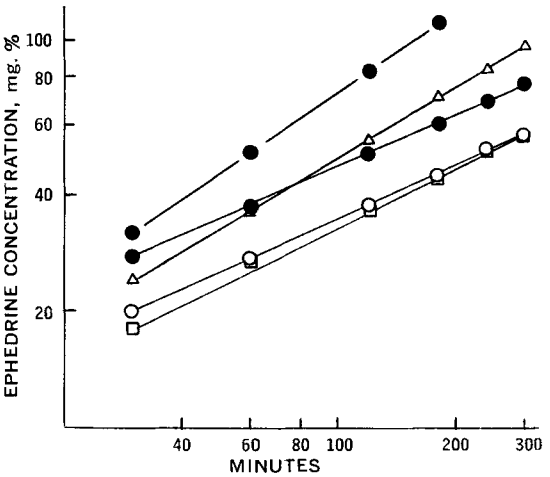

Figure 8—*Results of dilayses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 12.0. Each point on the figure represents the mean of two separate dialyses. Key:* ●, *ephedrine + water;* △, *ephedrine + water + surfactant;* ●, *ephedrine + oil;* □, *ephedrine + oil + surfactant; and* ○, *ephedrine emulsion.*

[13] Millipore Corp., Bedford, Mass.

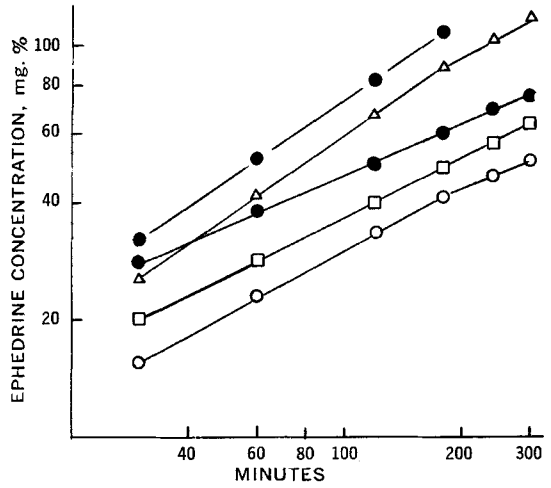

Figure 9—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 13.0. Each point on the figure represents the mean of two separate dialyses. Key:* ●, *ephedrine + water;* △, *ephedrine + water + surfactant;* ●, *ephedrine + oil;* □, *ephedrine + oil + surfactant; and* ○, *ephedrine emulsion.*

The blender was adjusted to the lowest speed, and the oil phase was rapidly poured into the water phase. An emulsion immediately formed, and the blender ran exactly 30 sec. The contents were transferred to a 250-ml. beaker and allowed to equilibrate for 1 hr. At the end of 1 hr., the contents were gently swirled by hand and transferred to dialysis cells. Dialysis procedures were initiated according to standardized procedures previously described.

The emulsion was tested as to type, o/w or w/o, by dilution techniques; all emulsions were found to be o/w.

RESULTS AND DISCUSSION

The results of Phase I, preliminary preparations, and Phase II, surfactant dialyses, were discussed previously in the *Experimental* section. It is well to reiterate that in Phase II the surfactants did not dialyze.

The results of the dialyses in Phases III–VI and VIII are reported in Figs. 1–11, and the data fit a log–log-type function with respect to time and concentration. In these plots, the control curves representing the dialyses of ephedrine from water and ephedrine from oil were repeated each time for ease of comparison. Thus, the

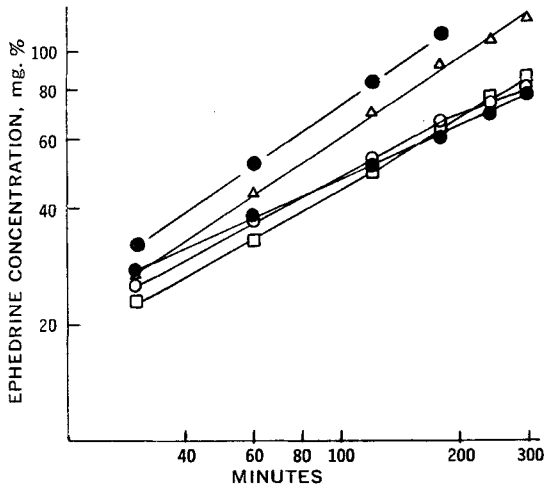

Figure 10—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 14.0. Each point on the figure represents the mean of two separate dialyses. Key:* ●, *ephedrine + water;* △, *ephedrine + water + surfactant;* ●, *ephedrine + oil;* □, *ephedrine + oil + surfactant; and* ○, *ephedrine emulsion.*

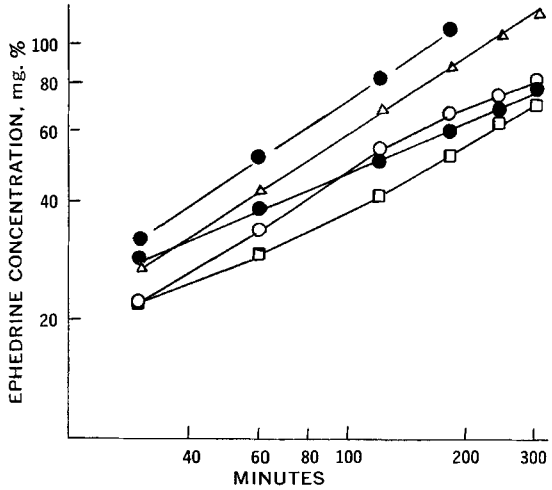

Figure 11—*Results of dialyses of ephedrine from mineral oil in water emulsified, liquid systems in which the emulsifier had an HLB value of 14.9. Each point on the figure represents the mean of two separate dialyses. Key:* ●, *ephedrine + water;* △, *ephedrine + water + surfactant;* ●, *ephedrine + oil;* □, *ephedrine + oil + surfactant; and* ○, *ephedrine emulsion.*

equations for the the curves had the same general formula:

$$y = bt^n \qquad \text{(Eq. 1}a\text{)}$$

$$\log y = \log b + n \log t \qquad \text{(Eq. 1}b\text{)}$$

where y = concentration of drug in milligrams percent, b = intercept, n = slope of the line, and t = time in minutes.

Since Figs. 1–11 cannot contain the 0% point, they exemplify the secondary or "steady-state" release pattern. The initial release rate was much faster and represents an equilibration period brought about by an initial surface release and/or a rapid diffusion of the drug from the aqueous phase until such release is controlled by either diffusion from the oil phase or diffusion through the interfacial area. The intercepts (not shown as such) of the log–log plots in Figs. 1–11 are indicative of the initial release pattern.

From the n values (Table I), some observations may be made about the secondary release rates from the various systems.

Drug Release from Water *versus* Drug Release from Oil—The release rate of ephedrine from the water (n = 0.725, antilog 5.31) was much greater than its release rate from the oil (n = 0.446, antilog 2.79). This was attributed to a slower diffusion rate of ephedrine through the more viscous oil (Table I).

Drug–Oil (n = 0.446, Antilog 2.79) *versus* Drug–Surfactant–Oil Release at Different HLB Values—Only HLB values 8 and 9 were below the control value of 0.446, and those were only slightly below. All others were above or considerably above the control value. This indicated that the surfactants in oil did not hinder the release rate of the drug; in most cases, as the HLB value approached

Table I—Calculated Slopes (n) of Ephedrine Concentration *versus* Time Plots Shown in Figs. 1–11[a]

	Ephedrine Emulsion		Ephedrine–Oil–Surfactant		Ephedrine–Water–Surfactant	
HLB	n	Antilog	n	Antilog	n	Antilog
4.7	0.435	2.72	0.447	2.80	0.472	2.96
6.0	0.496	3.13	0.460	2.88	0.535	3.43
7.0	0.427	2.67	0.472	2.96	0.493	3.11
8.0	0.435	2.84	0.440	2.75	0.673	4.71
9.0	0.418	2.62	0.442	2.77	0.562	3.64
10.0	0.423	2.65	0.453	2.84	0.640	4.37
11.0	0.473	2.97	0.446	2.79	0.667	4.64
12.0	0.457	2.86	0.497	3.14	0.610	4.07
13.0	0.547	3.53	0.507	3.17	0.704	5.06
14.0	0.548	3.53	0.600	3.98	0.677	4.75
14.9	0.610	4.07	0.598	3.96	0.660	4.57

[a] n_w = 0.725, antilog 5.31 (ephedrine–water). n_o = 0.446, antilog 2.79 (ephedrine–oil).

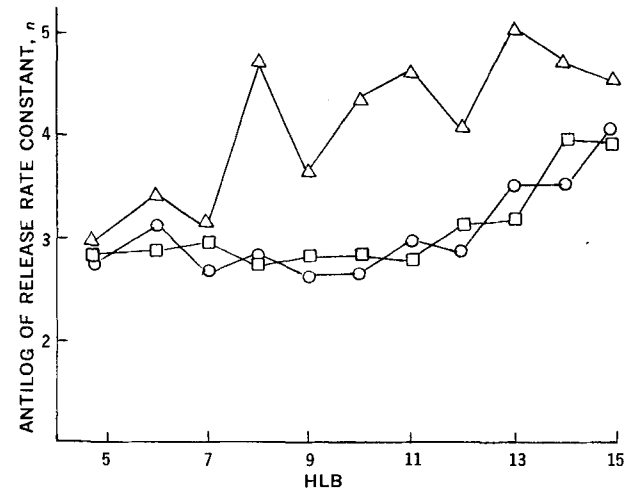

Figure 12—*Ephedrine dialysis rate constant as a function of HLB value of surfactant in various systems. Key:* ○, *ephedrine emulsion;* □, *ephedrine–oil–surfactant; and* △, *ephedrine–water–surfactant.*

14.9, the surfactant seemed to enhance the release of the drug from the oil (Table I).

Drug–Water (n = 0.725, Antilog 5.31) *versus* Drug–Surfactant–Water Release at Different HLB Values—In no case did the drug release from the drug–surfactant–water mixtures exceed the control value of 0.725, antilog 5.31, which indicated that the drug was interacting to a certain degree with the surfactants in water. This impaired release probably was due to a physical trapping of the drug in the micelles. As the HLB values increased from 4.7 to 14.9, the n values became larger, showing a greater release rate. This indicated that increasing the weight percent of polyoxyethylene groups tended to enhance the release rate of the drug (Table I).

Ephedrine Emulsion *versus* Controls at Different HLB Values—Figure 12 is a plot of the n values of all systems *versus* the HLB value of the surfactant in the separate systems. For each HLB value, no n value of the emulsified system was as large as the n value of the ephedrine–water control (n = 0.725, antilog 5.31) or the ephedrine–surfactant–water control. This again indicated that the emulsified system inhibited the release rate of the drug over the control values already described. However, for the control value of drug–oil (n = 0.446, antilog 2.79), the emulsified system did not show this inhibitory effect and, in fact, seemed to enhance the release rate of the drug over the drug–oil control at many HLB values.

Essentially, a zero slope was obtained with the n values of the emulsion and ephedrine–oil–surfactant control until an HLB value

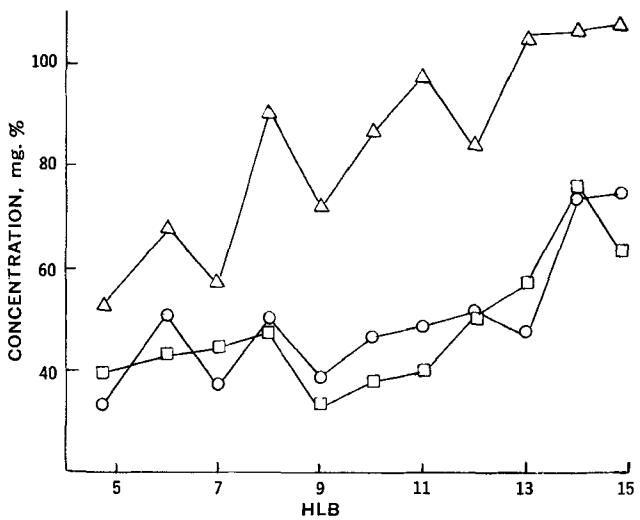

Figure 13—*Ephedrine concentration after 240 min. of dialysis as a function of HLB values of surfactant in various systems. Key:* △, *ephedrine–water–surfactant;* ○, *ephedrine emulsion; and* □, *ephedrine–oil–surfactant.*

of 12 was reached where the slope became positive. It was interesting to note the intertwining effect of the two values. At HLB 9–10, this was suddenly reversed, which could indicate the formation of the most stable emulsion.

From the close proximity of the curves and the intertwining effect of the release rates of the ephedrine–surfactant–oil and the emulsion systems at the various HLB values, it was concluded that the release rate from the oil system was the controlling factor in the release of the drug from the emulsified system.

The release of the drug from the water system was faster than the oil system at every comparable HLB value. Thus, the drug release from the oil was the rate-limiting factor.

Previously, it was shown that the surfactants did not retard the release of the drug from oil systems. Consequently, it would be expected from the control values that the rate of drug release from the emulsion would be the same or greater than the surfactant–oil system. However, in the emulsified system, at HLB values of 4.7, 7.0, 9.0, 10.0, 12.0, and 14.0, a lower rate of drug release was obtained than from the surfactant–oil system (Table I). This behavior was strongly indicative of the existence of an interfacial barrier between the oil globules and the water, which could be due to the arrangement and packing of the surfactants at the interface.

The concentration of ephedrine (milligrams percent) attained in the sink in a time interval of 240 min. reflects the net result of both release patterns during the time period in question (Fig. 13). The differences in the general shapes of Fig. 13 from Fig. 12 reflect the influence of the initial release rate on the total amount of drug dialyzed. The shape of the ephedrine–water–surfactant curves in Figs. 12 and 13 are the same except at HLB values of 13 and 14.9. The reasons for these apparent differences are not known. The absence of an oil–water interface in the ephedrine–surfactant–water system, the similarity of the curves, and the dissimilarity of the analogous emulsion curves further substantiate the possibility that transport of the drug through the interface in an emulsion system can be inhibited. This inhibition is apparently a function of the HLB value of the surfactant.

The fit of the data observed in the secondary or true rate of release to a log–log-type function (Freundlich isotherm type) indicates that a surface adsorption–evaporation process is occurring.

Inflection points were noted in all curves in Fig. 12 involving slope as a function of HLB. It is possible that at these points the shape of the micelle changes. Beecher (12) proposed that the micellar properties of nonionic surface-active agents could best be interpreted in terms of a rod-shaped micelle for the more lipophilic agents and a spherical micelle for the more hydrophilic agents. A change in shape would produce a change in the surface area of the micelle. Consequently, if the drug interacted with the micelle, the availability of the drug for dialysis would be changed.

Microscopic examination of the emulsions with the help of a micrometer eyepiece revealed that globules in all emulsions were essentially the same size; thus, change in the surface area of the droplets was not a factor in the fluctuations. The consistency of two separate dialyses conducted at each HLB value indicates these fluctuating points to be real differences and not random error in the experiments.

The distribution coefficients were attempted in accordance with the procedures previously described. Many problems became apparent in this study. In an effort to keep all parameters constant, no change was made in the concentration of the surfactants, drug, or phase volumes. The interfacial area between the oil and water was much less than when emulsified. Consequently, the water-soluble surfactant, polysorbate 60, settled out of solution and interfered with the analysis procedures from the water phase. The interference became appreciable around an HLB value of 10. This was not noticed at the lower HLB values; however, since the interference did become appreciable, all data must be regarded as less than exact. In an effort to salvage some of the experimentation, it was decided to sample the oil on the last set of distribution coefficients. This was described in the experimental procedures. Below an HLB value of 10, there was a greater loss of ephedrine from the water solution than was recovered in the oil solution. Above this value, recovery seemed more complete. This indicates that a significant amount of the drug was collecting at the interface, thereby indicating an interfacial barrier as postulated previously. Samples collected at the interface indicated a higher concentration of drug than would be expected, lending further support to the postulation. A control study indicated that the ephedrine

was stable over a 7-day period and that results were not influenced by degradation.

REFERENCES

(1) E. Sagarin, "Cosmetics, Science, and Technology," Interscience, New York, N. Y., 1957, pp. 998, 999.

(2) L. D. Lockie and J. B. Sprowls, *J. Amer. Pharm. Ass., Sci. Ed.*, **40**, 72(1951).

(3) J. A. Wood, L. W. Rising, and N. Hall, *J. Pharm. Sci.*, **51**, 669(1962).

(4) W. I. Higuchi, *Proc. Amer. Ass. Coll. Pharm. Teachers Seminar*, **13**, 162(1961).

(5) T. Koezumi and W. I. Higuchi, *J. Pharm. Sci.*, **57**, 87(1968).

(6) J. A. Wood and L. W. Rising, *J. Amer. Pharm. Ass., Sci. Ed.*, **42**, 481(1953).

(7) J. W. Rhyne, W. J. Payne, and C. W. Hartman, *ibid.*, **49**, 234(1960).

(8) R. Y. Spittle and C. W. Hartman, *ibid.*, **49**, 325(1960).

(9) J. H. Fincher, D. N. Entrekin, and C. W. Hartman, *J. Pharm. Sci.*, **55**, 23(1966).

(10) A. H. Goldberg, W. I. Higuchi, N. F. H. Ho, and G. Zografi, *ibid.*, **56**, 1432(1967).

(11) A. H. Goldberg and W. I. Higuchi, *ibid.*, **58**, 1341(1969).

(12) P. Beecher, *J. Colloid Sci.*, **16**, 49(1961).

ACKNOWLEDGMENTS AND ADDRESSES

Received May 10, 1971, from the *Department of Pharmaceutics, School of Pharmacy, University of Mississippi, University, MS 38677*

Accepted for publication August 26, 1971.

Presented to the Basic Pharmaceutics Section, APhA Academy of Pharmaceutical Sciences, San Francisco meeting, March 1971.

Abstracted in part from a dissertation submitted by W. G. Waggoner to the Graduate School, University of Mississippi, University, MS 38677

Supported by the N.D.E.A. Fellowship Program and the University of Mississippi Research Institute of Pharmaceutical Sciences.

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Normal and Promoted GI Absorption of Water-Soluble Substances III: Absorption of Antibiotics from Stomach and Intestine of the Rat

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Abstract □ Absorption of soluble antibiotics was studied in the doubly ligated stomach of the rat, in the ligated small intestine, and in the intact GI tract. When surface-active absorption promoters are administered with the antibiotic in the ligated stomach, promoted absorption occurs but the onset is slow. When the drug–promoter combination is put in the ligated intestine, blood levels of the drug are elevated to severalfold the normal levels and the rise is extremely rapid. Absorption promoters exert a smaller influence in the intact GI tract. Their effectiveness is confined to the first 30 min., after which approximately normal levels of drug are seen. Emptying of the liquid dose from the stomach at 6–8 min. was followed by an immediate rapid rise of blood levels characteristic of the rapid response of the intestine to promoters. Accordingly, the blood level *versus* time curves obtained when promoters are employed in the intact rat are interpreted to be a result of a rapid but transient promoted absorption in the duodenum–small intestine, with little contribution from promoted gastric absorption.

Keyphrases □ Surfactants—role in enhancing antibiotic absorption, stomach, small intestine, and GI tract compared, rats, toxicity □ Antibiotics, soluble—comparison of doubly ligated stomach, ligated small intestine, and intact GI tract absorption, rats, effect of surfactants □ Absorption, soluble antibiotics—role of surfactants in doubly ligated stomach, ligated small intestine, intact GI tract, rats □ Surface-active absorption promoters—influence on water-soluble antibiotics in stomach and intestine, rats

Absorption of normally nonabsorbed or poorly absorbed water-soluble drugs from a Thomas gastric fundic pouch of the dog is greatly increased by certain surface-active agents (1). Nonionic, anionic, cationic, and zwitterionic surface-active agents were shown to promote the absorption of several types of antibiotics. Further work demonstrated similar promoted absorption in other segments and in the intact GI tract of the dog.

Another study demonstrated a greatly increased absorption of vitamin B_{12} from the stomach and the intact GI tract of the rat when an absorption promoter was added (2). The present study was initiated to characterize the response of the rat to absorption promoters and, particularly, to distinguish the response of the stomach and the intestine.

EXPERIMENTAL

Handling of Animals—Male Wistar and CFE strain rats, weighing 150–300 g., were fasted in individual cages with wide-mesh screen floors for approximately 15–20 hr. prior to operation and dosing. Blood samples were obtained by amputating the tip of the tail. Enough blood was collected to fill the capillarity capacity of from three to six 0.63-cm. (0.25-in.) diameter filter pads. The filter pads were placed directly on agar plates and were assayed for microbiological activity of the drug by standard disk-plate procedures[1].

Surgical Procedure—The animal was anesthetized with ether, and the stomach was exposed through a midline incision in the abdominal area. A ligation was placed at the pyloric sphincter. Aqueous drug solution was administered by stomach tube, after which an esophageal ligation was made at the cardiac sphincter, care being taken not to occlude blood vessels. The abdominal incision was then closed with wound clamps. After the rat recovered from the anesthesia in a restraining cage, it was conscious throughout the remainder of the experiment. Blood samples were periodi-

[1] Cephalothin and penicillin V were assayed against *Bacillus subtilis* (ATCC 6633). Cephaloridine was assayed against *Sarcina lutea* (PC1-1001-FDA).