Critical Reviews™ in Therapeutic Drug Carrier Systems, 18(1):77–140 (2001)

Improved Drug Delivery Using Microemulsions: Rationale, Recent Progress, and New Horizons

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ABSTRACT: Microemulsions are excellent candidates as potential drug delivery systems because of their improved drug solubilization, long shelf life, and ease of preparation and administration. The formulation of microemulsions for pharmaceutical use requires a thorough understanding of the properties, uses, and limitations of microemulsions. Three distinct microemulsions—oil external, water external, and middle phase—can be used for drug delivery, depending upon the type of drug and the site of action. In this article, we present an examination of microemulsions as drug carrier systems, starting with general information and moving to a thorough review of the microemulsion literature, with a special section devoted to microemulsion-based gels.

KEYWORDS: pharmaceutical microemulsion, micelles, microemulsion based gels, targeted colloidal drug delivery, solubilization of hydrophilic and hydrophobic compounds.

I. INTRODUCTION

The design and development of new drug delivery systems with the intention of enhancing the efficacy of existing drugs is an ongoing process in pharmaceutical research. It is necessary for a pharmaceutical solution to contain a therapeutic dose of the drug in a volume convenient for administration. Of the many types of drug delivery systems that have been developed, one in particular—the colloidal drug delivery system—has great potential for achieving the goal in drug targeting.

This review article will focus on defining this current technology of microemulsions in relation to pharmaceutical applications. Section II discusses the historical development of microemulsions and various other applications of microemulsions. Section III delineates the conditions necessary to produce microemulsions, as well as some interesting characteristics

0743-4863/01 $5.00

of microemulsion formulations. It also discusses the methods most commonly employed to determine microemulsion properties. Section IV discusses microemulsion formulations in the pharmaceutical industry, beginning with an introduction to various methods of drug delivery and the important factors that influence each method. This section then discusses microemulsion formulations for drug delivery using nonionic surfactants, anionic surfactants, phospholipids and cholesterol, and other surfactants. Each formulation is reviewed in terms of its application. Section V discusses microemulsion-based gels (MBG), which are a direct spin-off of microemulsion technology. This section addresses the advantages and disadvantages of gel formulations and discusses various gel formulation strategies and the current level of application of each formulation. The final section summarizes and discusses future research for microemulsions as a drug delivery system.

A. Types and Properties of Colloidal Drug Delivery Systems

A few of the most widely examined colloidal drug delivery systems are micelles, microemulsions, macroemulsions (hereafter referred to as *emulsions*), niosomes, liposomes, and nanoparticles.[1] These colloidal systems are shown schematically in Figure 1. A comparison of physical properties of these colloidal drug delivery systems is given in Table 1. The first system, normal

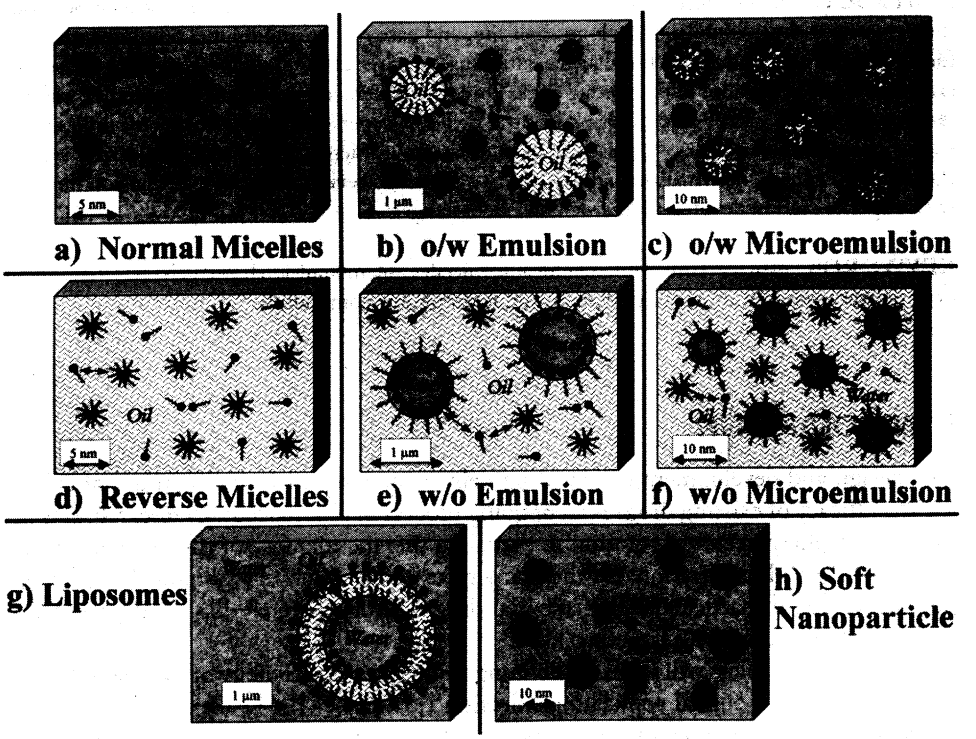

FIGURE 1. Various types of colloidal drug delivery systems.

TABLE 1
Physical Characteristics of Micelles, Microemulsion, Emulsions, and Liposomes

Delivery system	Advantages	Disadvantages
Micelles	Low viscosity Small droplet size Easy preparation Long shelf-life	Low solubilization Potential toxicity of surfactant
Microemulsions	High solubility of drug Small droplet size Easy preparation Long shelf-life	Large amount of surfactant Drug solubility influenced by environmental conditions Potential toxicity of surfactant
Emulsions	Small amount of surfactant High solubility of drug into carrier	High viscosity Instability Short shelf life Large droplets
Vesicles and liposomes	Made from lecithin and cholesterol also present in the body	High viscosity Difficult to prepare Often disintegrate once administered
Nanoparticles	Long storage life In vaccinations, slow degradation in body	Limited solubility of drug Difficult to prepare Difficult to control size Polymers which represent constituents are usually not bioacceptable

micelles (Figure 1a), are optically isotropic and thermodynamically stable liquid solutions consisting of water and amphiphile. Micelles do not tolerate large amounts of apolar solvents such as alkanes because of their very limited capacity to solubilize oil. With higher solubilization of oil, an emulsion will be formed in which oil droplets are dispersed in water containing oil-saturated micelles. Emulsions (Figure 1b) are metastable colloids with droplet sizes generally larger than 1 μm. Because of this large droplet size, emulsion drops will separate over a period of time and form a two-phase system (i.e., oil phase and aqueous phase). Emulsions are optically turbid dispersions and can only be obtained by mechanical mixing of the components because of their thermodynamic instability. However, because of the large amount of oil solubilized in an oil-in-water (o/w) emulsion, a large amount of hydrophobic drug can be dissolved in the dispersed phase. Furthermore, the amount of surfactant used in these systems can be as low as 1%.

Under certain conditions, the oil droplets in an emulsion can be made small enough that they do not refract light, hence forming a transparent dispersion. This dispersion is called a microemulsion (Figure 1c), which are transparent because of their small droplet size (generally < 100 nm). Microemulsions are thermodynamically stable, which implies that they

form spontaneously at certain concentrations of oil, water, and surfactant, and the formation is limited only by the diffusion of the molecules. It has been reported[2] that the change in free energy of dispersions shows a minimum at an equilibrium droplet size in the range of 100–1000 Å for microemulsion systems. Microemulsions require a relatively large amount of surfactant in order to stabilize the large interfacial area created by the nanodroplets. Microemulsions also often require the addition of cosurfactants such as alcohols, amides, and sulphoxides to attain an appropriate fluidity or viscosity of the interface. However, the choice of a surfactant system is very important in controlling the required surfactant concentration. A surfactant that strongly favors orientation at the oil–water interface will require a much lower concentration than will a surfactant that partitions strongly into either the bulk oil or bulk water phases. Like emulsions, oil-in-water (o/w) microemulsions are excellent solubilizing agents for hydrophobic drugs.

If water is added to an oil/surfactant solution instead of adding oil to water plus surfactants, reverse micelles (Figure 1d) can be formed. In reverse micelles, the polar groups of the surfactant molecules are oriented toward the hydrophilic interior, and the hydrophobic chains are extended in the oil phase. With higher solubilization of water beyond the solubility limit of reverse micelles, water-in-oil (w/o) emulsions (Figure 1d) are formed. Hence, w/o emulsions are excellent solubilization agents for hydrophilic drugs. Finally, as with o/w microemulsions, a water-in-oil (w/o) microemulsion (Figure 1e) can be formed under certain conditions. In addition, w/o microemulsions are also thermodynamically stable, are spontaneously formed, and are excellent solubilizing agents for hydrophilic drugs.

A special type of colloidal system can be generated when hydrophilic and hydrophobic properties of the surfactant are balanced. In this case, a lamellar liquid crystal is formed that is viscous and anisotropic.[3] Vesicles (Figure 1g) are spherical bilayers formed when a lamellar liquid crystal phase is shaken with water. It is important to emphasize that double-tailed surfactants such as lecithin and phospholipids produce vesicles in water or saline. Niosomes are a special class of vesicles made from nonionic surfactants.[4] Liposomes are vesicular structures consisting of hydrated bilayers.[5] Liposome structures used for pharmaceutical purposes consist of a phospholipid backbone, but other classes of molecules can form bilayer-based vesicular structures as well (for example, certain ionized cholesterol esters as cholesterol hemisuccinate).[6]

A final colloidal system to consider is composed of nanoparticles that are solid, colloidal particles consisting of macromolecular substances that vary in size from 10 nm to 1000 nm.[7] Nanoparticles of polymers can be used as drug delivery systems by dissolving, entrapping, adsorbing, attaching, or encapsulating the drug.[8] Nanoparticles, also called nanopellets, can have a shell-like wall called a microsphere or a polymer lattice.[9,10]

B. Molecular Aggregates of Surfactants

A general hypothetical phase diagram of the sort that can be expected in a system consisting of surfactant (+ cosurfactant) (S), oil (O), and water (W) systems is shown in Figure 2. The major feature of this phase diagram is that all the previously discussed classes of compartmentalized drug carrier systems, except for the special case of liposomes, can be represented on this one phase diagram of oil, surfactant, and water. Hence, each of these colloidal systems simply represents different concentrations of oil, water, and surfactant for a given system.

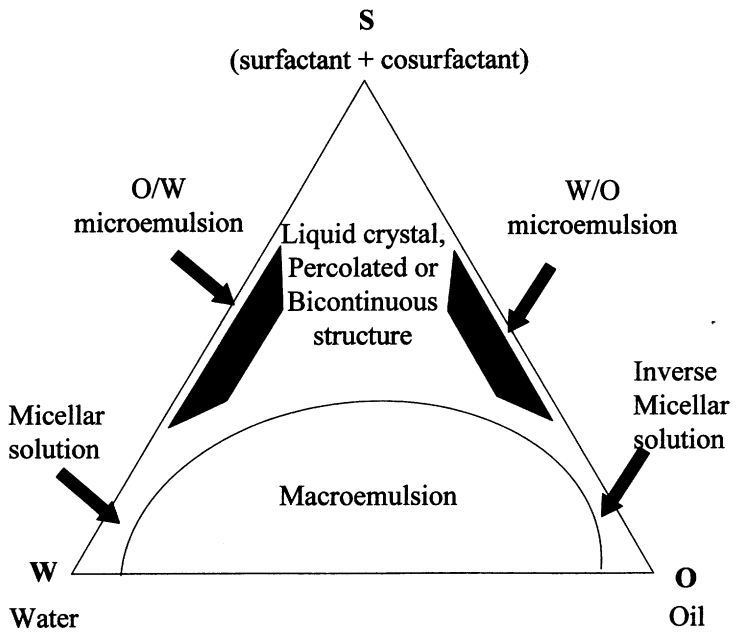

FIGURE 2. Hypothetical phase regions of microemulsion systems of oil (O), water (W), and surfactant + cosurfactant (S).

Surfactant molecules act like building blocks to make various molecular aggregates, such as those shown in Figure 1, by controlling parameters such as surfactant structure, concentration, electrolyte concentration, pH, pressure, and temperature. Thus, when a water-soluble surfactant is dissolved in water above a critical concentration, known as the critical micelle concentration (CMC), the surfactant molecules form spherical aggregates, with their hydrocarbon chains pointing toward the interior of the micelle and the polar group in contact with the bulk water phase. Such an arrangement removes the hydrocarbon chain from contact with water. Thus, normal micelles form from water-soluble surfactants, but other structures such as cylindrical micelles, lamellar micelles, and various liquid crystalline phases result from molecular association of such surfactant molecules, depending on the factors mentioned above. Similarly, some surfactants form reverse micelles when dissolved in organic solvents. In this case, the polar group is pointing inward, and hydrocarbon chains of surfactant molecules are in the organic solvent. For some surfactants, a critical amount of water is required to form the reverse micelles.[11] It should be also pointed out that the aggregation number, or number of molecules per micelle, is rather small (< 16) for reverse micelles, but much larger (65–300) for normal micelles.[12,13] Moreover, the critical micelle concentration is well defined for the normal micelles, whereas for the reverse micelles, micellization occurs over a broad range of surfactant concentrations.

The type of molecular aggregates that a surfactant may form depends on the molecular structure of the surfactant, the surfactant concentration, and physicochemical conditions such as temperature, pressure, pH, and electrolyte concentration. Israelachvili et al.[14] showed

that the type of structure that can be formed by a surfactant molecule depends on its packing parameter (v/al), which is defined as the ratio of volume of the hydrophobic chain (v) to the product of the head group area (a) and the chain length (l). The packing parameter for a particular environment will determine the curvature of the interface. A value of the packing parameter lower than unity facilitates the formation of structures in which the polar interface is curved *toward* the hydrocarbon phase—that is, structures of oil-in-water type. Conversely, a value larger than unity will give *reverse* curvature and favor water-in-oil structures.

C. Advantages and Disadvantages of Colloidal Drug Delivery Systems

The various colloidal systems applicable to drug delivery shown in Figure 1 have different advantages and disadvantages that should be considered for selecting a system for a given application. Table 2 lists some of the important advantages and disadvantages of drug delivery systems for pharmaceutical applications. Ideally, a colloidal delivery system should be designed to have low viscosity, small droplet size, simple preparation technique, long shelf life, low toxicity to the patient, high solubility of drug, controlled drug-release rate, slow degradation, and target specificity when administered.

The first system in Table 2, micelles, has low viscosity, small aggregate size, simple preparation, and long shelf life. However, the drug loading is very low in these systems.[15] Microemulsions have advantages similar to micelles, with the additional advantage of having a high drug-loading capability. The disadvantages of microemulsions stem from the use of a large concentration of surfactant and cosurfactant necessary for stabilizing the nanodroplets. The surfactants must be nontoxic for use in pharmaceutical applications. Furthermore, microemulsion stability is influenced by environmental parameters such as temperature and pH, and these parameters change upon microemulsion delivery to patients. Thus, the effect of such changes on microemulsion stability must be evaluated in the formulation development process.

TABLE 2
Advantages and Disadvantages of Different Drug Delivery Systems

	Micelles	Microemulsions	Emulsions	Liposomes
Spontaneously obtained	Yes	Yes	No	No
Thermodynamically stable	Yes	Yes	No	No
Turbidity	Transparent	Transparent	Turbid	Turbid
Size range	< 0.01 microns	< 0.1 microns	0.5–5 microns	0.025–25 microns
Cosurfactant used	No	Yes	No	No
Surfactant concentration	< 5%	> 10%	1–20%	0.5–20%
Dispersed phase concentration	< 1%	1–30%	1–30%	1–30 %

The next system, emulsions (or macroemulsions), offers the advantages of lower concentration of surfactant while still offering high drug loading. The drawback of this system is its high viscosity, larger droplet size, shorter shelf life, and instability. The next system, vesicles or liposomes, is significantly advantageous in drug delivery applications. The most commonly employed components are cholesterol and lecithin, which are biocompatible and hence eliminate toxicity concerns. It should be emphasized that cholesterol and phospholipids are similar to lipids found in tissues and cells. However, these systems are difficult to prepare, have high viscosity, and often disintegrate once administered.

The final system under consideration is the nanoparticle, which has a long storage life and slow degradation once administered because of the solid state of the particle. In general, nanoparticles are not influenced by environmental conditions, although they need to be stabilized in some form to prevent settling during storage. One disadvantage of nanoparticles is that they are difficult to prepare, and the actual drug preparation normally requires an additional step in order to adsorb or to attach the drug to the particle. Also, the prepared nanoparticles often show a wide size distribution, as opposed to microemulsions, which are monodispersed. Another disadvantage is the limited solubility of drug offered by adsorption on nanoparticles. Finally, the polymers generally used as nanoparticles in this application are usually not biocompatible, so toxicity is another concern.[16]

D. Microemulsions as Drug Carrier Systems

Some of the important properties of microemulsions are that they improve therapeutic efficacy of the drug and allow reduction in the volume of the drug delivery vehicle, thus minimizing toxic side effects.[17] The presence of surfactant raises the permeability of the cell membrane, which allows for easier absorption.[18] In some cases, the capacity of the microemulsion to solubilize large amounts of lipophilic and hydrophilic drugs at the same time can be advantageous as well.[19]

In addition to these advantages, microemulsions are expected to be easy to administer to children and adults who have difficulty swallowing solid dosage forms. They also offer several benefits for oral administration, including increased absorption, improved clinical potency, and decreased toxicity. Therefore, microemulsions have been reported to be ideal for oral delivery of drugs such as steroids, hormones, diuretics, and antibiotics.[20]

Some factors limit the use of microemulsions in pharmaceutical applications. The need for pharmaceutically acceptable ingredients limits the choice of microemulsion components (e.g., oil, surfactant, and cosurfactant), leading to difficulties in formulation. Furthermore, the concentration of surfactant and cosurfactant used must be kept low for toxicological reasons. For intravenous use, the demands on the toxicity of the formulations are rigorous, and very few studies have been reported so far.

E. Solubilization of Drug in Surfactant Aggregates

It is well recognized that the internal core of normal micelles forms an environment suitable for solubilization of nonpolar molecules.[21] Thus, solubilization of oil within micelles has been studied extensively. McBain and Hutchinson[22] reported extensive studies of the

moles of solubilized oil per mole of surfactant in micelles and showed that in most micellar solubilization, the ratio of mole of solubilized oil to mole of surfactant used is 1:10. Only in special cases can this solubilization ratio reach 1:1 (i.e., one molecule of oil solubilized per molecule of surfactant in the micelle). However, compared to loosely packed micelles, tightly packed micelles and cylindrical micelles exhibit greater solubilization capacity for oils.[23] Microemulsions, as stated previously, are a special class in which solubilization is much greater than that observed in most micellar solutions. For one surfactant molecule, it may be possible to dissolve 10–30 oil molecules (in o/w microemulsions) or 10–300 water molecules (in w/o microemulsions).[22] This is possible because microemulsion droplets consist of a core of oil or water having the same properties as bulk oil or bulk water. The drug dissolves or partitions in the oil and water domains in the microemulsions. Thus, the presence of bulk oil or water as dispersed phase in microemulsions allows substantially higher solubilization capacity than the conventional micellar solutions. This observation is also consistent with the fact that all microemulsion systems can become micellar solutions if the dispersed oil phase is decreased, but every micellar solution cannot necessarily grow into a microemulsion system if it does not meet the necessary conditions for the formation of microemulsions, as discussed in Section IIIA.

II. HISTORICAL DEVELOPMENT OF MICROEMULSIONS

A. Earlier Studies on Microemulsions

In 1943, Hoar and Schulman[24] first described water-in-oil microemulsions, which they referred to as transparent water-in-oil dispersions. They recognized the importance of very low interfacial tension in causing spontaneous emulsification of the added water in oil. Furthermore, they were able to calculate the radius of the water droplets in microemulsion using the formula

$$R = 3 \text{ (volume of water)} \prod \text{total area of water/oil interface}$$

Subsequently, these systems were investigated by Schulman and Riley[25] and J.H. Schulman and J. A. Friend.[26] Using low angle X-ray diffraction and light scattering, they confirmed the size of the droplets with the calculated size of the droplets in the microemulsion. In these investigations, they were able to confirm that the lamellar structure of surfactant is broken down by the penetration of oil and cosurfactant molecules, resulting in the fluidization of the film. The fluidization phenomenon was also emphasized by Schulman and McRoberts[27] from their studies on the structure of oil-in-water microemulsions having solubilized oils. Thus, from 1943 to 1948, Schulman and his coworkers established the pioneering concepts for the structure as well as the requirements for the formation of microemulsions. However, it was not until 1959 that the term *microemulsion* was used to describe such transparent oil-water-surfactant systems. Stoeckenious et al.[28] used electron microscopy to delineate the structure of microemulsions using negative staining. The existence of both water-in-oil and oil-in-water microemulsions was confirmed by the use of the electron microscopy. Schul-

man and his colleagues also emphasized the importance of transient negative interfacial tension as the driving force causing spontaneous emulsification.[29] In 1967, Prince[30] proposed a theory that identifies negative interfacial tension resulting from high surface pressure of the film as being responsible for the formation of microemulsion. The negative interfacial tension is due to depression of interfacial tension between the water and oil phase.

For air/water interface, the surface pressure of the film, π_f, is defined as

$$\pi_f = \gamma_o - \gamma_f \tag{1}$$

However, at the oil/water interface, one can write the surface pressure of the surfactant film, π_f as

$$\pi_f = (\gamma_{o/w})_o - (\gamma_{o/w})_f \tag{2}$$

where $(\gamma_{o/w})_o$ is interfacial tension of the clean oil/water interface (i.e., absence of surfactant film), and $(\gamma_{o/w})_f$ is interfacial tension at the oil/water interface in the presence of surfactant film.

Equation (2) can also be written as

$$(\gamma_{o/w})_f = (\gamma_{o/w})_o - \pi_f \tag{3}$$

Therefore, for the film that can generate very high π_f, [i.e., $\pi_f > (\gamma_{o/w})_o$], the $(\gamma_{o/w})_f$ becomes negative. This is a transient phenomenon, because the surfactant distribution among oil, water, and interface will ultimately yield very low positive interfacial tension (i.e., 10^{-3} to 10^{-5} mN/m).

The $(\gamma_{o/w})_f$ can be decreased to very low values in two ways. One is by increasing surface film pressure π_f by using mixed surfactant films (for example, SDS and cetyl alcohol), which are known to exhibit very high surface pressure (π_f). The other is by adding medium chain length alcohols to oil. The ideal way to achieve ultralow interfacial tension is by first decreasing $(\gamma_{o/w})_o$ by adding cosurfactants and then by adding surfactant to create high value of π_f. Therefore, in making microemulsions, oil-soluble surfactants/cosurfactants are added to the oil phase, and water-soluble ionic surfactants are added to the aqueous phase.

B. Other Research

Chan and Shah,[31,32] in studying enhanced oil recovery, proposed that as the ionic strength of the solution increases, the CMC decreases, the aggregation number of the micelles increases, and the solubilization of oil within normal micelles increases. This is because the repulsive forces between the micelles are reduced with increasing numbers of ions in solution by compression of the electrical double layer around micelles. At a particular ionic strength, termed *optimal salinity,* there are equal volumes of oil and brine solubilized, producing a balanced middle-phase microemulsion. Figure 3 shows the freeze-fracture electron micrograph of a middle phase microemulsion. It clearly shows discrete spherical structures embedded in a continuous aqueous phase. The system has been extensively studied by Reed and Healy.[33]

FIGURE 3. Freeze fracture scanning electron micrograph of a middle phase microemulsion. The spherical shapes are oil droplets suspended within the continuous aqueous phase. The black bar represents 0.5 μM.

It should be pointed out that other investigators[34-36] have proposed the possibility of bicontinuous structure or the coexistence of oil-external and water-external microemulsions in the middle phase. In very high surfactant concentration systems (15–20%), the existence of an anomalous microemulsion structure that contains neither conventional water-external nor oil-external phases has been proposed by Kotlarchyk[37] to account for some unusual properties of these systems. Not only is the transition to lamellae achieved by increasing the ionic strength, but it is also possible by changing the temperature if a nonionic surfactant is used.[40]

A characteristic feature of microemulsions prepared with nonionic surfactant is the sensitivity of their microstructure to temperature change. Each system is characterized by a narrow temperature range, the HLB temperature or phase inversion temperature (PIT) over which the hydrophilic–lipophilic properties of the surfactant are balanced for a given hydrocarbon–water system.[39] A temperature increase can cause a transition from o/w systems at

low temperature and high concentrations to bicontinuous microemulsions at or near the HLB temperature.[40] Further temperature increase beyond PIT favors the formation of w/o systems.[12,41] These possible changes are shown schematically in Figure 4.

C. Applications of Microemulsions

After the 1973 oil embargo, considerable research was carried out for enhanced oil recovery using microemulsions. In this application of microemulsions, the middle phase—or bicontinuous microemulsions—were of considerable interest in removing oil trapped in the porous media of oil reservoirs.[42]

From 1982 to the present, considerable work has been done in the area of producing nanoparticles of simple materials such as metals,[43,44] semiconductors,[45] and metal carbonates,[46] as well as various advanced materials such as superconductors,[47,48] magnetic materials,[49] photographic materials,[50] varistors,[51] nanocomposites,[52] and latexes[53] using microemulsions as nanoreactors. In this case, two water-in-oil microemulsions containing water-

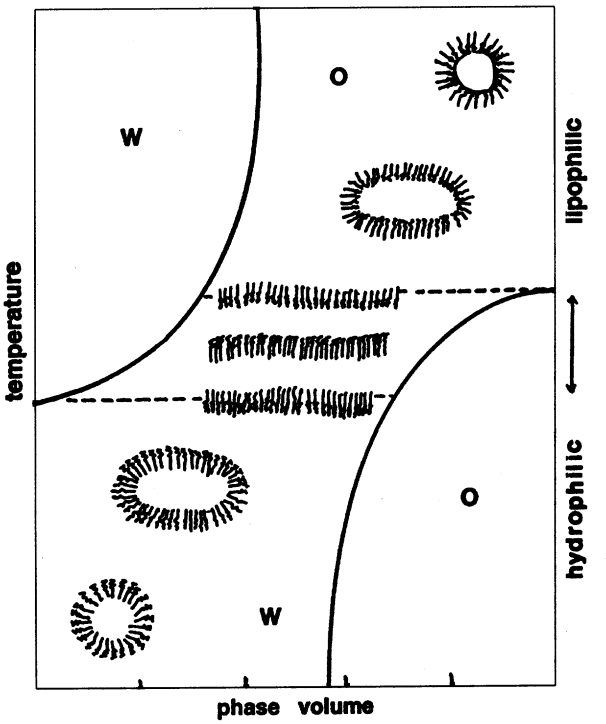

FIGURE 4. Schematic representation of transition from o/w systems at low temperature and high water content to bicontinuous microemulsions close to HLB temperature to w/o systems at higher temperatures. (Reprinted from Attwood, D., Colloidal drug delivery systems, 1994, p. 36,[60] by courtesy of Marcel Dekker, Inc., New York, NY)

soluble salts are mixed together or oil-soluble reactant is directly added to the water-soluble reactant in the aqueous core of the microemulsion. The collisions between droplets cause precipitation of water- insoluble compounds as nanoparticles or nanocrystals.[54] Further details on various types of nanoparticles prepared can be obtained from various reviews.[54,55] In addition, oil-in-water microemulsions are of considerable interest in areas such as lubrication and metal working industries, pesticide formulations, and agricultural sprays.[56,57]

The use of microemulsions in pharmaceutical technology is a relatively recent development.[58] Various possible advantages of microemulsions as carriers of drugs have been described by Gasco.[58] Considerable research has been devoted in the last decade to exploring microemulsions in relation to drug storage, stability, low dosage level, viability, side effects, controlled release, biological response, and homogenous distribution for possible use as drug delivery systems.

III. MICROEMULSION FORMULATIONS: METHODS TO PRODUCE AND CHARACTERIZE MICROEMULSIONS

A. Conditions Necessary to Produce Microemulsions

Microemulsions are fascinating systems in that nature prefers to have a dispersed system of oil, water, and surfactant having large total interfacial area, rather than separate phases of oil and water with much smaller interfacial area. In order to form microemulsions, three major factors must be considered.[59]

First, emulsifiers or surfactants must be carefully chosen so that an ultra-low interfacial tension (< 10^{-3} mN/m) can be attained at the oil/water interface. The ultra-low interfacial tension at the oil/water interface is a prime requirement to produce microemulsions. It is this very low interfacial tension that leads to spontaneous emulsification of oil in water or water in oil.

The second requirement is that the concentration of emulsifiers or surfactants must be high enough to provide the number of surfactant molecules needed to stabilize the microdroplets produced by an ultra-low interfacial tension. Because microemulsions are in the range of 100–1000 Å in diameter, 30% of oil dispersed in water with 200 Å droplet diameter will create 10^6 cm^2 of total interfacial area per milliliter of microemulsion; therefore, the large concentration (10–40%) of surfactant is required to stabilize the newly created interface of microemulsion droplets. However, it is precisely the fine tuning and the right choice of structure of the surfactant and cosurfactant that can reduce the concentration of surfactant required for microemulsion formation. The emulsifier partitions into three compartments: water, oil, and the interface between the water and oil. By proper adjustment of hydrophilic and hydrophobic groups of the surfactant, the surfactant molecules can preferentially partition into the interface and minimize their concentration in the bulk oil and water phases. Therefore, the understanding of the partitioning behavior of surfactant in water, oil, and the interface is of considerable importance for the formulation of microemulsions.

The third major consideration in formulating microemulsions is the flexibility or fluidity of the interface to promote the formation of microemulsions. Therefore, short-chain alcohols (C_4 to C_7) are often added as cosurfactant in surfactant + water + oil systems to

produce microemulsions.[60] The penetration of short-chain alcohols into the interfacial film produces a more fluid interface by allowing the long hydrophobic tails of the C_{16} or C_{18} surfactants to move freely at the interface. In summary, the three important conditions for producing microemulsions are:

1. ultra-low interfacial tension at the oil/water interface (i.e., < 10^{-3} mN/m)
2. sufficiently high concentration (10–40%) of surfactant to cover the newly created surface within the microemulsion
3. sufficiently low fluidity and low surface viscosity of the interfacial film to spontaneously form microdroplets with small radius of curvature (50–500 Å)

B. Phase Equilibria of Microemulsion Systems

Phase diagrams of the type shown in Figure 2 are useful in formulation studies as a means of delineating the area of existence of the microemulsion region. The method used to construct such diagrams depends on the mutual solubilities of the components, but in general it is convenient to use the titration method, which allows a large number of compositions to be examined relatively quickly. The titration method consists of weighing quantities of surfactant, cosurfactant, and oil, which are then mixed to form a monophasic solution. The constantly-stirred mixture is then titrated with water at constant temperature. After each addition of water, the container should be stoppered to minimize loss of volatile component and the system examined for clarity, birefringence, flow properties, and stability. After coarse determination of the microemulsion region, a more detailed study of this region of the phase diagram is required to assess the long-term stability of the systems in this region.

An alternative to the titration method is to take surfactant, oil, and water and mix them together to form an emulsion, which is then titrated with the cosurfactant until the mixture becomes clear to locate the microemulsion region.[61]

From Figure 2, we can see that at high oil concentration, the surfactant forms reverse micelles capable of solubilizing water molecules in their hydrophilic interior. Continued addition of water in this system may result in the formation of a w/o microemulsion in which water exists as droplets surrounded and stabilized by the interfacial layer of the surfactant/cosurfactant mixture. At a limiting water content, the isotropic clear region changes to a turbid, birefringent one. Upon further dilution with water, a liquid crystalline region may be formed in which the water is sandwiched between surfactant double layers. Finally, as the amount of water increases, this lamellar structure will break down and the water will form a continuous phase containing droplets of oil stabilized by a surfactant/cosurfactant interfacial film.

C. Dynamic Behavior of Microemulsions

Microemulsions are dynamic, self-organizing solutions in which aggregation/disintegration processes operate simultaneously. In this process, dynamic exchange of matter between dispersed phases occurs continuously, resulting in an overall equilibrium. The dynamic process comprises: (1) the exchange of water between bound and free state; (2) the exchange of counterions between ionic head groups of the surfactant and core water; (3) the exchange of co-

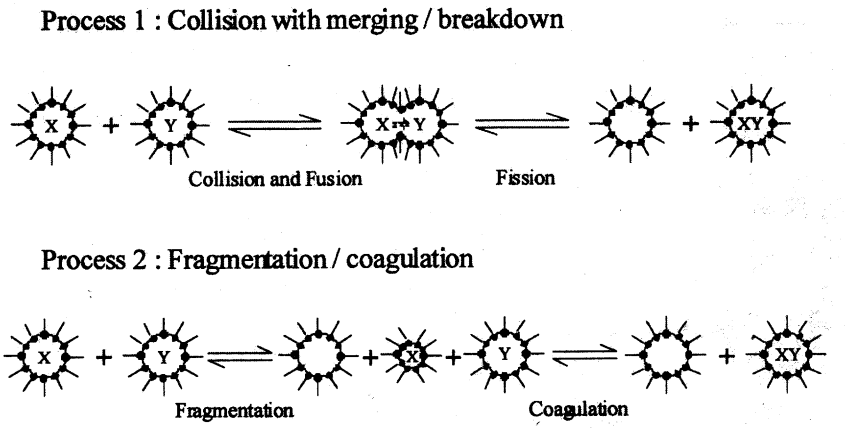

FIGURE 5. Dynamic behavior of microemulsion.

surfactant between the interfacial film, the continuous phase and the dispersed phase (if soluble in this phase); and (4) the exchange of surfactant between the interfacial film and the aqueous phase.[11] Microemulsions are thermodynamically stable, but there are continuous exchanges of components between droplets by two types of processes:

1. droplet collisions accompanied by temporary merging of the droplets into a larger droplet (fusion) followed by breakdown of this larger droplet (fission)
2. partial breakdown (or fragmentation) of droplets with loss of droplet fragments, which can later associate with other droplets (coagulation)

These two processes are represented in Figure 5. For a w/o microemulsion the first process has been found to hold good, while the second process of coagulation/fragmentation has been found to be good for o/w microemulsion.[62,63] The collision between aggregates is governed by diffusion. In the absence of interaction, the rate constant for intermicellar collisions (k_c), is the upper limit for solute exchange process. At room temperature in a solvent of low viscosity, typical values of k_c are of the order of 10^9 $dm^3.mol^{-1}.s^{-1}$. Following collision, the opening of a water channel (fusion) required for the transfer of a solute involves the formation of a transient interfacial positive curvature in the region of contact of colliding droplets. The opening of the surfactant layer has shown to be the rate-controlling step.[64] The facility of the process in a given system is characterized by the intermicellar exchange rate constant (k_{ex}). If every collision leads to coalescence, the exchange rate coefficient (k_{ex}) should be of the order of (k_c). In most microemulsions this is not the case, and the ratio k_{ex}/k_c thus reflects the fraction of collisions that results in matter exchange.[64,65] The intermicellar exchange process is also dependent on attractive interactions between the surfactant tails upon the approach of two droplets.

For a given formulation (nature of oil and aqueous phase), the nature of surfactant molecules determines the exchange rates through the elasticity (or "rigidity") of the interface. For rigid droplet-type structures (such as those found in AOT surfactant) the k_{ex} can be four orders of magnitude lower than k_c, which means that only one out of 10^4 collisions lead

to merging and matter exchange. On the other hand, reverse micelles formulated with nonionic surfactants exhibit fluid interfaces and exchange much faster. Table 3 shows the typical values of exchange rate coefficients (k_{ex}) of selected w/o microemulsion systems. By considering a typical droplet concentration of 10^{-3} mol.dm^3, the results of Table 3 indicate that time between interdroplet exchanges (estimated as $1/k_{ex}$ [drops]) is in the range of 1 microsecond to 20 milliseconds. As can be seen from the table, the exchange rate depends not only on the nature of surfactant but also on specific formulation and temperature.

TABLE 3
Intermicellar Exchange Rate of Different Microemulsion Systems

Microemulsion	Method	k_{ex} (dm^3.mol.$^{-1}$.s^{-1})	Reference
Water/$C_{12}EO_5$/heptane	TRFQ		254
T = 21 C, RC = 8		8×10^8	
T = 40 C, R = 8		3×10^8	
T = 40 C,R = 16		9×10^8	
Water/TX-100[d]/toluene	TRFQ		199
T = 25, R = 7.7		2×10^8	
R = 13		6×10^8	
R = 15		9.5×10^8	
Water/DTACe/hexanol/heptane	FQ		65
T = 25, R = 10	(CFMIO)[f]	4.6×10^6	
R = 20		9.2×10^6	
R = 25		1.1×10^7	
Water/CTABg/hexanol/dodecane	TRFQ		252
T = 25, R = 20		1.4×10^8	
Hexane in place of dodecane		1.1×10^7	
Water/AOT0/heptane	TRFQ		253
T = 25,R = 11		1.4×10^7	
R = 22		1×10^7	
R = 11, + additives:			
[hexanol]/[AOT] = 1.5		7.5×10^6	
[benzyl alcohol]/[AOT] = 1.5		3.3×10^8	
40 v/v% toluene in oil phase		1.6×10^6	
Dodecane in place of heptane		5.0×10^7	

[a] pentaoxyethylene dodecyl ether
[b] time-resolved fluorescence quenching
[c] water to surfactant molar ratio
[d] polyoxyethylene (9.5) octyl phenyl ether
[e] dodecyl trimethyl ammonium chloride
[f] fluorescence quenching in continuous flow method with integratin observation
[g] cetyl trimethyl ammonium bromide
[h] sodium bis (2-ethyl hexyl) sulfosuccinate

D. Modeling of Microemulsion Systems

Experimental studies of surfactant solutions reported in the literature are numerous. Many experimentally observed features of surfactant self-assembly are now well understood qualitatively. For example, Mukerjee[66-68] showed how cooperativity of self-association is responsible for the formation of large aggregates. The physicochemical features of surfactant self-assembly were elucidated by the pioneering work of Tanford,[69,70] who demonstrated that the hydrophobic effect is responsible for cooperative self-assembly, while the interactions between the polar head groups of surfactants provide the anticooperativity that keeps these aggregates constrained to a finite size.

A great deal of work has also been done with theoretical and computer modeling of self-assembly of surfactants and microemulsions using statistical and molecular thermodynamic approaches. Some of the work in this area is highlighted below.

1. *Reiss's Entropy Theory of Spontaneous Emulsification of a Two-Phase Fluid System*

Reiss[71] used a statistical thermodynamic approach to show that emulsification is an entropy-driven phenomenon under conditions where interfacial energy is positive and small. The assumptions used for this approach are:

1. The total free energy of the system (ΔG) is the sum of the free energy before mixing ($\Delta G'$) and the free energy of mixing ($\Delta G''$).

$$\Delta G = \Delta G' + \Delta G'' \tag{4}$$

$$\text{where } \Delta G' = \Sigma N_i 4\pi r_i^2 \sigma \tag{5}$$

   $\Delta G''$ should be sufficiently negative that the net free energy change ΔG will also be negative, leading to a dispersed equilibrium state.
2. The spherical droplet is fixed to a laboratory frame of reference, and the dividing surface used to define it is similarly fixed.
3. When dispersion occurs, clusters of only one size are produced.

The internal partition functions (motions relative to the center of mass) of the clusters were estimated by referring them to free energies of the laboratory drop of the same size. The spherical clusters were assumed to interact with the center of mass, and this interaction was further assumed to be that of equivalent hard spheres.

The final expressions describing the change in free energy of the system are

$$\Delta G' = 4\,\pi\, r^2\, \sigma\, N_D \tag{6}$$

$$\Delta G'' = -kTN_D\left[\ln\left\{\frac{Vn^{1/2}}{v_2}\left(\frac{12}{\pi}\right)^{3/2}(1-y)\exp\left[\frac{3y(y-2)}{2y(1-y)^2}\right]\right\} - \ln N_D + 1\right] \tag{7}$$

and

$$\Delta G = 4\pi r^2 \sigma N_D - kTN_D\left[\ln\left\{\frac{Vn^{1/2}}{v_2}\left(\frac{12}{\pi}\right)^{3/2}(1-y)\exp\left[\frac{3y(y-2)}{2y(1-y)^2}\right]\right\} - \ln N_D + 1\right] \quad (8)$$

where k is the Boltzmann constant; T is temperature; $y = 4\pi r^3 C/3$, $C = N_D/V$; σ is the interfacial tension; N_D is the number of clusters with radius r, each containing n molecules; V is the total volume of the system; and v_2 is the volume per molecule in bulk liquid phase 2 (the phase assumed to contain the interface).

For spontaneous dispersion to occur, it is necessary that $\Delta G = 0$.

These calculations show that for interfacial tension below about 2 dynes/cm, spontaneous dispersions can occur, although with large tensions the dispersed particles are not macroscopic. However, for interfacial tensions below 5×10^{-4} dynes/cm, macroscopic particles are formed.

The major drawback of this model is that it becomes inadequate when the surface tension becomes small. When this occurs, the thickness of the interfacial region will increase to a point where it becomes comparable to the dimension of the cluster.

2. Nagarajan–Ruckenstein Predictive Molecular Thermodynamic Approach

Nagarajan and Ruckenstein[72] developed a thermodynamic treatment of surfactant self-assembly in aqueous media that allows *a priori* quantitative prediction of the aggregation behavior of surfactants, starting from their molecular structure and solution conditions. The treatment combines the general thermodynamic principles of self-assembly with detailed molecular models for various contributions to the free energy of aggregation. The calculations show that as a result of solubilization, microemulsion structures are formed with a single surfactant. The core of the solubilizate in these structures is rather small. In developing a molecular model for the free energy of aggregation, they considered the contribution arising from (1) the transfer of surfactant tail from water to the hydrophobic core of the aggregate (assumed to behave like a liquid hydrocarbon); (2) the conformational free energy of the tails inside the aggregates resulting from the constrained location of head group on the aggregate surface; (3) the free energy of formation of the aggregate–water interface; and (4) the interaction between head groups of the surfactants at the aggregate surface. The thermodynamic treatment developed here is strictly predictive in the sense that it is free of information extracted from the aggregation behavior of surfactants.

3. Overbeek's Thermodynamic Model of Droplet Size Distribution and Phase Equilibria with Ionic Microemulsions

Overbeek[73] formulated the expression for free energy of droplet-type microemulsions containing ionic surfactants and nonionic cosurfactants on the basis of curvature-dependent interfacial free energy, the pressure difference between the droplets and the continuous medium, and the entropy of mixing between spherical droplets with the medium. Using this approach,

theoretical estimates on the droplet size distribution and its influence on phase equilibria can be obtained for a Winsor-type system in which the microemulsion exists in equilibria with excess water or oil. The equilibrium among droplets of various sizes was dealt via a mass-action approach. This thermodynamic analysis gave a good qualitative description of droplet sizes, size distribution, and interfacial tensions in Winsor-type equilibria in their dependence on the amounts of water, oil, and ionic surfactant and on the concentrations of salts and of cosurfactant. The free energy of curved electrical double layers forms the basis for understanding the influence of added electrolytes on the curvature dependence of the interfacial tension and on the phase behavior. Note that the droplet size distribution is essential for understanding the existence of a finite range of salt concentrations (and cosurfactant concentrations), where a microemulsion may be in equilibrium with both water and oil (Winsor III).

E. Solubilization of Drug Molecules in Microemulsions

The size and region of existence of a single-phase microemulsion domain within the phase diagram are strongly influenced by the presence of salts in the aqueous phase, the nature of the polar group and hydrocarbon group of the surfactant, the solvent, and the temperature. In general, increasing the ionic strength of the aqueous phase reduces electrostatic interactions among the surfactant polar groups, which results in more rigid interfaces, lower aggregation numbers, lower intermicellar attractions, and in most cases reduction of maximum solubilization capacity for the aqueous phase at a given set of conditions.[74] It has been concluded that solubilization and phase equilibria of w/o microemulsions are dependent on two phenomenological parameters: spontaneous curvature and elasticity of the interfacial film. Maximum solubilization can be obtained by minimizing both the interfacial bending stress of rigid interfaces and attractive interdroplet interaction of fluid interfaces at an optimal interfacial curvature and elasticity. Bansal et al.[75] studied the solubilization capacity of w/o microemulsions formed with fatty acid soaps and alcohols as a function of alkyl chain length of oil, soap, and alcohol and found that maximum solubilization was observed when chain length compatibility was reached. Recently, various articles have reported an increase in solubilization capacity of reverse micelles in the presence of mixed surfactant. The increase in solubilization was interpreted in terms of stability of the interfacial film of the reverse micellar droplet and size of the microdroplet.

F. Methods of Characterizing Microemulsions

The elucidation of the internal structure of a microemulsion can be very complex, and sophisticated physical techniques are required for this purpose. Small-angle X-ray scattering (SAXS), small angle neutron scattering (SANS), dynamic (or laser) light scattering (DLS), transmission electron microscopy (TEM), nuclear magnetic resonance (NMR), and time-resolved fluorescence quenching (TRFQ) methods have been increasingly used over the last 2 decades for this purpose. These methods can derive multiple data on microemulsion structure, and their application will be elaborated on in detail compared to other methods. Other methods—conductance, viscosity, ultrasonic interferometry, ultrasonic absorption, dielec-

tric permittivity, thermal conductivity, transient electric birefringence, infrared spectroscopy, calorimetry, etc.—are also frequently used for understanding the physicochemical states of microemulsions. Investigations worth noting in this direction are Friberg et al.,[76] Scriven,[77] Cazabat and Langevin,[78] Brunetti et al.,[79] Zana and Lang,[80] Fang and Venable,[81] and Hou et al.[82] For additional highlighting information, articles by Bellocq et al.,[83] Hansen,[84] Nilsson and Lindman,[85] Chatenay et al.,[86] Auvray et al.,[87] Bisal et al.,[88] Calje et al.,[89] Tabony et al.,[90] Lindman and Wennerstrom,[91] and Gradzielski and Hoffmann[92] may be consulted for specialized techniques.

1. Transmission Electron Microscopy (TEM)

Transmission electron microscopy (TEM) has been applied in limited studies but has potential use in understanding the microstructure of microemulsions under varied conditions of dispersant composition and concentration.[93–96] In principle, it is possible to measure droplets of the size range of most microemulsions using TEM. Several researchers[94–96] have reported results obtained using freeze-fracture electron microscopy (FFEM). In this technique, the microemulsion must be frozen rapidly enough to avoid phase separation or crystallization. The objective can be achieved by plunging the specimen into a liquid cryogen, by propane jetting, or by spray freezing. The sample is subsequently fractured, and its visibility is enhanced by depositing a platinum carbon layer *in vacuo*. The microemulsion sample, mounted on a support film or grid, is replicated by breaking apart the film, and the replica is then viewed by TEM and assumed to be representative of the bulk microemulsion. Recent developments in cryofixation have overcome many of the problems associated with artifact formation in earlier studies,[97–99] which were caused by slow cooling rates. Jahn and Strey[94] and Vinson et al.[95] have developed a mechanical plunging device for rapid freezing and have given detailed experimental procedures. Recent freeze-fracture TEM studies using rapid cooling rates and adequate environmental control of the samples before freezing (to prevent loss of volatile components) have investigated microemulsions of pentaethylene glycol dodecyl ether ($C_{12}E_5$), water, and n-octane.[94,95,100]

A complimentary technique is direct imaging, in which thin portions of the specimen are directly investigated in the frozen hydrated state by using a cryostage in the TEM. The development of glass-forming microemulsions that do not break down during cooling and in which neither the dispersed nor the matrix phase crystallizes during the cooling process has provided a way for direct studies of the microemulsion structures. The first type of such systems to be reported were o/w microemulsions with a noncrystallizing aqueous matrix obtained by adding propane–1,2–diol (propylene glycol) to the water in the ratio 1:3.[101,102] It was thought that propylene glycol functioned as cosurfactant because the usual addition of alcohol was not necessary. The fact that neither phase crystallized on cooling below 273 K meant that possible modifications of microemulsion structure by crystal growth were avoided. This fact was extended to w/o microemulsion by Green,[103] who used glycerol/water mixtures as the dispersed phase, a glass-forming oil (ethylcyclohexane) as the continuous phase, and didodecyldimethyl ammonium bromide as surfactant. Direct electron microscope imaging by vitrified microemulsion using freeze-fracture techniques revealed clearly separate droplets of 20 nm diameter.

2. Scattering Methods

Scattering methods that have been employed in the study of microemulsions include small-angle X-ray scattering (SAXS), small-angle neutron scattering (SANS), and DLS. SAXS and SANS have a lower size limit of 2 nm and an upper size limit of 100 nm, while the upper limit is a few microns for light scattering. These techniques are therefore ideally suited for study of microemulsions. In the case of monodisperse spheres interacting through hard sphere repulsion, the general expression for scattering intensity $I(Q)$ is defined as

$$I(Q) = n\,P(Q)S(Q) \tag{9}$$

where n is the number density of spheres and Q is the scattering vector ($Q = 4\pi\sin\theta/\lambda$ with θ = scattering angle and λ = wavelength). The form factor $P(Q)$ expresses the scattering cross-section of the particle, and the structure factor $S(Q)$ allows for interparticle interference. Analytical expressions may be used to calculate both $P(Q)$ and $S(Q)$ under favorable circumstances.

3. Small-Angle X-ray Scattering

SAXS techniques have been used by several researchers to gain information on droplet size in dilute microemulsion systems. The microstructure of the n-dodecyltetraoxyethylene glycol monoether ($C_{12}E_4$)/water/hexadecane system was studied by Shimobouji et al.[104] using SAXS. On the basis of analysis of surfactant concentration dependence of the peak position, it was concluded that an o/w microemulsion had been formed, with oil droplets being identified by a single broad peak on the scattering curves. The radii of the droplets were estimated to be between 3.5 and 140 nm depending on surfactant concentration. Bohlen,[105] using SAXS, reported a decrease in droplet size in a poly (oxyethylene) alkyl ether/ n-alkane/ water microemulsion from 94 to 22 nm as the molecular size of nonionic surfactant decreased in the order $C_{12}E_5 > C_{10}E_4 > C_8E_3$. The droplet size was also found to decrease with increasing surfactant concentration.

Interpretation of the scattering curves from concentrated microemulsions presents a problem. Zemb et al.[106] and Barnes et al.[107] interpreted the SAXS scattering from a w/o system composed of dodecyl dimethyl ammonium bromide and water with various alkanes as the oil using the disordered open connected model. This model successfully described the structural transitions that occurred in the system as the water content was increased. At low water content the structure resembled a disordered hexagonal phase, which at higher water content changed to isolated water spheres in oil. The approximate regions of the phase boundaries could be determined using the model.

The recent use of synchrotron radiation sources has improved small-angle X-ray methodology, resulting in increased performance of SAXS.[108,109] By this means, the sample-to-detector distances may be up to 4 m, as compared to 30–50 cm with laboratory-based X-ray sources. These large sample-to-detector distances result in less diffuse profiles, allowing better interpretation of the curves. Another advantage of synchrotron radiation is that it can be used even when the amphiphiles in the microemulsion are poor X-ray scatterers, allowing a wider range of microemulsion to be examined. North et al.[108] used synchrotron SAXS in a study of the 3-component w/o microemulsion system AOT, water, and dodecane and

found that analysis of the low Q value data gave more reliable results than analysis of high Q value data, which were affected by polydispersity. All microemulsions were chosen close to a critical phase transition at 25 °C. A significant change in the small-angle scattering profile at low Q values was observed in all samples due to critical scattering on approaching the phase transition within the microemulsion. Hilfiker et al.[109] used synchrotron SAXS to look at spherical deformity in the AOT, water, and n-hexane w/o microemulsion system. The spheres were thought to deform into prolate and oblate ellipsoids, the deformity being more likely when the temperature was increased.

4. Small-Angle Neutron Scattering

Small-angle neutron scattering has proved a valuable technique for the study of microemulsions, although it suffers the disadvantage of requiring a central facility. Howe et al.[110] and Robinson et al.[111] reported an extensive investigation of the AOT, water, and alkane microemulsion system using this technique. These workers achieved good SANS contrast by using the fact that hydrogen and deuterium have a different sign in their neutron scattering length (−3.74 and 6.67 fm, respectively). The negative scattering for hydrogen indicates a phase shift of 180° between scatter by hydrogen and deuterium. Contrast was achieved between the water and oil phases by selective H/D isotopic substitution. These workers found an approximately linear relationship between the size of the water core radius and the concentration ratio (D_2O/AOT) with heptane as the oil.[112] Cebula et al.[113] used SANS in combination with light scattering to investigate the water, xylene, sodium dodecyl benzene sulphonate, and hexanol system. The information from this experimental technique was consistent. For the system investigated, the radius of the water core of the droplet was found to increase with increase in water volume fraction while maintaining a constant water/oil interfacial area. In further studies, Cebula et al.[114,115] reported SANS and light scattering measurements on microemulsions formed with water, potassium oleate, hexanol, and dodecane with a water/potassium oleate volume fraction of 1.44. The results from both techniques indicated little variation in droplet diameters within a wide region of the microemulsion domain.

There are a number of important differences between SANS and SAXS techniques. Neutrons are absorbed less readily than are X-rays; therefore, the sample is more stable in neutron beams than in X-ray beams. The source-to-detector distances used in SANS are much larger (10–30 nm)[116] than are those of SAXS (4 m).[109] A powerful characteristic of neutron scattering is the possibility to selectively enhance the scattering power of various parts of the microemulsion using protonated or deuterated molecules, while in X-ray scattering materials must be selected as good X-ray scatterers. The X-ray contrast between oil and water is small, while selective deuteration improves contrast (signal-to-background ratio) in the SANS method. The rate of data collection is faster with SANS because of the longer wavelengths used (1 nm compared with 0.1 nm for X-rays) and larger sample size. SANS profiles suffer from incoherent scattering at high Q values, making interpretation of small signals difficult, whereas with SAXS there is little background scattering at high Q values, and, consequently, the effective Q range for SAXS experiments extends to higher values. In view of the limitations of the two techniques, it is preferable to use SANS or SAXS in combination with other techniques—that is, complementary use of several techniques can better help reveal microemulsion structure.

5. Dynamic Light Scattering

Dynamic light scattering analyzes the fluctuations in scattering intensity that occur over very short time intervals resulting from the Brownian motion of the particles. The diffusion coefficient of the scattering centers may be calculated from decay of the correlation function, and in the absence of interparticle interference, the hydrodynamic radius of the particle r_H can be determined from the diffusion coefficient D using the Stokes–Einstein relationship

$$D = k_B T/6\pi\eta\, r_H \tag{10}$$

where k_B is the Boltzmann constant, T is absolute temperature, and η is viscosity of the solvent. With most microemulsions, extrapolation to infinite dilution is not possible, so errors caused by interparticle interference will affect r_H for these systems and should be corrected.[97]

Cebula et al.[113] used DLS to confirm their light scattering and SANS results in the system water, xylene, sodium dodecyl benzene sulphonate, and hexanol, as discussed above. The stability of the microemulsion was demonstrated by lack of change in correlation functions after the sample was subjected to ultrasonic radiation. Bedwell and Gulari[117] reported that addition of small amounts of electrolyte to microemulsions of AOT, water, and heptane resulted in an increase in diffusion coefficient D, which they interpreted as a decrease in the hydrodynamic radius of the particles. In a further examination of this system,[118] these researchers reported an increase in droplet radius from 6 to 300, nm with increase in temperature and dispersed phase volume. Chang and Kaler[119] used DLS in an examination of the 5-component w/o system consisting of sodium 4-(1-heptylnonyl) benzene sulphonate, isobutyl alcohol, water, dodecane, and sodium chloride. Droplet sizes increased from 6 to 30 nm, with increasing volume fraction of water, up to a limited value. From 25% to 75% water, the systems were turbid, and D values were found to be constant, indicating the presence of a bicontinuous structure. This was confirmed by the presence of light scattering studies on the same system. Rosano et al.[120] investigated the influence of the nature of surfactant, cosurfactant, and oil on microemulsion formation using DLS and transmittance data. Increasing the amount of surfactant (potassium soaps of straight chain fatty acids) decreased the particle size. They reported an increase in size with increasing amounts of alcohol for the droplets in o/w and w/o microemulsions. They attributed this in the o/w case to the excess alcohol's dissolving in the oil droplets causing their swelling. In the w/o system, the excess alcohol was thought to extract surfactant from the interface, resulting in decreased stabilization of droplets and hence increased size.

Muller and Muller[121,122] reported investigations on w/o microemulsions prepared with potassium oleate (as surfactant), liquid paraffin (as oil), and various alcohols (as cosurfactants). DLS was used to determine the effect of the alcohols on microemulsion stability by sizing the systems at regular time intervals, an increase in particle size indicating a deviation from the stable system. These researchers reported an almost constant droplet size in the middle of the microemulsion region over a prolonged period of time. Zulauf and Eicke[123] studied the AOT, water, and isooctane system using DLS and presented evidence for a clear distinction between inverted micelles and microemulsion regions according to the degree of hydration or amount of solubilized water, respectively.

Cazabat and Langevin[78] reported a DLS study on a w/o microemulsion composed of sodium dodecyl sulphate and water with cyclohexane or toluene as oil phase and butanol,

pentanol, or hexanol as cosurfactant. No attempts were made to interpret the diffusion data at large water volume fractions, but a theoretical treatment was proposed for the variation in diffusion coefficient with water content at small volume fractions. This treatment assumed that the microemulsions behaved as solutions of spherical particles interacting with a potential that was the sum of a hard sphere repulsion force VHS and a small attractive force VA. A recent DLS study by Trotta et al.[124,125] investigated the influence of increasing amounts of alcohol on the diffusion coefficients of microemulsions droplets. The o/w system studied was AOT, isopropyl myristate, and water with 10 alcohols of varying water solubilities, ranging from butanol to pentane-1-ol. All experiments, performed by adding increasing amounts of cosurfactant to the system, showed the presence of maximum diffusion coefficient corresponding to an optimum amount of cosurfactant for microemulsion formation.

IV. DRUG DELIVERY WITH MICROEMULSION FORMULATIONS

A. Introduction to Pharmaceutical Microemulsions

The two main processes that govern the release of drugs from compartmentalized systems are transfer of drug from the dispersed phase to the continuous phase, and diffusion of the drug through a membrane or interface from the continuous phase to the sink solution.[126,127] In properly selecting a suitable microemulsion system for drug solubilization and delivery, it is first necessary to have a general understanding of the behavior of the drug, both in the delivery system and *in vivo.* Of particular importance are various parameters such as aqueous and oil solubility of drug, partition coefficient data, droplet size, membrane permeability data across body tissues encountered during delivery,[126] and rate of diffusion in both phases.[127,128] Because of the high stability of microemulsions, it is often difficult to measure the concentration of drug in both phases directly.[129]

Also needed when choosing a microemulsion formulation for drug delivery is a detailed understanding of the components making up the microemulsion system. The surfactant, cosurfactant (if necessary), and oil best suited to the desired performance characteristics of the drug formulation must be chosen from a wide variety of possibilities. For example, the type of oil used in the microemulsion formulation is an important consideration not only in determination of phase structures and stability, but also in the level of drug solubilization.[130]

As previously mentioned, the major advantages of microemulsion drug delivery systems include:

- ease of preparation
- clarity
- stability
- ability to be filtered
- ability to encapsulate drugs of different HLB in the same system because the microemulsion can solubilize oil-soluble, water-soluble, and interface-soluble drugs
- low viscosity

Because of these unique physical properties, microemulsions have attracted a great deal of interest in recent years as drug delivery vehicles.[19,131-134]

Specific advantages exist for different types of microemulsion systems. For example, water-in-oil (w/o) microemulsions offer protection of water-soluble drugs and sustained release (with concomitant increased bioavailability) of water-soluble material. Similarly, oil-in-water (o/w) microemulsions present increased solubility, sustained release, and bioavailability of oil-soluble material. Middle-phase (or bicontinuous) microemulsion systems provide a concentrated formulation of both oil-soluble and water-soluble drugs. When tailoring a specific microemulsion system for drug delivery, the following requirements must be followed for pharmaceutical microemulsion formulations:

1. All components of the microemulsion system *must* be biocompatible.
2. There should be sufficient drug solubility in the microemulsion system.
3. The system should have a long shelf life.
4. The rate of release and bioavailability of drug should meet the need of the patient.

Few studies have been done on rate of release of drugs from well-defined micellar structures. Some of the pertinent literature follows.

Figure 6 compares the pharmacokinetic profile of patient- administered with cyclosporine entrapped in microemulsion (Sandimmune® and Neoral®) and without microemulsion (Sandimmune). The bioavailability of drug was found to increase 5.2-fold when the drug was injected with Sandimmune and Neoral. Leslie et al.[135] demonstrated that mid-

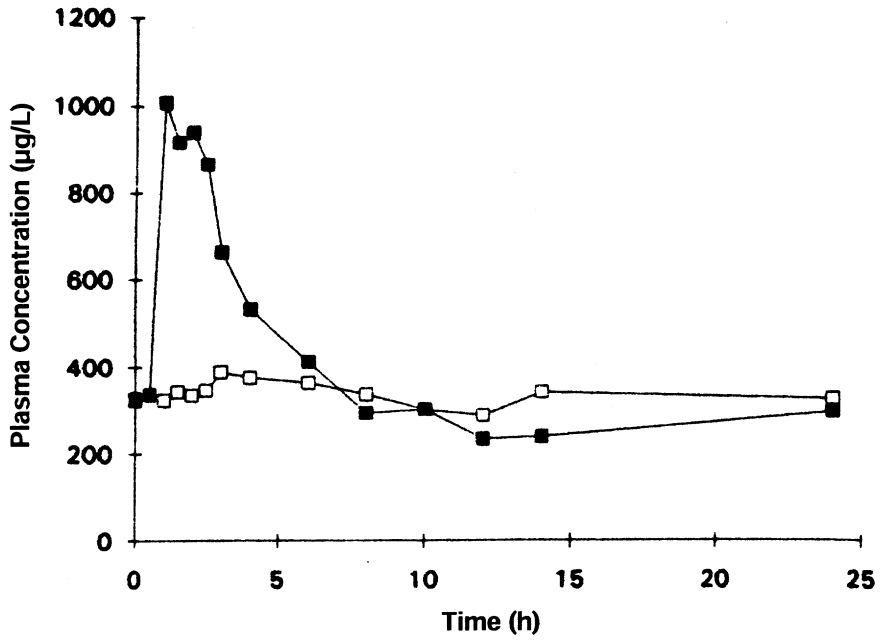

FIGURE 6. Whole blood cyclosporine concentration versus time profiles in a liver transplant recipient following single dose of Sandimmune 350 mg (□) and Sandimmune and Neoral (■) as determined by specific monoclonal antibody radioimmunoassay. (Reprinted with kind permission of Harvey Whitney Books.[258])

dle-phase microemulsions show high encapsulation of protein and bovine hemoglobin and slow release rate.

Ritschel et al.[136,137] compared the bioavailability of the drug in the microemulsion with that in commercial form using dog and rat models. The higher relative bioavailability of the microemulsion systems compared to the commercial preparation was attributed to their lower droplet size, although differences between the two microemulsion formulations suggested that an additional mechanism might be involved. Later, Ritschel[61] investigated the gastrointestinal absorption of three peptides using a series of o/w microemulsion formulations, concluding that the systemic peptide uptake from microemulsion in the gastrointestinal tract was also dependent on type of lipid phase of the microemulsion, digestibility of the lipid used, and types of surfactant in the microemulsion. A detailed hypothesized mechanism of peptide absorption from microemulsions given perorally was proposed.

1. Biocompatibility

As previously mentioned, the major disadvantages of using microemulsion formulations as vehicles for drug delivery systems are the high surfactant and cosurfactant concentration (> 10% in general) and the subsequent possible side effects resulting from the surfactants and cosurfactants required to create these formulations. Most of the microemulsion formulations used for technological applications in the oil recovery, nanoparticle, paint and ink, and other industries are composed of surfactants, cosurfactants, and oils incompatible with human physiology. Short-chain alcohols other than ethanol, for example, are not biocompatible.

The surfactants used for pharmaceutical microemulsions should be nonirritating. Phospholipids, particularly lecithin, offer a possible nontoxic alternative emulsifier for parenteral use. The cosurfactant should be carefully chosen as well. Most existing short- and medium-chain alcohols currently used as cosurfactants are useless for pharmaceutical microemulsions because of their toxicity and irritancy.[131] Furthermore, evaporation of these alcohols can destabilize the system, resulting in decrease of formulation shelf life.[60] Therefore, the development of biocompatible surfactants and cosurfactants for use in pharmaceutical microemulsions is becoming a very important technological challenge.

2. Solubility and Stability

A microemulsion is not an inert vehicle. Adding new components such as drugs to the system may affect its phase behavior. For instance, solubilized drug molecules may affect the spontaneous curvature of the surfactant in different ways, either by being incorporated into the surfactant film or by changing the polarities of the polar and/or apolar phases.[138] However, it is important to note that in some water-in-oil microemulsions investigated as vehicles for nanoparticles growth, the solubilization of certain salts was found to actually increase, not decrease, the stability of the microemulsion. Hence, the effect of drug solubilization on microemulsion stability may or may not be a disadvantage in a particular microemulsion formulation.[58,60,139]

Drug stability is also an important consideration. The effect, whether beneficial or detrimental, that the microemulsion will have on the drug must be determined. Figure 7 demon-

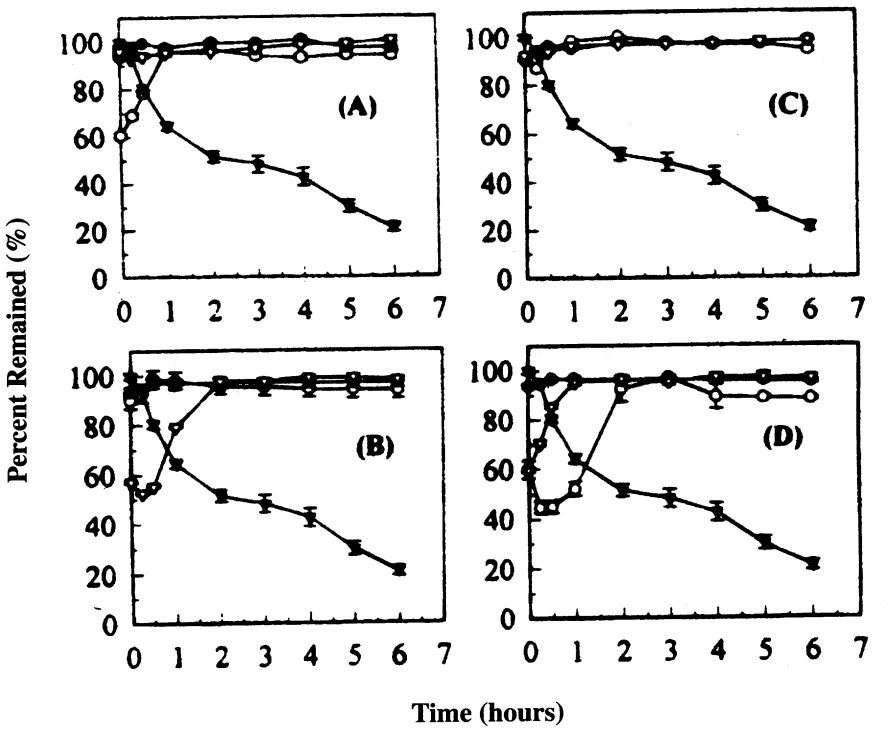

FIGURE 7. Stability of insulin in microemulsion formulation dissolved in 0.1 N HCl at 37 °C: (**A**) ○, SO750/ethanol; ●, SO750/1-propanol; ∇, SO750/1-butanol; (**B**) ○, MO500/ethanol; ●, MO500/1-propanol; ∇, MO500/1-butanol; (**C**) ○, MO750/ethanol; ●, MO750/1-propanol; ∇, MO750/1-butanol; (**D**) ○, ML310/ethanol; ●, ML310/1-propanol; ∇, ML310/1-butanol; ▼, insulin only (n = 3). (Reprinted from Hsiu et al. J Pharm Sci 1996 85(2):143[257] by permission of John Wiley & Sons, Inc.)

strates the ability of a microemulsion system to protect insulin from acidic degradation at 37 °C for at least 6 hours. Changing the o/w partitioning of the drug can alter the rate of release of drug from a microemulsion formulation. For example, as can be seen in Figure 8, by adding octanoic acid, Gasco et al.[140] increased the lipophilicity of propranolol and the partitioning of propranolol to the oil phase, which resulted in a decrease in the drug release rate.

It is possible to control drug release rate by microemulsion structure and composition. For example, Table 4 shows how a lecithin w/o microemulsion decreased drug release rate substantially when compared to aqueous solution.

3. *Bioavailability*

Drug bioavailability is another factor that must be considered when choosing a microemulsion formulation. Bioavailability is the fraction or percentage of a dose that reaches the systemic circulation intact when not directly injected into the circulation. By maximizing bioavailability, one maximizes the dose concentration or blood level of the drug. Also, if the drugs

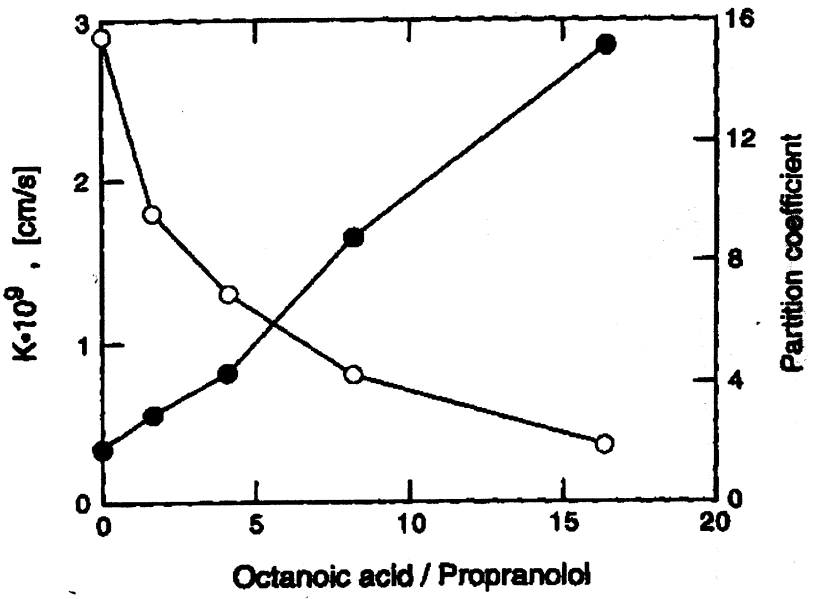

FIGURE 8. (●) Isopropyl myristate/buffer partition coefficient and (○) permeability coefficient *K* of propranolol over a hydrophilic membrane from Tween 60–isopropyl myristate–butanol–water microemulsions with varying octanoic acid/propranolol ratios. (Reprinted from 161 with permission from Kluwer Academic/Plenum Publisher.)

TABLE 4

Half-Life Values (h) of Technetium Activity in Rabbits Injected with Technetium in w/o Microemulsion or Aqueous Solution

Rabbit	Microemulsion	Aqueous Solution
1	132	10.6
2	151	12.5
3	57a	17.9
4	46a	12.1
5	69a	9.8
6	122	8.2
7	189	9.2

[a] These animals showed biexponential decay; half-lives reported are those of monoexponential functions with the same intercept and the same blood concentration versus time integrals as those of the biexponential fit.

Reprinted with permission from Bello et al., J Pharm Pharmacol, 1994;46:508.[255]

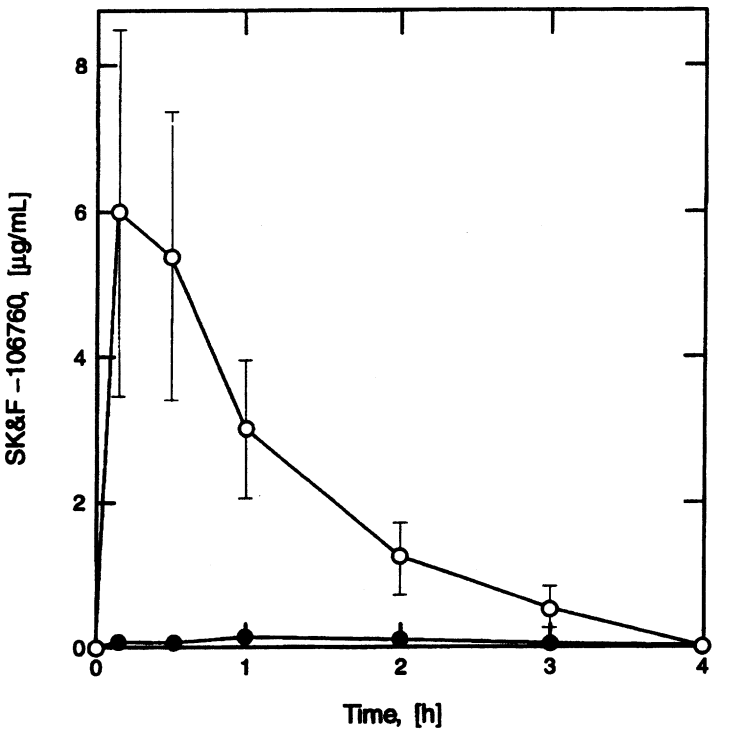

FIGURE 9. Plasma concentration of SK&F-106760 as a function of time after intraduodenal administration from (○) and aqueous solution or (●) a microemulsion. (Reprinted from 176 with permission from Plenum and Chapman & Hill.)

being administered are expensive to produce, maximizing bioavailability increases cost effectiveness. The bioavailability of a drug can be increased dramatically using microemulsion formulations. For example, as can be seen in Figure 9, the bioavailability of SK&F-106760, a water-soluble RGD fibrinogen receptor antagonist, is much greater in a microemulsion formulation than in aqueous solutions. The low oral bioavailability resulting from poor membrane permeability of many peptide drugs, including the one found in Table 5, can be increased by adsorption enhancement with microemulsions.

B. Methods of Drug Delivery

All three types of microemulsions, oil-in-water, middle-phase, and water-in-oil, can be used as drug delivery vehicles. Each type of microemulsion system can dissolve water-soluble, oil-soluble, and interface-soluble drugs or related compounds. The extent to which each type of compound is solubilized, however, depends on the microemulsion's structure and composition. Jayakrishnan et al.[141] studied the solubilization of hydrocortisone by w/o microemulsions and found that the amount of drug incorporated into the microemulsion depends on

the concentration of both the surfactants (Brij 35/Arlacel 186) and the cosurfactants (short-chain alcohols). The limit of solubilization depends on the partition coefficient of the drug in oil and water, and on the relative volume of oil and water in the system. The water-external (o/w) microemulsion presumably can be diluted by the aqueous phase of the stomach and the intestine. The oil-external (w/o) microemulsion will probably go through an inversion or destabilization upon dilution with stomach or intestinal aqueous phase. Depending on the structure of the surfactant molecules in the microemulsion, the surfactant may enhance the penetration rate or rate of transport of drug through the intestinal wall.[142] It has been reported that the rate of penetration of drug is much faster from microemulsion formulations than from other drug delivery vehicles.[61]

TABLE 5
Introduodenal Bioavailabilities of an RGD Peptide (SK&F 106760) in the Rat from w/o Microemulsion of Different Composition and Particle Size

ME	Composition[a] (%, w/w)	Droplet diameter[b] (mean ± sd)	%F[c] (mean ± sd) $n = 3$
ME1	Captex 200/Capmul MCM/Centrophase 31/ Cremophor EL/Saline (68.3/8.3/1.6/16.5/5.3)	26.4 ± 11.0	29.1 ± 7.1
ME2	Captex 355/Capmul MCM/Tween 80/ Saline (65/2V10/3)	15.2 ± 4.2	27.4 ± 8.9
ME3	Captex 200/Centrophase 31/Cremophor EL/ Saline (76.5/1.6/16.6/5.3)	585.2 ± 303.7	19.4 ± 11.8
ME4	Captex 200/Capmul MCM/Myverol 18-92/ Cremophor EL/Saline (76.5/9.3/1.0/10.0/3.2)	29.5 ± 10.1	14.4 ± 4.4
ME5	Captex 200/Capmul MCM/Centrophase 31/ Tween 80/Saline (76.6/9.3/2.1/8.7/3.3)	18.3.± 4.8	7.4[d]
ME6	Myvacet/Capmul MCM/Myverol 18-92/ Cremophor EL/Saline (76.9/9.1/1.0/9.8/3.2)	16.5 ± 4.0	5.4 ± 2.2
ME7	Captex 200/Dicaprin/Centrophase 31/ Cremophor EL/Saline (76.5/9.3/1.0/10.0/3.4)	760.1 ± 296.7	2.5 ± 1.9
ME1-C[e]	same as in ME1	same as in ME1	5.3 ± 3.6
Saline	NA	NA	0.5 ± 0.3

[a] For chemical names of excipients, refer to Table 1.[133]

[b] Based on particle number distributions (peptide-free microemulsions).

[c] $F = (AUC'd/AUCiV) \times (Dose_{iv}/Dose_{id}) \times 100$; where AUC is the area under the plasma concentration-time curve in mg.min/ml; the administered dose of the peptide (mg/kg) was 6.5 for ME1 and ME3, 8.4 for ME2, and 10 for saline and the rest of the microemulsions; the administered microemulsion volume was 3.3 ml/kg.

[d] $n = 1$

[e] Administration of ME1 (peptide-free) first, followed in 15 min with 10 mg/kg of peptide in saline.

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Pharmaceutical microemulsions can be delivered by three routes: parenterally (by injection), orally, and topically (to the skin and eyes). The method by which the drug is delivered dictates the constraints that will be encountered in formulation with regard to solubility, bioavailability, toxicity, and site targetability of the microemulsion/drug system.

1. Parenteral Delivery

Microemulsions have advantages over conventional parenteral (injectable) drug delivery systems for a wide variety of reasons. Parenteral administration (especially via the intravenous route) of drugs with limited solubility is a major problem in industry because of the extremely low amount of drug actually delivered to a targeted site. Until recently, not many studies have been published in which a microemulsion was used to administer drugs by the intravenous route,[143-145] but a number of pharmaceutically acceptable microemulsions for parenteral drug delivery have appeared in recent years.[129,130] Microemulsion formulations have distinct advantages over macroemulsion systems when delivered parenterally, because fine particle microemulsions are cleared more slowly than coarse particle emulsions and, therefore, have a longer residence time in the body. Both o/w and w/o microemulsions can be used for parenteral delivery.[1] The type of microemulsion employed is usually determined by the intended route of delivery as well as the role that the microemulsion will play. Although the literature contains details of many microemulsion systems, few of these can be used for parenteral delivery because of toxicity of the surfactant, cosurfactant, and/or oil phases. As already discussed, most short-chain alcohols other than ethanol are not acceptable for parenteral use. An alternative approach was taken by von Corswant and Thorén[138] in which C_3–C_4 alcohols were replaced with the parenterally-acceptable cosurfactants polyethylene glycol (400)/poly(ethylene glycol) (660) 12-hydroxy-stearate/ethanol, while maintaining a flexible surfactant film and a spontaneous curvature near zero to obtain an almost balanced middle-phase microemulsion. The middle-phase structure was preferred in this application because it has been able to incorporate large volumes of both oil and water with a minimal concentration of surfactant.[146]

2. Oral Delivery

Oral drug formulations are generally preferable to parenteral drug formulations. They are less frightening to children and so are easier to administer to them as well as to people with difficulty swallowing solid dosage forms. Microemulsion formulations offer several benefits over conventional oral formulations for oral administration, including increased absorption, improved clinical potency, and decreased drug toxicity.[147] Therefore, microemulsions have been reported to be ideal for oral delivery of drugs such as steroids, hormones, diuretics, and antibiotics. Table 6 lists the common food emulsifiers and their legal status, and Table 7 lists some common excipients used to formulate lipid microemulsions for oral drug delivery.

Pharmaceutical drugs of peptide and protein origins are highly potent and specific in their physiological functions. However, most are difficult to administer orally. With an oral bioavailability of most peptides in conventional (i.e., non-microemulsion based) formulations of less than 10%, they are usually not therapeutically active by oral administration.[148]

TABLE 6
Food Emulsifiers and Their Legal Status

Chemical name	Abbr.	ADI value[a]	U.S. FDA EU no.	U.S. FDA 21 CFR
Lecithin	—	Not limited	E 322	§ 184.1400[b]
Monodiglycerides	MG	Not limited	E 471	§ 184.1505[b]
Acetic acid esters of monodiglycerides	ACETEM	Not limited	E 472a	§ 172.828
Lactic acid esters of monodiglycerides	LACTEM	Not limited	E 472b	§ 172.852
Citric acid esters of monodiglycerides	CITREM	Not limited	E 472c	§ 172.832
Diacetyltartaricacid esters of monodiglycerides	DATEM	50	E 472e	§ 184.1101[b]
Succinic acid esters of monodiglycerides	SMG	—	—	§ 172.830
Salts of fatty acids (Na, K)	—	Not limited	E 470	§ 172.863
Polyglycerol esters of fatty acids	PGE	0–25	E 475	§ 172.854
Propylene glycol esters of fatty acids	PGMS	0–25C	E 477	§ 172.856
Sodiumstearoyl-lactylate	SSL	0–20	E 481	§ 172.846
Calcium stearoyl-lactylate	CSL	0–20	E 482	§ 172.844
Sucroseesters of fatty acids	—	0–10	E 473	§ 172.859
Sorbitanmonostearate	SMS	0–25	E 491	§ 172.842
Sorbitan tristearate	STS	0–15	E 492	—
Polysorbate-60	PS 60	0–25	E 435	§ 172.836
Polysorbate-65	PS 65	0–25	E 436	§ 172.838
Polysorbate-80	PS 80	0–25	E 433	§ 172.840

[a] Acceptable daily intake in mg/kg body weight per day
[b] Generally recognized as safe (GRAS)
[c] Calculated as propylene glycol
Reprinted from Krog NJ, Food emulsions, 1997, p. 142,[256] by courtesy of Marcel Dekker, Inc., New York, NY.

Because of their low oral bioavailability, most protein drugs are only available as parenteral formulations. However, peptide drugs have an extremely short biological half-life when administered parenterally. Because of this, multiple injections are generally required for parenteral administration of peptide drugs.

Gomez-Orellana and Paton[149] reviewed the recent patent literature related to formulation strategies to improve oral bioavailability of proteins. The peptide drug by far most frequently studied in relation to oral bioavailability is cyclosporine, an immunosuppressant drug commonly delivered orally to transplant patients to help prevent organ rejection. A microemulsion-based formulation of this drug, named Neoral, has been introduced to replace Sandimmune, a crude oil-in-water emulsion cyclosporine formulation. Neoral is formulated with a finer dispersion, giving it a more rapid and predictable absorption profile and less inter- and intrapatient variability.[150-152] It was found that no penalty was paid by converting stable renal transplant recipients from Sandimmune to Neoral. In fact, doing so

TABLE 7
Common Excipients Used to Formulate Lipid Microemulsions for Oral Drug Delivery

Excipient (HLB)	Chemical Definition	Manufacturer[a]
Arlacel 80 (4.3)	sorbitan oleate	ICI Americas
Arlacel 186 (2.8)	monoolein: propylene glycol (90:10)	ICI Americas
Capmul MCM (5.5–6.0)	C_8/C_{10} mono-/diglycerides from coconut oil	Abitec
Captex 200 (oil)	C_8/C_{10} diesters of propyiene glycol from coconut oil	Abitec
Captex 355 (oil)	C_8/C_{10} triglycerides from coconut oil	Abitec
Centrophase 31 (4.0)	Liquid Lecithin	Central Soya
Cremophor EL (13.5)	polyoxyethylene glycerol triricinoleate 35 DAC	BASF
Labrafac CM 10 (10)	C_8/C_{10} polyglycolyzed glycerides from coconut oil	Gattefosse
Labrafil M 1944 CSD (3–4)	primarily oleic acid ($C_{18:1}$) polyglycolyzed glycerides from apricot kernel oil	Gattefosse
Labrafil M 2125 CS (3–4)	primarily linoleic acid ($C_{18:2}$) polyglycolyzed glycerides from corn oil	Gattefosse
Labrasol (14)	C_8/C_{10} polyglycolyzed glycerides from coconut oil	Gattefosse
Miglyol 812 (oil)	C_8/C_{10} triglycerides from coconut oil	Huls, America
Myvacet (oil)	distilled acetylated monoglycerides	Eastman Chemicals
Myverol 18-92 (3.7)	distilled sunflower oil monoglyceride (90% glyceryl linoleate)	Eastman Chemicals
Soybean Oil	primarily oleic (25%) and linoleic (54%) triglycerides	Croda
Tagat TO (11.3)	polyoxyethylene (25) glycerol trioleate	Goldschmidt Chem.
Tween 80 (15.0)	polyoxyethylene (20) sorbitan oleate	BASF

Amount of added water needed for lecithin "gelation" is expressed as wo (gel) = $[H_2O]/[\text{lecithin}]$.[223]

[a] ICI Americas is located in Wilmington, DE; Abitec is located in Columbus, OH; Central Soya is located in Fort Wayne, IN; BASF is located in Parsippany, NJ; Gattefosse is located in Westwood, NJ; Huls, America is located in Piscataway, NJ; Eastman Chemicals is located in Kingsport, TN; Croda is located in Mill Hall, PA; Goldschmidt Chem. is located in Hopewell, VA.

resulted in effective, safe, increased drug exposure, reduced intrapatient variability, and the potential for reduced chronic rejection and thus greater long-term graft survival.[153] Therefore, microemulsion formulations have much potential in increasing the viability of oral delivery of cyclosporine, and further research can lead to similar results for other peptide drugs.

3. Topical Delivery

Topical administration of drugs can have advantages over other methods for several reasons, one of which is the avoidance of hepatic first-pass metabolism of the drug and related

toxicity effects. Another is the direct delivery and targetability of the drug to affected areas of the skin or eyes. Recently, there have been a number of studies in the area of drug penetration into the skin.[125,154] For example, Gasco et al.[155] studied the transport of azelaic acid, a bioactive substance used for treating a number of skin disorders, from a microemulsion system to abnormal skin. Several groups have been investigating the use of microemulsion formulations as ocular drug carriers.[156] The results of these studies seem promising. It was found that *in vitro* corneal penetration of indomethacin using microemulsions, for example, was more than 3-fold that of currently available commercial eye drops.[137] Microemulsions were also found to prolong the time of drug release, increasing the duration of time the drug spends in the body. An example of this is found in Figure 10, which shows the concentration of the drug timolol in rabbit eyes versus time. At first glance, it seems that timolol in solution is more effective than timolol in a microemulsion. However, by looking closely one can see that the concentration of timolol in solution decays much more rapidly than that in the microemulsion. Hence, the microemulsion formulation exhibits a longer presence in the body.

C. Pharmaceutical Microemulsions Using Nonionic Surfactants

Interest in nonionic surfactants for pharmaceutical microemulsion formulations is increasing because of their low irritation and high chemical stability. Traditionally, nonionic surfactants of the polyoxyethylene class have been used in the formulation of these microemulsions.[157] Ho et al.[147] formed microemulsions using polyglycerol fatty acid esters as non-

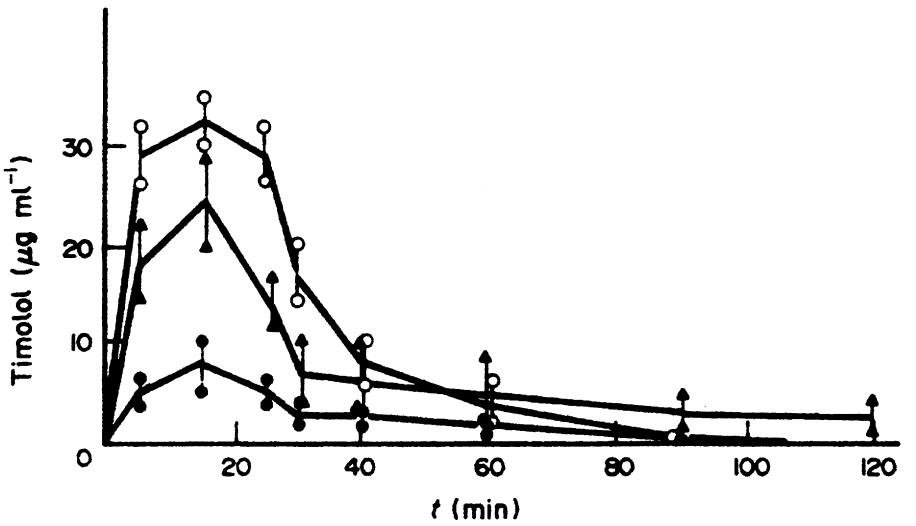


FIGURE 10. Aqueous humour concentration-time profiles following multiple installations in rabbits' eyes: (●), timolol alone; (○), timolol as an ion-pair in solution; (▲), timolol as an ion-pair in microemulsion. (Reprinted from J Pharmaceut Biomed, 7(4), Gasco et al., Microemulsions as topical delivery vehicles: ocular administration of timolol, p. 433, 1989,[188] with permission from Elsevier Science.)

ionic surfactants and short-chain alcohols as cosurfactants. From tests such as stability and viscosity measurements, they concluded that an oral insulin delivery system employing microemulsions as carriers is applicable. Jayakrishnan et al.[141] reported the solubilization of hydrocortisone by w/o microemulsions containing a mixture of the nonionic surfactants Brij 35 (polyoxyethylene 23 lauryl ether) and Arlacel 186 (glycerol monooleate-propylene glycol), isopropanol as cosurfactant, water and n–alkane. The influence of oil-soluble surfactant (Arlacel 186) concentration, the oil chain length, and the alcohol concentration on the amount of water solubilized in the w/o microemulsion were studied. Solubilization of water in microemulsions was found to increase with increased chain length of the alkane between C-8 and C-16 and increase in concentration of Arlacel 186, with a maximum water solubilization found for a 5:1 weight ratio of Arlacel to Brij surfactants. A formulation containing 10 mL decane, 4 mL isopropanol, 5 g Arlacel 186 and 1 g of Brij 35 was found to incorporate about 8 mL of water. Ritschel, et al.[137] studied this microemulsion, further modified by the addition of silicium dioxide (6–10%) to obtain a gel-like structure, for its effectiveness for delivery of cyclosporine A. Arlacel 186/Brij 35 microemulsions containing isopropanol as cosurfactant with several oil phases, including lauric acid hexyl ester, oleic acid, oleyl ester, glycolyzed ethoxylated glycerides of natural oils, and a mixture of branched fatty acids with 13 molecules of ethylene oxide were investigated.

In other formulations, Osipow[158] reported studies on o/w microemulsions prepared by mixing the nonionic surfactants polysorbate 60 (Tween 60) and sorbitan monooleate (Span 80) with glycerol as cosurfactant and liquid paraffin as oil. Attwood et al.[159] examined the influence of surfactant ratio (polysorbate/sorbitan monooleate) and oil content on the phase diagram of microemulsions. Ktistis[160] and Attwood et al.[159] reported studies on o/w microemulsions prepared using polysorbate surfactants, sorbitol, isopropyl myristate, and water. Gasco et al.[161] used microemulsions of similar composition using polysorbate 60 with phosphate buffer to study the *in vitro* release of propranolol. Several researchers have prepared o/w microemulsions using polysorbate 20 (20.8%) in combination with water/glycerol mixtures (58.3%), decanol/dodecanol (2:1 wt. ratio) (14.1%), and 1-butanol (6.8%) for use in topical delivery of azelaic acid. Malcolmson et al.[130] examined the effects of varying the nature (i.e., size and polarity) of oil on the solubilization of testosterone propionate into o/w microemulsions produced by Brij 96 (polyoxyethylene 10-oleyl ether). Their results suggest that unless a drug has a significant solubility in the dispersed phase and this dispersed phase is localized within the microemulsion core, there may be no advantage in solubilizing drug in an o/w microemulsion rather than a micellar system. Lee et al.[162] used nonionic hydrophilic surfactants (Poloxamer 338) to direct the lipid microemulsion system away from reticuloendothelial system (RES)-rich tissues so that they are able to reach other target tissues. The regions of existence of microemulsions of water and isopropyl myristate were studied by Carlfors et al.[163] as a function of the hydrophile–lipophile balance (HLB) of nonionic surfactant mixtures in order to find microemulsions stable at skin temperature. They observed that the addition of lidocaine lowered the phase inversion temperature (PIT) of the system and increased the temperature range for microemulsion stability. Therefore, nonionic surfactant-based microemulsion formulations for pharmaceutical applications show significant potential and promise as a next-generation delivery system.

The use of a nonionic surfactant, such as n-alkyl polyoxyethylene ether (C_mE_n) (where m is hydrocarbon chain length and n is the number of oxyethylene units), to stabilize a microemulsion is particularly attractive because it is generally possible to create a microemulsion

without the use of a cosurfactant. This has important advantages from a pharmaceutical viewpoint. First, as already discussed, most cosurfactants are not pharmaceutically acceptable. Second, it is not always possible to dilute a microemulsion containing cosurfactant, whereas microemulsions stabilized only by surfactant appear to be infinitely dilutable.[58] Third, nonionic surfactants are generally recognized as the least toxic of the surfactants and are currently used in a variety of pharmaceutical products.[131] Microemulsions formed with $C_{12}E_5$ surfactant have been the subject of detailed studies[85,86,164] and have been well defined over the past few decades.

D. Pharmaceutical Microemulsions Using Anionic Surfactants

Many pharmaceutical microemulsions have been developed recently using anionic surfactants instead of or in addition to nonionic surfactants. For example, addition of nonionic surfactant sorbitan monolaurate (Span® 20, Arlacel® 20) to the anionic surfactant Aerosol OT (AOT, Dioctylsulfosuccinate sodium salt) increases the phase boundary of w/o microemulsions of AOT.[165] With hexadecane as the oil phase the maximum solubilization occurs for a 1:1 weight ratio of sorbitan monolaurate to AOT. This microemulsion formulation is suitable for topical drug delivery applications because irritancy caused by the presence of various medium-chain length alcohol cosurfactants is absent.

In further studies, Osborne et al.[166] have shown that sorbitan monolaurate acts as a cosurfactant for the AOT microemulsion system and significantly affects its phase diagram. They have also observed that the mixing time for the waxy AOT solid with viscous sorbitan monolaurate is quite long, but this can be overcome by using ethanolic AOT solution or AOT-75 (5% ethanol, 20% water, and 75% AOT). Very low concentrations of ethanol are accepted pharmaceutically because of ethanol's low irritancy and toxicity compared to other alcohols. Maximum water incorporation into the microemulsion occurs for ratios of 55:45 and 60:40 for AOT-75 to sorbitan monolaurate and is independent of the ratio of hexadecane to surfactant mixture in the hexadecane range of 20–50%.

Addition of an oil (isopropyl myristate) to the w/o ternary microemulsion systems of AOT/water/medium chain alcohol can result in the formation of an o/w microemulsion. Trotta et al.[124,125,129] and Gasco et al.[161] produced o/w microemulsions with butanol as cosurfactant. A typical formulation is AOT (14 wt.%), isopropyl myristate (19.6 wt.%), buffer pH 6 (56 wt%), and butanol (10.4 wt.%). Of course, this formulation has limited applicability because of the toxicity of butanol.

In an attempt to gain insight into the characteristics and properties of a microemulsion vehicle fundamental to the topical delivery of drugs, Osborne et al.[167] investigated the w/o microemulsion region formed by water/octanol/AOT. They found that the delivery of the polar water portion of the microemulsion is highly dependent upon the composition of the microemulsion. Skodvin et al.[168] investigated the interaction between pilocarpine chloride and chloramphenicol, and the microemulsion systems AOT/octanoic acid/water, sodium octanoate/octanoic acid/water, and didodecyldimethylammonium bromide (DDAB)/dodecane/water. They found that pilocarpine chloride interacts with the colloid structures as an electrolyte, while chloramphenicol seems to be located in the interfacial (w/o) membrane. García-Celma et al.[169] investigated the use of microemulsions composed of Tween 80 (polyoxyethylene-20 sorbitan monooleate) surfactant and isopropyl myristate and isopropyl palmi-

tate oils for topical administration of antifungal drugs clotrimazole, ciclopirox olamine, and econazole nitrate. They reported that antifungal drugs with solubilities ranging from slightly soluble to practically insoluble in both water and oil can be successfully dissolved in a pharmaceutically acceptable ternary microemulsion system. In another application, Berthod et al.[170] attempted to use an o/w microemulsion system as a mobile phase for rapid screening of illegal drugs in sports using reverse-phase HPLC. The system heptane/water/sodium dodecyl sulfate (SDS)/n-pentanol was used to quantify 11 drugs in a reversed-phase liquid chromatography column. Although the limit of detection (LOD) with this method is not as low as in other techniques, microemulsion systems have the ability to assay water- and oil-soluble drugs at once. Hence, anionic surfactants used in microemulsion formulations also have significant potential as drug delivery vehicles.

E. Pharmaceutical Microemulsions using Phospholipids and Cholesterol

Aboofazeli and Lawrence[180] showed phospholipids, particularly lecithin, to be very good surfactant candidates for microemulsion formulations because of their very low toxicity. One problem in using lecithin as surfactant for microemulsions is that lecithin is slightly too lipophilic to spontaneously form the zero mean curvature lipid layers needed for balanced (middle-phase) microemulsions. Another problem is that in using lecithin in microemulsions, it is necessary both to adjust the HLB of the lecithin and to destabilize the lamellar liquid crystalline phases that have a strong tendency to form in these systems. Alteration of HLB can be achieved by adding a short-chain alcohol that makes the polar solvent less hydrophilic. In addition, incorporation of these weakly amphiphilic cosolvents in the polar parts of the lipid layers increases the area of the lipid polar head to produce the required spontaneous curvature of the lipid layers while also serving to decrease the stability of lamellar liquid crystalline phases.

These authors[180] have reported the phase properties of lecithin/propanol/water/n-hexadecane systems. About 2–3 wt % lecithin is the minimum amount required to form a microemulsion at all mixing ratios. The propanol concentration should be in the range 10–15 wt% of the aqueous solvent, with the concentration decreasing slightly with increase in oil content. From an examination of the microstructure of the system, these authors have shown a gradual transition from oil droplets in a water continuous phase through a middle-phase structure and then to water droplets in oil as the propanol concentration is decreased.

Oil-in-water microemulsions prepared using lecithin in combination with n-butanol as cosolvent have been recently reported by Attwood et al.[171] Figure 11 shows the influence of lecithin (egg)/butanol ratio on the phase properties in a system containing isopropyl myristate as oil phase. As the lecithin content increased with change of this ratio from 1:0.6 to 1:0.33, the amount of isopropyl myristate incorporated into the microemulsion increased from 8–19 wt % for the 1:0.6 ratio to 24–42 wt % for the 1:0.33 ratio. From a formulation viewpoint, the increased oil content obtained with the 1:0.33 ratio may provide a greater opportunity for solubilizing poorly water-soluble compounds. Substitution of soy lecithin for the egg lecithin produced differences in the size and position of the o/w microemulsion regions that were attributed to differences in the fatty acid impurities in the two types of lecithin, both of which were stated to contain 94–95% of phosphatidylcholine. These stud-

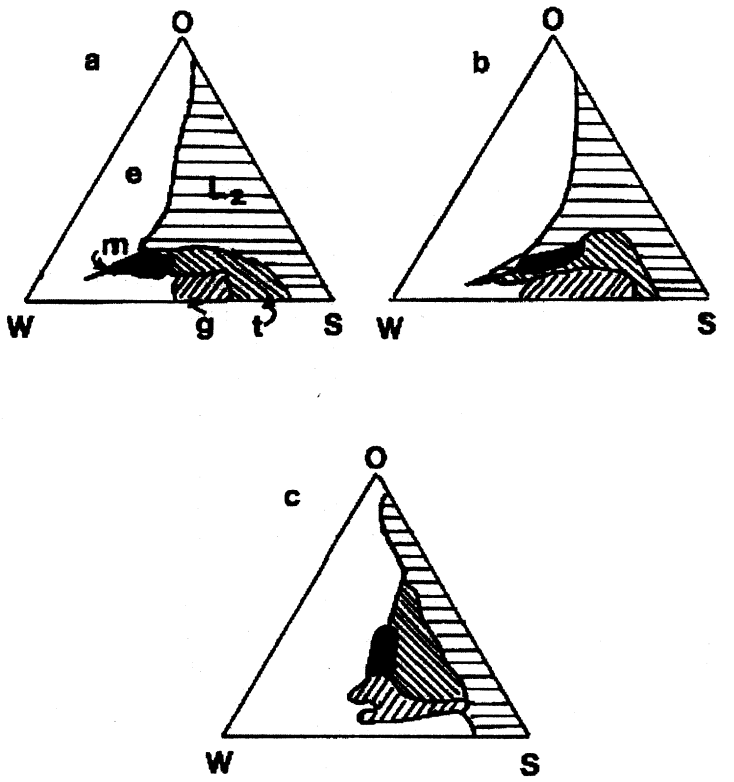


FIGURE 11. Partial phase diagram of the system (egg lecithin + butanol) (S)/isopropyl myristate (O)/water (W) showing stable o/w microemulsion (m), gel (g), monophasic turbid (t), unstable emulsion (e), and isotropic (L2) regions for lecithin/butanol weight ratios of **a**, 1:0.6; **b**, 1:0.45 and **c**, 1:0.33. (Reprinted from Int J Pharm, 84, Attwood et al., Phase studies in oil-in-water phospholipids microemulsions, p. R5, 1992,[179] with permission from Elsevier Science.)

ies were extended by Aboofazeli and Lawrence,[180] who recently reported phase properties for systems comprising water/lecithin/alcohol/isopropyl myristate in which the alcohols were n-propanol, isopropanol, n-butanol, s-butanol, t-butanol, and n-pentanol.

Other workers have used both o/w and w/o lecithin microemulsions as vehicles to deliver a range of drugs but have not reported detailed examination of phase properties. Fubini et al.[172] and Trotta et al.[124] have prepared microemulsions of egg lecithin, isopropyl myristate, butanol, and water with 3 lecithin/butanol ratios within the range examined by Attwood et al. [171] A variation of this formulation reported by Gallarate et al.[173] contained octanoic acid to enhance the solubility of timolol in the oil phase and also to facilitate ion-pair formation. The formulation used was egg lecithin (28.7 wt %), 1-butanol (14.9 wt %), and isotonic phosphate buffer pH 7.4 (0 wt %). The oil phase (16.4 wt %) contained solutions of octanoic acid and isopropyl myristate at several weight ratios (10.6:89.4, 15.9:84.1,21.1:78.9, and 28.4:71.6). An o/w microemulsion designed for the topical ad-

ministration of diazepam[125] contained a mixture of egg lecithin (5.6 wt %) and polysorbate 20 (10.1 wt %) as surfactant in combination with benzyl alcohol (8.9 wt %) as cosurfactant, and a series of mixtures of water and propylene glycol with weight ratios of 10:0, 9:1, 8:2, 7:3, 6:4, and 5:5 that constituted the continuous phase (56.3 wt %). Water-in-oil microemulsions containing egg lecithin (13.81 wt %), water (11.03 wt %), hexanol (8.6 wt %), and ethyl oleate (66.5 wt %) were shown to be suitable reservoirs for incorporation of deoxorubicine and 1-demethoxy-daunorubicine.[174,175] Constantinides[133] and Constantinides et al.[176-178] also have made an extensive study of the development of microemulsion formulations composed of medium-chain glycerides and medium-chain fatty acids for improved intestinal absorption of water-soluble compounds and proteins.

Considerable interest has been shown in the use of microemulsion drug delivery systems composed of phospholipids for the parenteral delivery of lipophilic drugs. Attwood et al.[179] studied the phase properties of the system isopropyl myristate/lecithin (egg and soya)/butanol/water. Aboofazeli and Lawrence[180] and Saint Ruth et al.[181] investigated the use of various short-chain alcohols, including ethanol, as cosurfactants in similar systems. Microemulsion systems containing phospholipids are rapidly captured after intravenous injection by the reticuloendothelial system (RES)-rich organs, such as the liver and the spleen, and by inflammatory cells. This can be advantageous in treating diseases localized to the RES182 or to macrophages.[183] Other than this specific case, however, rapid uptake of colloidal carriers by the RES is a significant disadvantage. Accumulation of drugs in the liver and spleen could be hazardous in the case of many drugs. In addition, rapid removal of drug by the RES prevents a targeted or sustained delivery of the drug.

Microemulsions also provide unique advantages as opthalmological carrier systems. No impairment of visibility is encountered, as is found with eye oils. Surfactant-containing multicomponent systems for ocular application have been developed and characterized.[184,185] Hasse and Keipert[186] studied o/w microemulsions made of lecithin and Macrogol-1500-glyceroltriricinoleate surfactants, polyethylene glycol 200 and propylene glycol cosurfactants, and isopropyl myristate oil for ophthalmic delivery of the drug pilocarpine hydrochloride. Such a preparation could be used as an alternative to pilocarpine eye oil, which affects visibility.

Kriwet and Müller-Goymann[187] studied the relationship between the colloidal structure of a topical formulation and the drug release *in vitro* as well as the influence of the microstructure on the stratum corneum drug permeability. They found that phospholipids are able to interact with the structures of the stratum corneum if they are applied as a microemulsion, as opposed to liposomal formulations and other investigated systems, where an enhanced drug permeability effect is not detectable. Gasco et al.[188] compared the effect of topical administration of timolol via microemulsion with aqueous solutions. Microemulsion-based delivery with lecithin resulted in a better concentration-time profile (3.5 times greater area under the curve for timolol in aqueous humour) than the aqueous timolol solution. The effect of short-chain alcohols as cosurfactants on pseudo-ternary phase diagrams composed of lecithin, water, and dodecane containing 1.0 wt% lidocaine was investigated by Choi et al.[189] in relation to the application of lecithin-based microemulsions for transdermal drug delivery. Halbert et al.[190] incorporated the cytotoxic drugs etoposide and methotrexate-α-benzyl-γ-cholesteryl diester into a microemulsion composed of egg phosphatidyl choline–cholesteryl oleate at a molar ratio of 2:1. They showed that it is possible to include these cytotoxic agents into microemulsion systems that possess a degree of toxicity when tested *in vitro*. It is quite evident that phospholipid and cholesterol-based micro-

emulsions have great potential as drug carrier systems, primarily because of their inherent biocompatibility. Such systems could become significant players in the future of targeted drug delivery applications.

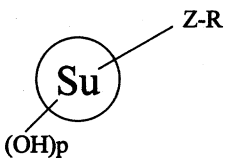
F. Pharmaceutical Microemulsions Using Other Surfactants

1. Sugar-Based Surfactants

Sugar-based surfactants have potential pharmaceutical applications because of their low toxicity, biocompatibility, and excellent biodegradability.[191] They offer an attractive alternative to the generally more convenient ethylene oxide (EO)-based nonionic surfactants discussed previously.[192] Sugar-based surfactants are prepared from natural and renewable resources that allow researchers to devise manifold structural combinations suited for modulating surfactant properties.[193] Furthermore, the phase behavior of this class of nonionic surfactant is much less influenced by temperature than the phase behavior of typical EO-based nonionic surfactants.[194] Sugar-based surfactants have the general formula shown in Figure 12. Bolzinger et al.[195] investigated the relationship between microstructure and efficacy of a sucrose-ester-based microemulsion as a drug carrier system. Sucrose esters are biodegradable surfactants whose hydrophilic and lipophilic properties can be adjusted by varying fatty acid chain lengths. Their pharmaceutical and industrial applications in microemulsion formulations have been reviewed by Garti et al.[196]

2. Fluorinated Surfactants

Fluorinated surfactants are uniquely characterized by very strong intramolecular bonds and very weak intermolecular interactions. They display exceptional thermal, chemical, and biological inertness, low surface tension, high fluidity, low solubility in water, and high gas-



Su: carbohydrate
p:2,4....
Z: O;OCO;S
R: n-C_nH_{2n+1}

FIGURE 12. Structure of sugar-based surfactant.[40] (Reprinted from Murdan et al., Water in sorbitan monostearate organogels [water-in-oil gels], J Pharm Sci 1999, 182[40] by permission of John Wiley & Sons, Inc.)

dissolving capacities. Because of this, microemulsions made from fluorinated surfactants are among the most promising candidates for producing suitable blood substitutes and other biocompatible fluids.[197,198]

Few articles exist in the literature describing the use of microemulsions as delivery systems for cytotoxic alkaloids. Alkaloids continue to represent one of the most potent groups of compounds for treating a large number of pathologies,[199] and future research in the delivery of these compounds using microemulsion formulations is essential.

V. DRUG DELIVERY WITH MICROEMULSION-BASED ORGANOGELS

A. Introduction and Potential as Drug Delivery Devices

In the late 1980s, a new class of formulations was described in the literature that has since been applied to numerous applications. The formulations are gels produced from water-in-oil microemulsions, and they have since been referred to (credited to Howe et al.[200]) as microemulsion-based organogels (MBG). The sol-gel techniques used for producing MBG have become popular in the last few decades because of their low processing temperatures and high chemical homogeneity. One of the most important advantages of doped sol-gel materials is their ability to preserve the chemical and physical properties of the dopants.[201] This advantage distinguishes these materials as unique hosts for a variety of biologically important molecules that can be exploited in a number of biomedical applications. MBG, which are classified as organogels because of the organic continuous phase, have been investigated in recent years for a number of applications, including biosynthetic media,[202–208] moisture sensors,[209,210] synthetic templates,[211] purification and separation media,[212] and carriers for liquid crystals.[213] MBG have also been shown in recent literature to have application as delivery devices for hydrophobic and hydrophilic drugs and vaccines.

As described previously, w/o microemulsions consist of nanodroplets of water dispersed in a continuous oil medium and stabilized by a layer of amphiphilic surfactant molecules. These hydrophilic and hydrophobic domains offer unusual solvent properties for macromolecules, especially for those that are amphiphilic. The unique structure of microemulsions may lead to pregel state conditions different from those encountered in usual gelling systems with polymers and colloids. The organogels formed also possess rather unique structural features.

The application of MBG to drug delivery is interesting for numerous reasons. A disadvantage of any aqueous-based formulation designed for the clinical environment is the potential for microbial contamination, and water-based hydrogels are no exception.[214] In these systems, metabolic action can result in the breakdown of gel structure, and pH changes and redox reactions can reduce drug efficacy.[215,216] These problems are overcome by the use of an organic continuous medium, such as that used in organogels, with the aqueous phase being the dispersed phase. Furthermore, in w/o microemulsions, these aqueous domains are in the nanometer size range and are consequently orders of magnitude smaller than typical bacteria. This makes microbial contamination in MBG much lower than that in hydrogels.[217] One problem with organogels is that most are more fluid than hydrogels and are quite soluble in the oil used as the continuous medium. Because this solubility would pose

problems in clinical applications, it is important to note that gelatin-based MBG are an exception and are not very soluble in the parent oil.[218]

Another advantage of MBG in pharmaceutical applications is that the gel-like reverse micellar systems are able to solubilize macromolecules (i.e., drugs) of different physicochemical properties. A hydrophobic active entity can be dissolved or dispersed in the continuous phase before gel formation. Likewise, a hydrophilic substance can be dissolved in the aqueous phase of the water-in-oil microemulsion prior to gel formation. Finally, an amphiphilic surface-active entity can be incorporated into the interface between the dispersed and continuous phases, although the additional contribution of this entity to the stabilization of the microemulsion droplets must be considered. Finally, organogels are interesting agents for drug delivery because of their unique biocompatibility and chemicophysical stability.[219] Moreover, the gels have been observed to be stable for a period of three months, with no changes in chemical composition or rheological properties.[220]

B. Gelatin Gels

Three major types of MBG have been extensively researched in the last decade, including gels based on gelatin (a polymeric material), lecithin, and, more recently, sorbitan monostearate. The formation of MBG from water-in-oil (w/o) microemulsions was first described by Haering and Luisi[221] and Quellet and Eicke.[222] The preparation of the gelatin gels was slightly different. Haering and Luisi reported stirring the gelatin powder in the prepared hydrocarbon micellar solution at room temperature for 30 minutes prior to heating to 50–60 °C. When the gelatin is dissolved at the elevated temperature, the turbid two-phase solution is cooled under vigorous shaking until the gel is formed. Quellet and Eicke[222] first allowed the gelatin to swell in water for 2 hours at room temperature. The system was then heated to 60 °C for 10 minutes to allow dissolution and was subsequently added to the surfactant/oil micellar solution, quenched at 17 °C, and allowed to set 1 week. Both studies, and most of the subsequent work with gelatin gels, used AOT anionic surfactant (bis 2-ethylhexyl sodiumsuccinate) and isooctane in their formulations.

The gelation of the gelatin microemulsions has been found to be highly dependent on the concentrations of water and gelatin in the system. Figure 13 shows a phase diagram of the gelatin-in-water/AOT/isooctane system.[223] The figure shows the different phases formed as a function of w_o, the water-to-surfactant molar ratio. The gel phase is identified as Phase II. This figure shows that the minimal water content to obtain a gel corresponds to about w_o {25, independent of AOT and gelatin concentration. The gel region also increases in size, with an increase in AOT concentration from Figure 13a, 100 mM, to 13b, 140 mM. However, the gelatin concentration required to obtain a gel also increases with increasing surfactant concentration.

More recent formulations using gelatin-based MBG have attempted to use pharmaceutically acceptable surfactants and oils.[217] The most acceptable surfactants have been found to be Tween 21, Tween 81, and Tween 85, which are ethoxylated sorbitan nonionic surfactants. However, MBG could not be formulated using these surfactants alone, so mixtures of up to 85% of these surfactants with the remainder being AOT were proposed. The oil phase investigated was isopropyl myristate in these formulations.

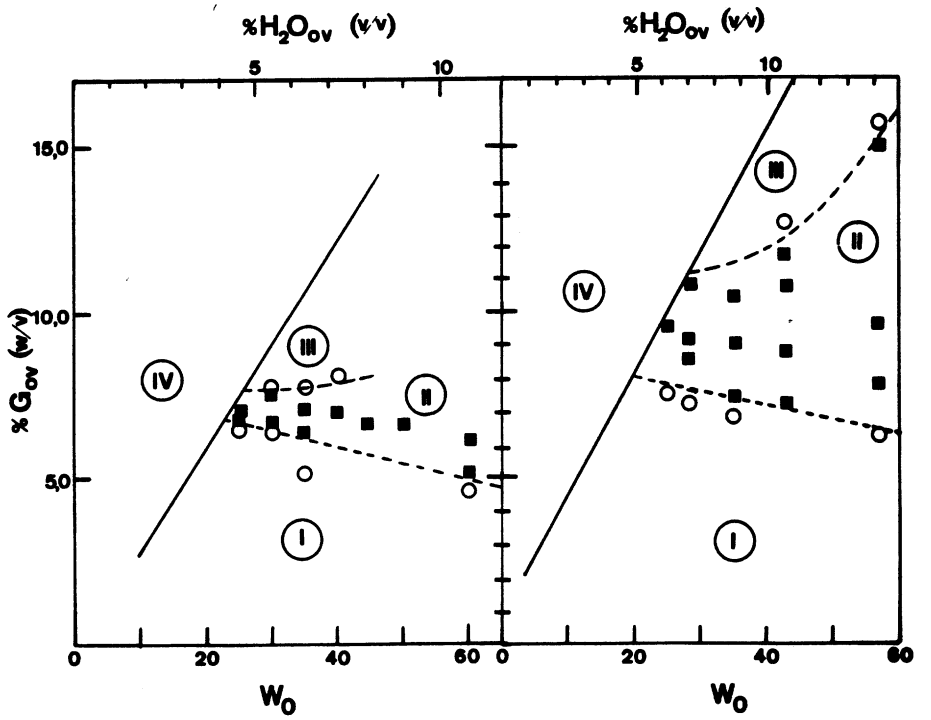


FIGURE 13. Stable regions of AOT/isooctane gels, with 100 mM (left side) and 140 mM (right side) AOT using gelatin Bloom 250. The 4 regions represent the different states of the micellar solutions: (I) liquid micellar gelatin solution; (II) gel phase; (III) two-phase region; (IV) insolubility of part of gelatin. (Reprinted from J Phys Chem, 90(22), 1986, p. 5892,[221] Fig. 1, with permission from American Chemical Society.)

1. Microstructure of Gelatin Gels

The structure of gelatin-based gels has been studied by pulsed NMR spectroscopy.[224] These studies reveal that in gelatin-based gels, the gelatin forms an "infinite" cluster with helical structures. The water in these gels is strongly bound to AOT or gelatin (from differential scanning calorimetry, DSC), but the electrical conductivity of the gels is significantly larger than that of AOT reverse micelles without gelatin. The schematic model of MBG proposed by Atkinson et al.,[225] in which this gelatin network is proposed to coexist with a population of conventional w/o microemulsion droplets, is shown in Figure 14. Gelatin incorporation into w/o microemulsions makes it possible to obtain gel matrices of comparable viscosity to those attainable using hydrogels. In addition, in contrast to the majority of other organogel formulations that are not microemulsion based, MBGs are electrically conductive, giving them potential applications in iontophoretic drug delivery, as described below.

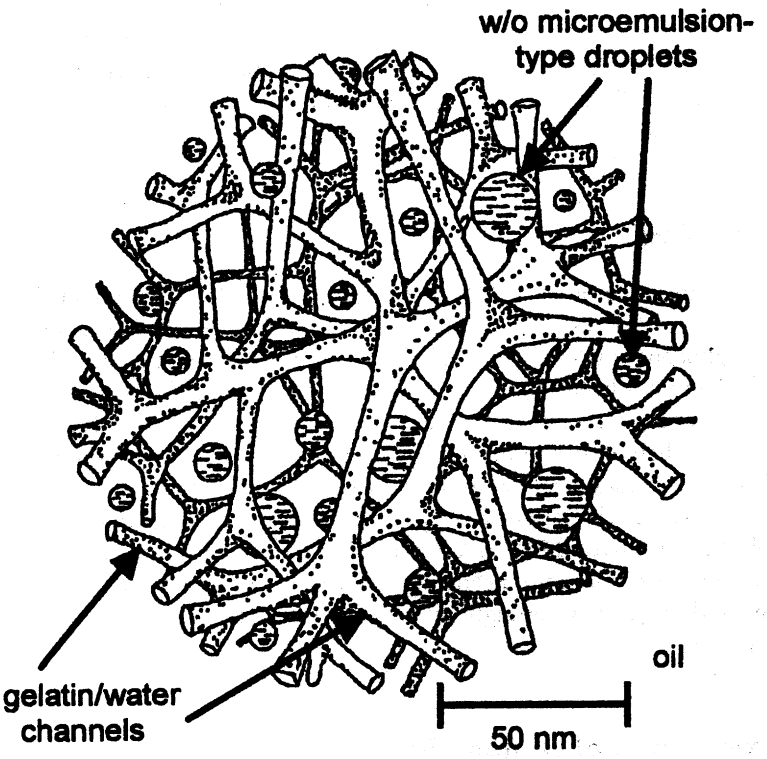


FIGURE 14. Proposed structure of gelatin MBG's based on small angle neutron scattering. (Reprinted from J Conrol Rel, 60, Kantaria et al., Gelatin-stabilised microemulsion-based organogels: rheology and application in iontophoretic transdermal drug delivery, p. 355, 1999,[218] with permission from Elsevier Science.)

2. Application of Gelatin-Based Gels to Drug Delivery

Because of the presence of electrically conductive channels within the gel matrix, gelatin-based gels have been investigated by Kantaria et al. [218] for iontophoretic drug delivery applications. For this investigation, the model drug sodium salicylate was passed through excised pig skin, and pharmaceutically acceptable surfactants and oils were used (with 15% or more AOT necessary to form the gel). Drug delivery rates with MBGs were broadly comparable to those attainable using aqueous drug solutions, and the transdermal flux was found to be proportional to the drug loading and the iontophoretic current density. The preparation and contamination advantages of delivery using MBG make this type of delivery an interesting subject.

One distinct problem in incorporating gelatin-based gels into drug delivery formulations is the inability to remove AOT from the formulation entirely. The attempted use of other surfactants has shown that the interaction of gelatin with AOT is very important, and gelatin is in fact not soluble in other surfactant/organic solvent microemulsions, even at high water con-

tent.[226] The presence of AOT makes gelatin gels less attractive to potential users because of the toxicity of AOT, so interest in gelatin gel usage has shifted to enzymatic reactions. If the AOT can be successfully replaced with a pharmaceutically acceptable surfactant, however, these gels could once again emerge as a major drug delivery candidate.

C. Lecithin Gels

The use of lecithin in producing MBGs was first described by Mahjour et al.[227] and Scartazzini and Luisi.[228] These gels use highly pure lecithin, with the best composition being 95–97% of phosphatidyl choline and 3–5% of lysophosphatidyl choline.[229,230] Also, the choice of solvent has been found to be important for the production of completely biocompatible organogels, retaining no irritant or allergic reactions. One advantage of lecithin-based gels is in their ability to form in a variety of solvents and even in mixtures of solvents. This versatile nature allows for modification of the viscoelastic properties of the gel in order to satisfy specific applications. Table 8 lists the solvents that are able to induce gelation in the solvent/lecithin/water system.[223] This table also lists the value of w_o (water-to-lecithin concentration ratio) needed to induce gelation. The biocompatibility and versatility of lecithin gels have brought about great interest in this type of system as a drug delivery device.

1. Microstructure of Lecithin Gels

Gelation is observed by an increase in viscosity by as much as a factor of 10^6 and passes through a maximum viscocity within a narrow range of added water, as illustrated in Figure 15. These viscosity values, and in particular the viscoelastic behavior of lecithin-based gels, are particularly interesting because the lecithin solutions contain no polymeric material and hence this phenomenon cannot be explained by the existence of a cross-linked polymeric network.

Structural investigations have led to some general theories of the origin of lecithin gels. ^{31}P-NMR and ^{2}H-NMR show no sign of anisotropy, which would suggest liquid crystalline phases.[231,232] In addition, small-angle neutron scattering (SANS) and small-angle X-ray scattering (SAXS) do not support the existence of cubic liquid crystalline order,[223] which means lecithin gels are a completely isotropic solution. However, rheology[233,234] and light scattering data[235] have shed light on the structure of lecithin gels. The gels form by self-association of lecithin molecules into long cylindrical reverse micelles and subsequent formation of an entanglement network. Because the observed dramatic increase of the viscosity is caused by the addition of water, it is proposed that this aggregation into cylindrical micelles is water induced. The water addition induces unique one-dimensional aggregation of lecithin molecules, as opposed to typical spherical growth of micelles in most microemulsion systems. The structural model proposed by Luisi et al.[223] for lecithin gels is shown in Figure 16A. This figure shows the structural transition of lecithin gels as water content is increased in these systems. The analogy to polymer solutions is shown in Figure 16B.

TABLE 8
Complete Representation of Solvents Able to Produce "Gelation" of Lecithin

Solvent	w_o (gel)	Solvent	w_o (gel)
Ethyl laurate	4	*N*-nonane	2
Butyl laurate	7	*N*-decane	2
Ethyl myristate	5	*N*-undecane	2
Isopropyl myristate	4	*N*-dodecane	1
Isopropyl palmitate	3	*N*-tridecane	1
Butyl stearate	3	*N*-tetradecane	2
Ethyl oleate	4	*N*-pentadecane	1
Ethyl erucate	2	*N*-hexadecane	1
Ethyl pentadecanoate	4	*N*-heptadecane	1
Isooctane	3	2,3-dimethylbutane	4
Cyclopentane	8	1-hexene	6
Cyclohexane	6	1-octene	4
Cycloheptane	7	1,7-octadiene	7
Cyclooctane	7	paraffin	0
Cyclodecane	12	(1R)-(+)-*trans*-pinane	6
Methyl cyclohexane	7	(1R)-(+)-*cis*-pinane	10
Tert-butyl cyclohexane	4	(1S)-(-)-*trans*-pinane	4
Bicyclohexyl	4	(1S)-(-)-α-pinane	4
Phenylcyclohexane	12	tripropylamine	4
1,3,5-triisopropylbenzene	3	tributylamine	2
Octylbenzene	6	triisobutylamine	3
Trans-decaline	5	trioctylamine	2
N-pentane	3	*N,N*-dioctylamine	2
N-hexane	3	Dibutyl ether	6
N-heptane	2	2-dodecen-1-ylsuccinic anydride	6
N-octane	2		

Reprinted from Coll Polym Sci, Organogels from water-in-oil microemulsions, Luisi et al., 268(4), p. 356,Table 5, 1990, © Springer-Verlag.[223])

2. *Lecithin Gels as Drug Delivery Vehicles*

The use of phospholipids *in vivo* is interesting because of the special kinds of aggregates they can build, including bilayers of membranes and reverse micellar aggregates, as proposed by De Kruijff et al.[236] The ability of lecithin gels to incorporate guest molecules is an interesting question, because there are in actuality three compartments where a guest molecule can be located. The small amount of water in these gels is essentially tightly bound to the phosphatidyl moeity of lecithin and so is expected to be significantly different than bulk water.[237] However, lipophilic substances are expected to be dissolved in lecithin gels as in the bulk organic phase. The differences in guest molecules present in the water and organic compartments of lecithin gels is shown in Figure 17.[223] The figure shows the UV absorp-

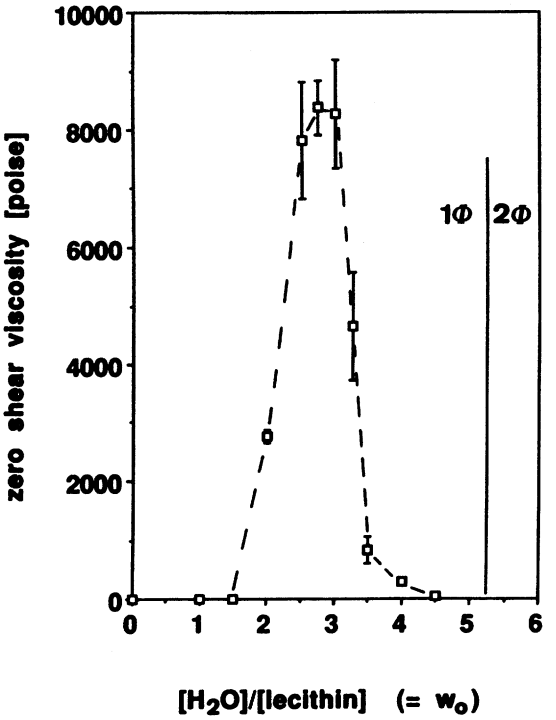


FIGURE 15. Zero shear viscosity, η_s, vs. ratio of added molecules of water per molecule of lecithin (w_o) for a lecithin concentration of 200 mM (ϕ = 0.145) in isooctane, T = 20 °C. The line indicates the phase boundary between the one-phase region, 1 ϕ, and the two-phase region, 2 ϕ. (Reprinted from Coll Polym Sci, Organogels from water-in-oil microemulsions, Luisi et al., 268(4), p. 356, 1990, © Springer-Verlag.[223])

tion spectra of perylene (A), a lipophilic substance, and erythrosin (B), a hydrophilic substance. Also shown are the UV spectra for perylen (A = dotted line) and erythrosin (B = line B+) without lecithin in isooctane and in water, respectively. The results for perylen show no shift in spectra between the bulk compartment and the gel phase. This verifies that the organic phase is not affected by the presence of the gel. The results for erythrosin, however, show that the water pool is significantly affected by the gel, as seen by the shift in spectra. This guest molecule, which is bulky and aromatic, is likely located at the micellar interface and shows different properties than the same molecule in bulk aqueous solution. Other hydrophilic substances, such as ascorbic acid and hydrophilic amino acids, are easily dissolved in lecithin gels without disruption of the guest molecule. These molecules, once forced into the gel, may sequester some of the water and build their own water shell inside the gel.[223]

The use of lecithin gels as drug delivery vehicles has shown exciting results in initial studies. As an example of application of lecithin gels, transdermal delivery experiments were carried out with the proteinase inhibitor tetra-p-amidinophenoxyneopentane (TAPP-Br). This drug has been shown to have antitumor capabilities, with *in vitro* and *in vivo* experiments showing consistently smaller tumor sizes than in control experiments.[230] Despite these therapeutic effects, significant chronic toxicity of the compound has been observed, which

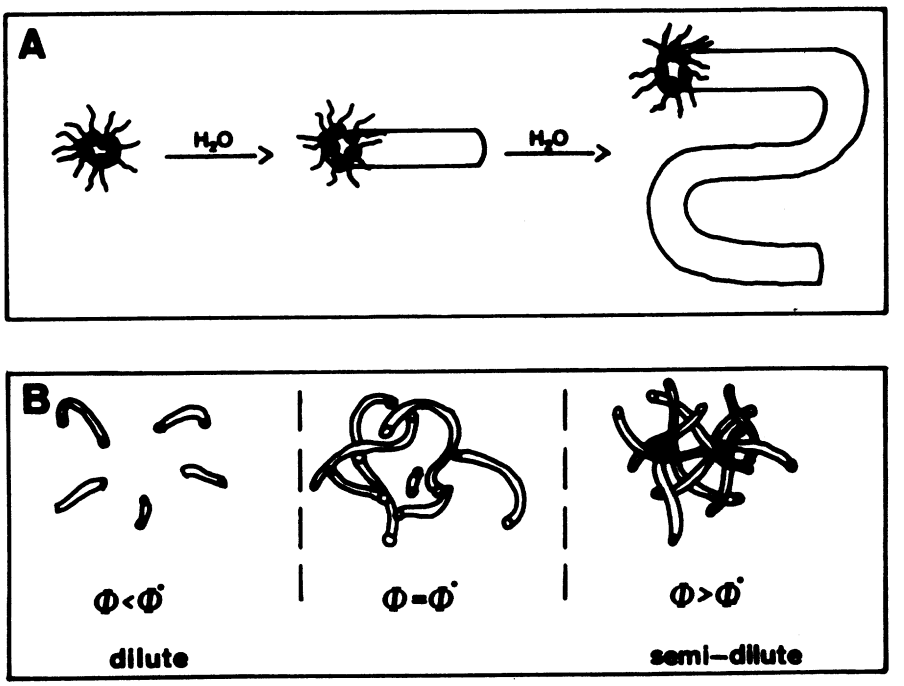


FIGURE 16. Schematic representation of a simple model for the structure of lecithin reverse micelles as a function of w_o. (**A**) Addition of water induces one-dimensional growth of the small lecithin aggregates into rod-like aggregates. Upon further addition of water, the rod-like aggregates grow into long, flexible cylindrical reverse micelles. (**B**) Analogy to polymer solutions: at a lecthin volume fraction $\phi < \phi^*$ (dilute regime), the micelles are not in contact; at $\phi = \phi^*$ the micelles start to overlap; and finally, at a lecithin volume fraction $\phi > \phi^*$ (semidilute regime), a transient network is formed. (Reprinted from Coll Polym Sci, Organogels from water-in-oil microemulsions, Luisi et al., 268(4), p. 356, 1990, © Springer-Verlag.[223])

could be circumvented by alternate routes of administration, such as vehiculation in gels. In order to investigate gel incorporation, lecithin gel containing TAPP-Br was applied on the skin directly surrounding the tumor legion. After different lengths of time, the relative size of the tumors developed by the mice were compared with those developed by mice treated with empty gels. The administration of lecithin gels containing TAPP-Br showed a significant reduction in the tumor mass compared to the empty gels and to mice subcutaneously injected with a Ha-ras-1-transformed cell line.

In a study by Willimann et al.,[238] lecithin gels obtained from isopropyl palmitate and cyclooctane (w_o = 3 and 12, respectively) were used in administration of scopolamine and broxaterol, with transdermal experiments done using a Franz diffusion cell and human skin obtained from cosmetic surgery. The transport rate of scopolamine was about 10 times greater in the gel than in an aqueous solution of the drug at the same concentration. However, the transport rate of scopolamine in the gel did not differ from that obtained in the microemulsion solution prior to gelation. The same variations in transport rates were observed for broxaterol, with the flux through the skin being directly proportional to the concentration of drug

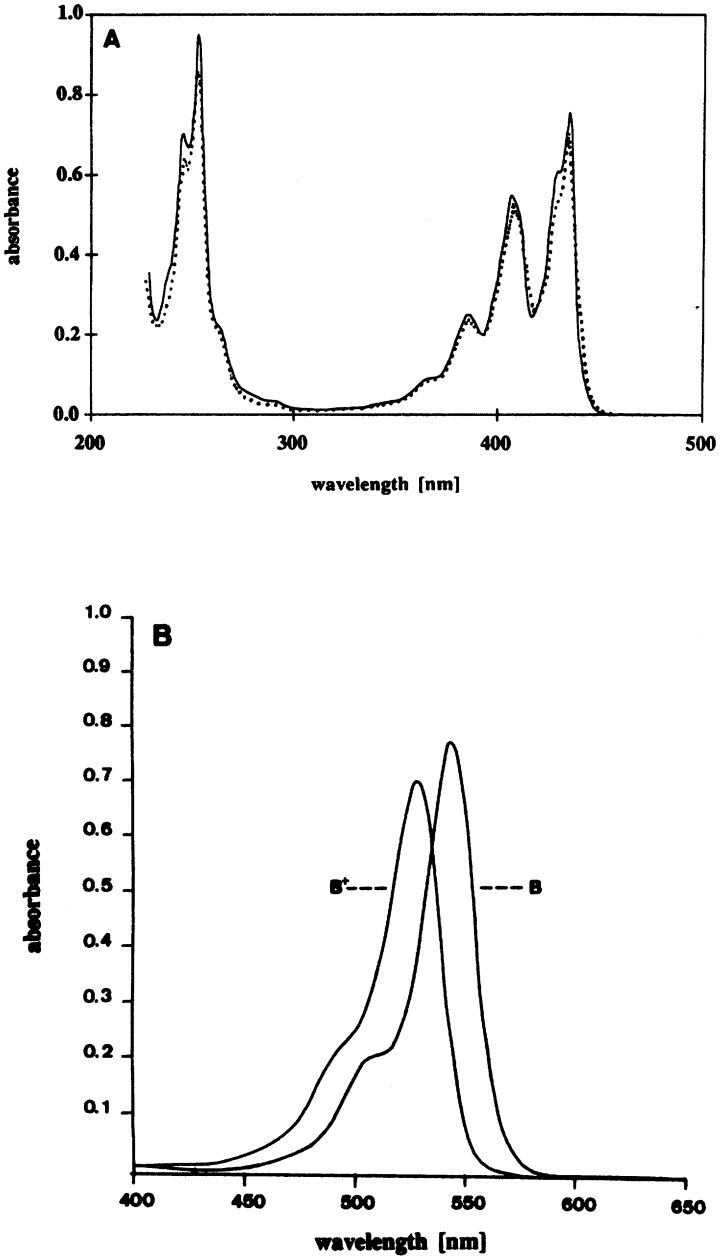


FIGURE 17. UV-absorption spectra of gels containing (**A**) perylen and (**B**) erythrosin. Gels are prepared with 50 mM lecithin (**A**) in isooctane and 200 mM lecithin (**B**) in *n*-octane with w_o = 2, respectively, and an overall concentration of 2 x 10^{-4} M for perylen and of 7.8 x 10^{-5} M for erythrosin. Also shown are the UV spectra for the same amount of perylen (dotted line) and erythrosin (**B***) without lecithin in isooctane and in water, respectively. The UV spectra were recorded at T = 23 °C using a cell of 0.1 cm path length. (Reprinted from Coll Polym Sci, Organogels from water-in-oil microemulsions, Luisi et al., 268(4), p. 356, 1990, © Springer-Verlag.[223])

in the gel. The flux of broxaterol was 47 μg/hr*cm^2 at a concentration of 75 mg/mL. Preliminary results with amino acids and peptides also showed successful transdermal transport.

One major advantage of lecithin gel administration is a reduction in the toxicity of drugs when administered in a gel-type vehicle. The toxicity of the lecithin microemulsion gel itself has been investigated in human skin irritation studies.[239] The study revealed a very low acute and a low cumulative irritancy potential for lecithin-based MBGs. The application of lecithin gels to pharmacology and cosmetics is therefore an area of great interest. The limited ability of the lecithin-bound water pools to solubilize certain hydrophilic drugs without structural changes is one disadvantage. However, the applicability of lecithin gels to incorporation of numerous hydrophilic and hydrophobic guest molecules has been demonstrated, with low toxicity.

D. Water-in-Sorbitan Monostearate Organogels

Multicomponent organogels based on water-in-oil microemulsions have also been prepared using sorbitan monostearate (Span 60) as the organogelator. As are lecithin-based gels, these gels are of interest as drug delivery vehicles because of the biocompatibility of sorbitan monostearate. These gels are synthesized by preparing a hot water-in-oil (w/o) emulsion and cooling to room temperature, at which time the w/o emulsion sets to an opaque, semisolid thermoreversible gel. This is a result of the reduced solubility of sorbitan monostearate in the solvent oil with cooling, which drives the surfactant to self-assemble into aggregates that form the microstructure of the gel.[240] The gelation process, observed by hot-stage light microscopy, begins by formation of inverse toroidal vesicles, which are analogous to typical inverse vesicles and liposomes except that they are not spherical in shape.[241] The short-lived vesicles give way to a network of tubules and fibrils containing the aqueous phase dispersed in the oil.[242] This microstructure, viewed by hot-stage microscopy, is shown in Figure 18, which shows both the w/o emulsion droplets at 60 °C (A) and the gel at 25 °C (B).[226]

The formation of these water-in-sorbitan monostearate organogels has been investigated with a variety of solvent oils, gelators, and additives to enhance gel properties. The gelation results using different solvent oils with sorbitan monostearate as the principal gelator show that more polar solvents such as ethanol and chloroform do not form gels, but that many alkanes (C > 5) and vegetable oils do form gels.[227] Gels have been found to form with sorbitan monostearate concentration as low as 1% w/v (0.02 M). The gel microstructure has been found to depend strongly on the solvent oil. The addition of polysorbate 20 (Tween 20), a hydrophilic nonionic surfactant, was found to have substantial effects on the gel lifetime and stability. Table 9[224] shows the effects of polysorbate 20 on both the solubility of sorbitan monostearate and the gel lifetime and stability with a variety of oils.[243] Microstuctural characterizations of the gels enhanced with polysorbate 20 show that the gels were composed of star-shaped clusters of surfactant tubules. These phenomena are as yet unexplained. However, the formation of these gels has been investigated in many publications.[244-246]

1. Water-in-Sorbitan Monostearate Gels as Drug Delivery Vehicles

The application of water-in-sorbitan monostearate gels to drug delivery has been attempted by Murdan et al.[240] These authors investigated the presence of a depot property when a gel

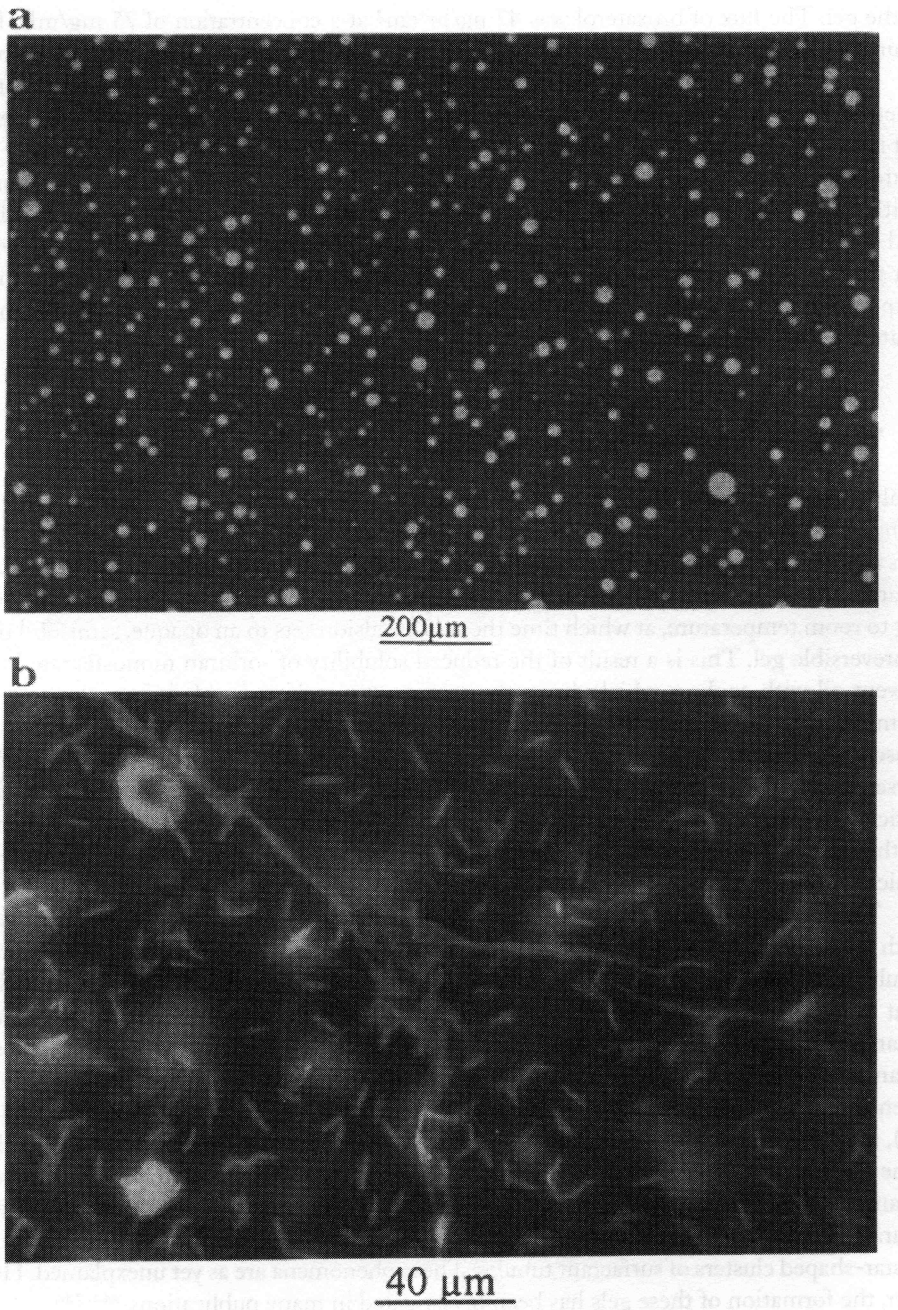


FIGURE 18. (**A**) Microstructure of a w/o emulsion at 60 °C: aqueous droplets (fluorescent CF solution) dispersed in the continuous oil phase. (**B**) Microstructure of a w/o gel at 25 °C: tubules and fibriles incorporating the aqueous fluorescent CF solution in the organic medium. Junction nodes are responsible for the integrity of the gel skeleton. (Reprinted from Murdan et al., Water in sorbitqn monostearate organogels (water-in-oil gels), J Pharm Sci 1999, 88(6):615[240] by permission of John Wiley & Sons, Inc.)

TABLE 9
Effects of Polysorbate 20 on Solubility of Sorbitan Monostearate in Hot Solvents (60 °C) and on Resulting Gels

Solvent	Effects of polysorbate 20 on solubility of sorbitan monostearate	Effects of polysorbate 20 on gel lifetime and stability
Hexane	⇓	⇓
Cyclohexane	⇓	⇓
Octane	⇓	⇓
Isooctane	⇓	⇓
cis-decalin	⇓	⇓
trans-decalin	⇓	⇓
Decane	⇓	⇓
Dodecane	⇓	⇓
Tetradecane	—	—
Hexadecane	⇑	⇑
Octadecane	⇑	⇑
Squalene	⇑	⇑
Ethyl oleate	—	⇑
Isopropyl myristate	—	⇑
Ethyl myristate	—	⇑
Cottonseed oil	—	⇑
Soybean oil	—	⇑
Sesame oil	—	⇑
Corn oil	—	⇑
Olive oil	—	⇑

increase (⇑), decrease (⇓), or no effect (—)

Reprinted from Murdan et al., Novel sorbitan monostearate organogels. J Pharm Sci 1999, 88(6):608[243] by permission of John Wiley & Sons, Inc.

containing a model radiolabeled antigen, bovine serum albumin (BSA), was given to mice by intramuscular administration. Isopropyl myristate was used as the organic solvent in the w/o gel. The performance of the w/o gel was compared to that of a w/o emulsion made by keeping the gel formulation at 60 °C, as well as to an aqueous solution of the antigen. After injection of the aqueous solution, almost all of the BSA is cleared within 8 hour. On the other hand, the w/o emulsion and the w/o gel release the antigen slowly over a few days, with similar release profiles. However, at 48 hours postinjection, the w/o gel exhibits depot performance superior to the w/o emulsion, with 20% of the injected antigen still present at the injection site. The rather short half-life of molecules at the injection site has been explained.[246] When the gel mass comes in contact with the interstitial fluid near the injection site, the latter penetrates into the organic gel via the sorbitan monostearate tubular network, resulting in gel breakdown into smaller fragments. Therefore, the surfactant tubular network aids in water penetration into the gel. Emulsification also occurs between the organogel and the aqueous phase because of the presence of surfactant. This mechanism results in the gradual erosion of the gel by oil droplets breaking off of the gel mass and leads to a relatively short duration of drug at the injection site.

Murdan et al.[246] made an interesting extension of water-in-sorbitan monostearate organogels in drug delivery applications by incorporating niosomes (vesicles with nonionic surfactant) into the aqueous phase of the gels, resulting in the formation of vesicle-in-water-in-oil (v/w/o) gels. The same model antigen, BSA, was used in a depot study and another antigen, haemagglutinin (HA), was used in an immunogenicity study. It was proposed that the vaccine in such v/w/o gels is entrapped in the niosomes, themselves located within the surfactant tubular network in organic medium. The depot study revealed that the BSA was cleared from the injection site over a period of days, once again showing a short-lived depot property resulting from interactions between the gel and local interstitial fluid. The immunogenicity study showed that the v/w/o gel, as well as one of the controls—the w/o gel with no vesicles—possess immunoadjuvant properties and enhance the primary and secondary antibody titers (of total IgG, IgG1, IgG2a, and IgG2b) to HA. The w/o gel (no vesicles) showed stronger immunoadjuvant properties than the v/w/o gel.[229] Another study revealed that when carboxyfluoroscein and 5-fluorouracil, an adjuvant for tetanus toxoid, is incorporated within the niosomes of a similar v/w/o gel containing cottonseed oil as organic phase, enhanced immunological activity is obtained over the free antigen or vesicles.[247,248]

These results indicate that water-in-sorbitan monostearate gels have performance as drug delivery vehicles similar to w/o emulsions made from the same formulation. The advantage of gel usage stems from the unique viscoelastic and thermoreversible nature of MBG. In addition, the novel v/w/o gels discussed here may offer additional advantages because of the ability to incorporate drugs into the internal or interfacial framework of the vesicles.

VI. SUMMARY

Microemulsions offer an interesting and potentially quite powerful alternative carrier system for drug delivery because of their high solubilization capacity, transparency, thermodynamic stability, ease of preparation, and high diffusion and absorption rates through skin when compared to solvent without the surfactant system. A number of factors must be considered when using microemulsions as drug delivery vehicles. First, the appropriate type of surfactant must be used in order to dissolve and protect the drug. The surfactant can either be cationic, anionic, nonionic, or catanionic (mixed cationic and anionic system). The type of drug dissolved and conditions of the target site will dictate the type of surfactant used to carry the medicine. Second, the concentration of surfactant largely influences the carrier system, because this surfactant concentration will ultimately determine drug delivery effectiveness. Last, the conditions of the target site are critical to the effectiveness of a surfactant delivery system. Parameters such as temperature and pH will influence the solubility of a drug inside the microemulsion system.

Although o/w microemulsions are promising solvent systems for drug delivery, compared to other drug delivery methods, their potential has not yet been reached. There are several barriers that need to be overcome before this technology can be used in practice. The main challenge is the control of drug diffusion and partitioning between the dispersed and continuous phases present.

Another factor influencing the use of microemulsions is the biological tolerance of the constituents of microemulsions, particularly the surfactants and cosurfactants, because sur-

factants can cause disruptions in biological membranes. A high surfactant concentration in the body over a long period of time may disturb some bodily processes.[142] Problems arising from the use of medium-chain-length alcohols as cosurfactants intended for topical administration has been addressed by Osborne et al.[166] These cosurfactants seem to be skin or eye irritants, and their use in topical delivery seems to be limited.

Nonionic surfactants such as ethoxylated alkyl ethers and sorbitan esters, as well as nonionic block copolymers (e.g., polyethylene oxide-block-polypropylene oxide) are generally less irritating and toxic than ionic surfactants.[249,250] Moreover, many nonionic surfactants have an advantage over charged surfactants in that they can form microemulsions even without cosurfactants.[251] Despite the reasonable tolerance of synthetic nonionic surfactants, particularly in topical applications, microemulsions prepared from phospholipids, sugar-based surfactants, and fluorinated surfactants seem to be preferred over those prepared by synthetic surfactants from a toxicity point of view. The preparation of microemulsion systems with these surfactants, however, is comparatively difficult, involving prolonged sonication in order to achieve the clarity normally associated with microemulsions. Complex size separation techniques are also often necessary to ensure uniformity of particle size.

The use of microemulsion-based gels (MBGs) in drug delivery applications has been described. It is evident that lecithin gels offer superior advantages in the drug delivery industry compared to gelatin gels. This is primarily a result of the biocompatibility of lecithin formulations and the relative ease with which they have been used. The use of gelatin gels in iontophoretic drug delivery remains an area of interest, however. More recently, water-in-sorbitan monostearate organogels have emerged as a leading candidate for drug delivery applications. These gels are biocompatible and show promising results in preliminary drug delivery studies. In general, the relative advantages of gel formulation compared to other innovative administration techniques such as microemulsion formulation, drug encapsulation in microspheres, and alternative routes of administration will ultimately determine the significance of MBGs in the pharmaceutical industry.

The use of microemulsions as drug delivery vehicles is an exciting and attractive area of research, offering not only many challenges to be overcome but also many potentially extraordinary benefits.

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