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PHASE DIAGRAMS OF LECITHIN-BASED MICROEMULSIONS CONTAINING SODIUM SALICYLATE

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ABSTRACT

Partial phase diagrams were constructed at 25°C to investigate the phase behaviour of systems composed of soybean lecithin, water, sodium salicylate, alcohol and isopropyl myristate. The lecithins used were the commercially available soy bean lecithins, namely E200 and E170 (phosphatidyl choline purities greater than 95% and 68-72% respectively). The cosurfactants employed were n-propanol, 2-propanol and n-butanol and these were used at lecithin/alcohol weight ratios (K_m) of 1:1 and 1.5:1. At a given K_m, the aqueous phase consisted of a 2%w/w sodium salicylate solution. Phase diagrams showed the area of existence of a stable isotropic region along the surfactant/oil axis (i.e., reverse microemulsion area). The extension of the microemulsion domain was influenced by the purity of surfactant, the lecithin/alcohol weight ratios and the kind of the alcohol.

Key words: Microemulsion, Cosurfactant, Partial phase diagram, Soybean lecithin, Sodium salicylate, Phase behaviour

INTRODUCTION

In 1943, Hoar and Schulman observed that in the presence of short chain alcohols (e.g., *n*- butanol), the unstable, turbid macroemulsions transform into solutions with particles of much smaller dimensions in comparison to the corresponding emulsions (1). Such systems were then defined as microemulsions (2). In general, microemulsions are self-emulsifying stable dispersion of oil and water, stabilized by interfacial film of surfactant molecules, frequently in combination with a cosurfactant. These systems are also characterized by their small particle size (generally less than 100 nm), stability toward wide range of temperature and they appear to be thermodynamically stable. These systems in contrast to the conventional emulsions are formed spontaneously without the need of any mechanical work. The structure, type, formation characteristics, stability, phase behaviour and the influence of different variables on the phase behaviour of microemulsions have been reviewed (3).

In the past three decades, microemulsions have been the focus of extensive research worldwide due to their importance in a variety of technological and pharmaceutical applications including drug delivery (4-9). Recently, investigations have focused on the use of microemulsion related systems called *microemulsion gels* which are formed by some surfactants under certain conditions (10-13).

In the previous studies (14, 15) the pseudo-ternary phase diagrams of water-isopropyl myristate (as oil) and lecithins (as surfactant) systems using a wide range of short chain alcohols (as cosurfactant) at four different surfactant/ cosurfactant weight ratios (K_m) were reported. The long term objective of this project is to evaluate the potential of lecithin-based microemulsions for an effective delivery of a hydrophilic solute (i.e., sodium salicylate as a model drug) transdermally. Therefore, as a starting point, we have investigated the influence of incorporation of sodium salicylate on the area of existence of solute-loaded lecithin microemulsions.

MATERIALS AND METHODS

Materials: Two commercially available soybean lecithins, Epikuron 200 (E200) and Epikuron 170 (E170) were obtained as a gift from Faratin Company (Lucas Meyer Representative in Iran) and used without further purification. Isopropyl myristate (IPM) and sodium salicylate were purchased from Sigma Chemical Company (Dorset, UK). *n*-Butanol, *n*-propanol and 2-propanol were obtained from Aldrich Chemical

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Company (Dorset, UK). Triple distilled water from a well-seasoned all glass steel was used throughout the study.

Construction of partial phase diagrams: Three component mixtures of lecithin-alcohol-IPM from 5% (w/w) total surfactant and 95% (w/w) oil to 50% (w/w) of each at 5% weight intervals were prepared and progressively enriched with 2%(w/v) sodium salicylate solution while being shaken for sufficient time in order to attain equilibrium at room temperature. The course of each addition was monitored through cross polaroids in order to determine the boundaries of any microemulsion and birefringence liquid crystalline domains. By repeating this experimental procedure for all different combinations of lecithin/alcohol to oil weight ratios, the phase boundaries of solute-loaded water in oil microemulsion domain were determined. No attempt was made to identify any other regions of the phase diagrams in detail.

In order to show the variation of components on a partial phase diagrams, the top apex of the triangle was used to represent the surfactant/alcohol ratio at a particular weight ratio (K_m), and the other apices to represent oil and sodium salicylate solution. The transparent, isotropic regions mapped on the phase diagrams were stable at least for one month at room temperature.

RESULTS AND DISCUSSIONS

The partial phase diagrams of systems containing E200, IPM, *n*-propanol, and sodium salicylate at two different K_m are shown in figure 1. The phase diagrams are overlaid because in all systems examined, samples became cloudy and ultimately phases were separated outside the microemulsion boundary and no significant difference in the phase behaviour was observed at high water concentrations. It should be noted that, since in most systems little difference was observed between phase behaviour of E200 and E170, only the phase diagrams for E200 systems containing *n*-propanol are presented. Any differences between the phase diagrams will be mentioned appropriately in the text. Tables 1 and 2 indicate the approximate amounts of water (containing 2% w/v sodium salicylate) solubilized in the microemulsion region, the corresponding surfactant/oil ratios and the total surfactant concentrations for E200 and E170 systems respectively, at each K_m tested.

Systems containing E200: In general, the following generalizations can be made about the systems which were examined.

a) Phase diagrams show the area of existence of a monophasic, stable, isotropic region along the surfactant/oil axis in all systems, regardless of the kind of alcohol and K_m.

b) At a given K_m , the amount of water incorporated in the microemulsion area increases with increase of total surfactant content.

c) At a given surfactant/oil ratio (particularly above the ratio of 15:85), the amount of solubilized water increases with the increase of K_m

d) The water solubilizing capacity of all systems below the surfactant/oil weight ratio of 15:85 (in a few cases, below 10:90), increases slowly.

e) The water solubilizing capacity, irrespective of the K_m and the type of lecithin, is least in the presence of *n*-butanol. Below surfactant/oil weight ratio of 20:80, a less pronounced difference in solubilizing capacity is observed between different studied K_m. These observations show dependence of water solubilizing capacity on the K_m of each system, particularly at the higher surfactant contents.

Systems containing E170: There was little difference in partial phase diagrams obtained when E200 was replaced by E170. Comparison of phase diagrams revealed that, irrespective of the kind of the alcohol and K_m, E200 produced, a larger microemulsion area (particularly at surfactant/oil weight ratios above 20:80), which was extended further into the water rich region of the phase diagrams. On the other hand, at the K_m of 1:1, the solubilizing capacity of systems composed of E170 was greatest in the presence of *n*-propanol and at the K_m of 1.5:1, 2-propanol possessed the greatest capacity to solubilize sodium salicylate solution. At high oil concentrations, no significant difference in the extent of microemulsion region was observed with lecithin type.

Attempts to use lecithin as an amphiphile for the preparation of an efficient microemulsion must take into consideration the characteristic solution properties of lecithin (16). These are; a) a very strong hydrophobicity due to a long hydrocarbon chains, b) a strong lipophobicity due to the heavily hydrated zwitterionic polar head groups, and c) a close balance between hydrophile and

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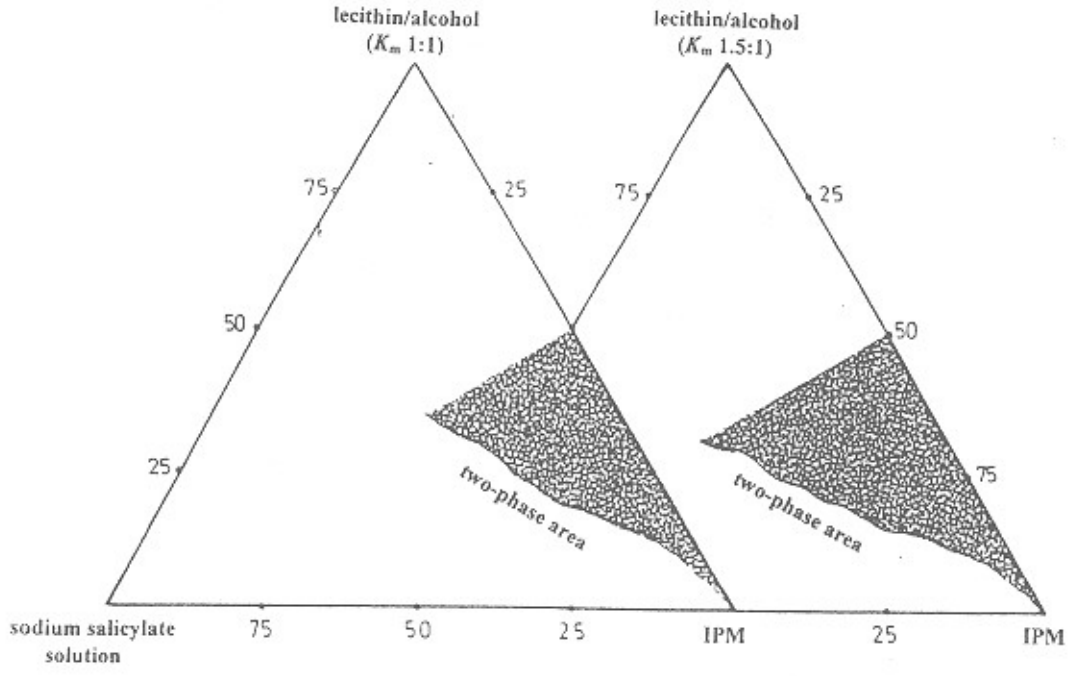

Figure 1: Partial phase diagrams of systems containing E200/n-propanol/IPM/ 2% sodium salicylate solution at different K_ms, constructed at 25 °C.

lipophile properties, which is slightly displaced towards the lipophilic side. In order to form lecithin microemulsions over a range of oil-water concentrations, the HLB of the lecithin should be adjusted by making the lipid more hydrophilic, the non-polar solvent more lipophilic or polar solvent less hydrophilic.

Lecithin as a naturally occurring biocompatible surfactant is capable of producing balanced microemulsions, in the presence of short chain alcohols (16). Alcohols (17-20) as cosurfactant, can decrease the polarity of the polar medium or incorporate into the lipid layer and change the critical Packing Parameter (CPP) of lecithin molecule (a parameter which determines the spontaneous curvature of the surfactant). These effects in turn, promote micellization of the lamellar or hexagonal phases and provide the appropriate conditions for the formation of microemulsions.

In the previous studies the phase behaviour of systems composed of lecithin/IPM/water prepared from a wide range of short chain alcohols at different K_ms were ivestigated and the influence of the kind of alcohol, differences in K_m and the surfactant purity were completely discussed (14,15). To elucidate the effect of loading a hydrophilic solute on the microemulsification process, the phase diagrams of systems containing similar components were constructed in the presence of sodium sallicylate. In general, at surfactant levels of 50% or less, phase diagrams were similar to those obtained in the absence of sodium salicylate, indicating that the extent of microemulsion realms was not affected significantly by incorporation of the solute. The general trend of changes induced by different lecithins and K_m, was also the same as that of solute free microemulsions (i.e., increased solubilizing capacity by increasing purity and K_m).

Musana (21) has reported the changes in the nature of the phase diagrams by incorporation of 10% and 20% w/v sodium salicylate solutions in systems composed of IPM/O200 (Ovothin 200, egg lecithin) and alcohols. A decrease in the extent of microemulsion region and the formation of liquid crystalline phase

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were noted upon the addition of sodium salicylate at lower surfactant concentrations in the oil rich part of the phase diagrams. The decrease in the microemulsion area due to the presence of birefringent phase was attributed to an increase in the size of aggregates, which led to the rearrangement in the structure of aggregates. Musana (21) has assumed that incorporation of the additives do not affect the partitioning of the alcohols in the system and, constructed in the presence of sodium Salicylate. the changes are most likely as a result of the presence of the incorporated additives. Considering the chemical structure of sodium salicylate, it can be assumed that the solute would be incorporated at different sites of the aggregates, due to its surface activity as a consequence of having amphipathic nature. This molecule is more likely to align itself at the interface with the aromatic rings between the hydrocarbon chains and the carboxyl groups of the head groups.

Table 1. Solubilizing capacity of sodium salicylate solution for w/o microemulsion systems containing E200/alcohol/IPM

alcohol	surfactant/oil weight ratio (%)	K_m : 1:1 % water[a]	% surfactant[b]	K_m : 1.5:1 % water[a]	% surfactant[b]
n-butanol	50:50	24.50	37.80	31.00	34.50
	45:55	21.60	35.30	27.30	32.70
	40:60	20.00	32.00	24.50	30.20
	35:65	18.60	28.60	23.00	27.00
	30:70	16.70	25.00	20.00	24.00
	25:75	14.90	21.30	18.40	20.40
	20:80	13.00	17.40	16.70	16.70
	15:85	7.00	14.00	9.10	13.60
	10:90	4.80	9.50	7.00	9.30
	5:95	2.40	4.90	4.80	4.80
n-propanol	50:50	32.20	33.90	41.20	29.40
	45:55	29.80	31.60	36.50	28.60
	40:60	27.30	29.10	33.30	26.70
	35:65	24.50	26.40	31.00	24.10
	30:70	23.00	23.10	28.60	21.40
	25:75	20.00	20.00	24.50	18.90
	20:80	13.00	17.40	20.00	16.00
	15:85	7.00	14.00	11.10	13.30
	10:90	3.60	9.60	4.80	9.50
	5:95	2.40	4.80	3.60	4.80
2-propanol	50:50	40.10	29.90	46.70	26.70
	45:55	41.20	26.50	42.80	25.70
	40:60	37.50	25.00	39.40	24.20
	35:65	33.30	23.30	34.40	23.00
	30:70	29.80	21.00	31.00	20.70
	25:75	23.00	19.20	24.50	18.90
	20:80	21.60	15.70	23.00	15.40
	15:85	13.90	13.00	14.90	12.80
	10:90	4.80	9.50	7.00	9.30
	5:95	2.40	4.80	3.60	4.80

[a] 2%w/v sodium salicylate solution
[b] Total surfactant content required

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The increased presence at the interface would probably results in the formation of liquid crystalline phases at low surfactant concentrations. In this investigation, the extent of drug loaded microemulsion domain on the phase diagrams were increased as much as possible. This provided the possibility to examine effects of increasing the amount of drug solution on the release pattern (in some cases, due to the presence of an extensive microemulsion area, systems with up to 30% drug solution could be prepared for the purpose of release studies). Based on the Musana's report, 10% and 20% sodium salicylate would cause the formation of liquid crystalline phase in the oil rich part of the phase diagram and decrease the extent of microemulsion domain, and it was expected that the extent of microemulsion

Table 2. Solubilizing capacity of sodium salicylate solution for w/o microemulsion systems containing E170/alcohol/IPM

alcohol	surfactant/oil weight ratio (%)	K_m : 1:1 % water[a]	% surfactant[b]	K_m : 1.5:1 % water[a]	% surfactant[b]
n-butanol	50:50	23.00	38.50	27.30	36.40
	45:55	20.00	36.00	24.50	34.00
	40:60	18.40	32.70	23.00	30.80
	35:65	16.70	29.20	20.80	27.70
	30:70	15.00	25.50	19.20	24.20
	25:75	13.00	21.70	17.50	20.60
	20:80	11.10	17.80	13.00	17.40
	15:85	4.80	14.30	4.80	14.30
	10:90	2.40	9.80	3.60	9.60
	5:95	1.20	5.00	2.40	4.80
n-propanol	50:50	28.60	35.70	32.80	33.60
	45:55	24.50	34.00	29.80	31.60
	40:60	23.00	30.80	25.20	30.00
	35:65	20.00	27.70	23.00	27.00
	30:70	16.60	25.00	20.00	24.00
	25:75	14.90	21.30	16.70	20.90
	20:80	13.00	17.40	15.00	17.00
	15:85	7.00	14.30	9.10	13.60
	10:90	4.80	9.50	7.00	9.30
	5:95	2.40	4.80	4.80	4.80
2-propanol	50:50	26.00	37.00	34.40	32.80
	45:55	23.00	34.60	31.00	31.00
	40:60	22.30	31.10	28.50	28.50
	35:65	19.20	28.30	26.00	25.90
	30:70	15.80	25.30	23.00	23.00
	25:75	13.00	21.70	21.60	19.60
	20:80	9.10	18.20	16.70	16.70
	15:85	7.10	14.00	9.10	13.60
	10:90	4.80	9.50	7.00	9.30
	5:95	2.40	4.80	4.80	4.80

[a] 2%w/v sodium salicylate solution

[b] Total surfactant content required

domain would in-crease in the presence of lower

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amounts of the solute and the liquid crystalline phase disappearance. It is well established that the spontaneous curvature of the surfactant is determined by the ratio between its hydrophobic volume, head group area and hydrocarbon length. The results obtained in this research could be explained by considering the partition of sodium salicylate molecules at the interface which affects the curvature of the interface and favours formation of the more balanced microemulsions.

As mentioned earlier, in this study, a 2% w/v sodium salicylate solution was used for the microemulsification and it was observed that all samples became turbid outside the microemulsion region. Although, solubilizing capacity of microemulsions was shown to change due to the influence of the solute, the concentration of sodium salicylate seemed to be insufficient for the formation of liquid crystalline mesophase. However, by increasing the concentration of sodium salicylate, the area per lipid polar group increases and thus the spontaneous curvature of the lipid layer changes from being curved toward planar format which favours the formation of birefringent region.

The solubilizing capacity of systems containing E200, regardless of the kind of the alcohols and difference in K_m when titrated with 2% w/v sodium salicylate solution was found to increase in comparison with those obtained in the absence of the solute. When E200 was replaced with E170, the same behaviour was observed with *n*-butanol indicating that this alcohol is not significantly different in its influence on the microemulsion domain. In contrast, the microemulsion domain of all systems containing 2-propanol and *n*-propanol decreased when they were loaded with sodium salicylate.

Sodium salicylate is an ionizable solute that is primarily distributed in the aqueous phase. The presence of a charged moiety and its counterion could possibly influence the effective head group area, leading to a decrease in CPP and in turn, in an increase in the solubilizing capacity. E170 contains not only phosphatidylcholine, but also phosphatidylethanolamine, and other phospholipids and glycolipids. Each of the non-phosphatidylcholine components present in E170 could behave as cosutfactant and influence the effective CPP of lecithin in a different manner. On the other hand, there is a possibility that due to the presence of these molecules, the more hydrophilic alcohols are distributed primarily in the aqueous phase and since these components would not be expected to be as effective as alcohols in increasing the flexibility of the interfacial layer, the total solubilizing capacity is slightly decreased.

In E200 systems, it was observed that the solubilizing capacity increased in the presence of 2-propanol irrespective of the K_m, whilst in E170 formulated systems the formation of the largest microemulsion domain was favoured by the presence of *n*-propanol at the K_m of 1:1, and 2-propanol at the other K_m value. Although not significant, it is difficult to explain the differences observed in the extent of phases obtained with the two lecithins.

This research was performed on solute-loaded microemulsions in which the main goal was to evaluate the release pattern of these systems. Clearly it was necessary to study the phase behaviour of these systems initially and to find out whether it was possible to map clear and extensive microemulsion regions in the presence of sodium salicylate on the phase diagrams. The results obtained in this study showed that the preparation of transparent and stable microemulsions in the presence of a water soluble solute is reproducible.

ACKNOWLEDGEMENTS

The authors are so grateful for the financial support from the Research Deputy of Shaheed Beheshti University of Medical Sciences, Tehran, Iran.

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