Topical Delivery of Diclofenac using Microemulsion Systems

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Abstract

The purpose of this study was to investigate microemulsions as delivery systems for diclofenac sodium (DS). Microemulsion systems were composed of: soybean oil, nonionic surfactants (Brij 58 and Span 80), and different alcohols: ethanol, and 1-buthanol as cosurfactant. The optimum surfactant: cosurfactant (S:CoS) weight ratios and microemulsion areas were detected by the aid of ternary phase diagrams. Five microemulsion with various values of: DS, 0.25-2.5% enhancer (menthol, farnesol), 4-20% oil, 20-70% bi-distilled water, 14-70% the mixture of surfactant and co-surfactant (w/w) were selected, and their physico-chemical properties like: pH, viscosity and conductivity were determined.

Drug solubility in plain oil and microemulsions as well as partition coefficient respectively was estimated. The conductivity results showed that DS-loaded microemulsions have higher conductivity values (19.8-22.6 μS/cm) than unloaded formulations (15.5-17.2 μS/cm), and a load of DS into the formulation had no negative effect on system stability. Moreover, viscosity measurements were examined as a function of shear rate, and Newtonian fluid characterization was observed for each microemulsion system. All formulations had appropriate observed pH values varying from 6.50 to 7.15 for a topical application. The steady-state flux of DS from microemulsions and commercial gel were evaluated using Franz diffusion cells using synthetic membrane of carboximethylcellulose (CMC).

The results suggest that microemulsions are potential vehicles for improved topical delivery of diclofenac. The release and the diffusion of DS from both microemulsions and commercial gels through a synthetic membrane established the advantages of microemulsions instead of commercial gels.

Keywords: microemulsions, diclofenac sodium, transdermal delivery, enhancer

Introduction

Microemulsions are modern colloidal drug carrier systems [1]. A microemulsion, made from water, oil, surfactants and co-surfactant is a thermodynamically stable system. The presence of the co-surfactant is often required in order to decrease the interfacial tension of this interface, because the fact that a low interfacial tension is essential in microemulsions obtaining. Microemulsions represent an interesting and potential quite powerful alternative carrier system for transdermal drug delivery because of their high solubilisation capacity, transparency, thermodynamic stability, facility of preparation, and high diffusion and absorption rates.

Transdermal drugs delivery with absorption into systemic circulation occurs as a convenient way of administration because:

- it is easier and painless;
- it avoids direct contact between the drug and the liver;
- it avoids the action of aggressive triggers of intestinal tract against the drug;

- it provides a sustained drug delivery;
- it provides a pharmacological action against skin diseases.

Drugs penetration through the skin is a complex process which depends on various factors such as:

- the nature of the drug (hydrophilic or lypophilic drug);
- the nature of the vehicle (gel-state, emulsions, microemulsions , complexation of the drug with cyclodextrines, liposomes and nanoparticles);
- the nature of the membrane (synthetic, natural);
- presence of the enhancers (isopropyl myristat, isopropyl palmitat, hydrogenated soybean phospholipids, non-ionic surfactants, terpenes, alcohols with long carbon chain C_8-C_{12} etc.).

Microemulsions as topical drug delivery contain [2]:

- oil phase: saturated and unsaturated fatty acids, long chain alcohols, triglycerides etc.
- aqueuse phase;
- surfactants, without toxical power: Tween 20, Tween 80, Span 20, Span 80, Azone, Plurol isostearique, Plurol oleic etc.;
- cosurfactants: short-chain alcohols (ethanol, propanol, isopropanol, propylene glycol etc.), long chain alcohols (1-buthanol, decanol, octanol etc.).

In the last few years, many researchers have studied the advantages of using microemulsions as transdermal drug delivery vehicles including certain drugs as: ascorbic acid, 5-fluorouracil, triptolids, non steroidal anti-inflammatory drugs, lidocaine etc. [3-6].

The objective of this present study is to make a comparison between the DS delivery and diffusion from microemulsions and gels through the methylcellulose membranes. The diclofenac as the main substance is a non-steroidal anti-inflammatory drug.

Diclofenac, mainly as the sodium and diethylammonium salts of diclofenac are usually used for the local symptomatic relief of pain and inflammation in localised rheumatismal soft-tissues and following trauma of tendons, ligaments, muscles and joints .

Concerning this research it was followed these steps:

- to establish the proportion of each W/O microemulsion compound using the pseudoternary phase diagrams;
- to prepare the pharmaceutical microemulsions with or without DS;
- to understand the microemulsions structure (drop size, conductivity and viscosity measurements);
- to establish the DS solubility and bioavailability phase behaviour and to find the partition coefficient W/O phases;
- the study of the enhancers influence on the penetration coefficient of DS from microemulsions in comparison with its diffusion from the gel-state through the CMC membranes.

Materials and Methods

Reagents and apparatus

Soybean oil (Sigma), synthetic membrane of CMC with the pore size 0.22μm (Sigma), enhancers: menthol, farnesol (Merck)), DS 99.9% (pharmaceutical laboratory), Brij 58, Span 80 (Merck), cosurfactants: ethanol, 1-buthanol (Sigma). All the reagents were of analytical grade.

Ultrasonic homogenizers SONOPLUS HD 3200, Mastersizer-MALVERN INSTRUMENTS, Multiparameter INOLAB 720, Spectrophotometer UV-VIS T 80 +, Centrifuge MIKRO 200 R, Viscometer DV-II+, Stereomicroscope OLYMPUS 240.

Preparation of W/O microemulsions

In order to prepare the microemulsions it has been used soybean oil, a mixture of surfactants with HLB 5.44 made from Brij 54 and Span 80 in 9:1 (w:w), cosurfactant (ethanol, 1-butanol). The optimal ratio S/CoS has been established using the phase diagrams. These phase diagrams were made using a consequently proportion of S/CoS starting with 1:1. On this mixture of soybean oil and enhancers was added CoS and finally the product was ultrasounded 300 s at a frequency of 20 kHz, amplitude of 40% and duty cycle of 5 s. Microemulsions were obtained by a light ultrasound stirring of the mixture with distilled water for 150 s. Considering the systems turbidity results, it was realized the phase diagrams and it was established the optimum rates S/CoS. DS was added after the microemulsions preparation and the final product was ultrasounded for 60 s. The concentration of DS in microemulsions is up to 1.5% (wt/wt)

Determination of n-octanol-water and soybean oil-water partition coefficients

Before the experiment, the organic phase was saturated with water for 24 hours. It was used 6 centrifugal tubes containing 5 mL organic saturated phase in which was added 5 mL aqueous solution of DS 10^{-4} M. Those 6 tubes were stirred for 24 hours. It was a centrifugal power of 3000 rpm for 10 minutes in order to separate the phases.

The DS concentration in the aqueous phase was determined using the spectrophotometric analysis of the green coloured complex DS-Cu (II) at the wavelength of 680 nm [7].

Determination of DS solubility

The DS saturated suspension solubility in distilled water was determined using ultrasounds for 60 s. The sample was filtered using an acetate-cellulose filter with 0.45 μm pore diameter. The DS concentration in clear solution was determined using the spectrophotometric analysis of the green complex DS-Cu (II) at the 680 nm.

In vitro penetration study

The permeation and the diffusion of DS from prepared microemulsions were evaluated using Franz diffusion cells (surface 2 cm^2 receiver liquid volume 4.2 mL) in a 37^oC thermostatic bath. The receiver liquid contains phosphate buffer 10 mM, NaCl 120 mM, KCl 2.7 mM, and this solution has pH = 7.4. It was used a CMC membrane with the pore size of 0.45μm as diffusion barrier. This membrane was hydrated for 24 hours with phosphate buffer pH=7.4 at 20^oC.

The donor compartment contains 1g of microemulsion. It was collected 1mL of sample at every hour from the receiver compartment and it was established the amount of DS delivered and diffused. After each sample was collected the same volume of phosphate buffer solution in the receiver compartment was added.

Results and Discussion

Using the phase diagrams, the microemulsions compounds ratio of mixture was determined. The optimal ratio S/CoS is different, depending on CoS implication degree: surfactant/ ethanol = 6:1, and surfactant/ 1-buthanol = 4:1 respectively (Fig.1).

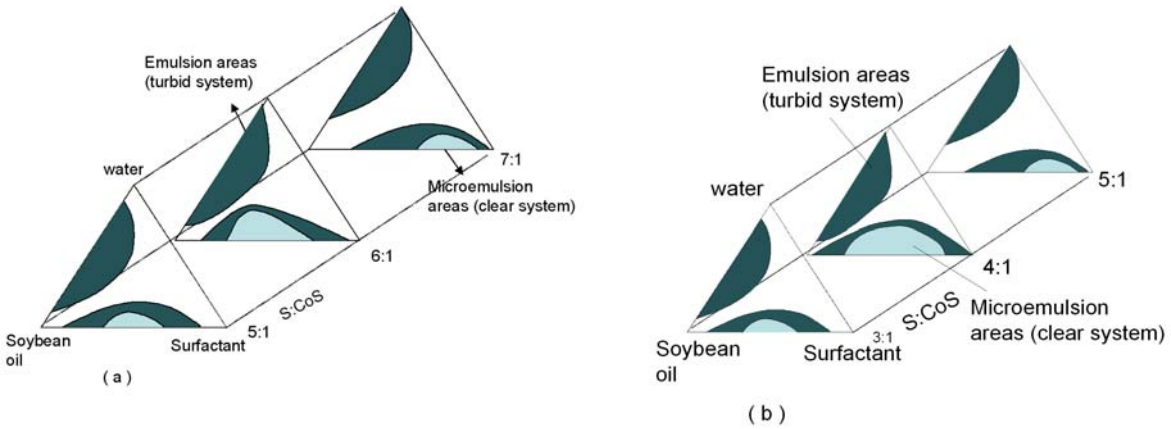

Figure 1.Ternary phase diagrams of W/O microemulsion systems
a) cosurfactant - Ethanol; b) cosurfactant – 1 Buthanol

According to phase diagrams and table 1, 1-buthanol as CoS is responsible for microemulsions obtaining as clear systems, with the largest amount of incorporated water.

Table 1.The composition of the W/O microemulsions

Microemulsion	Soybean oil (%wt)	Brij 58 (%wt)	Span 80 (%wt)	Cosurfactant (%wt)	Water (%wt)
ME	35	5	45	8	7
MB	30	5	45	12.5	7.5

ME – microemulsion prepared with ethanol
MB – microemulsion prepared with 1-buthanol

The results concerning particles size, shown in figure 2, relieve two points:

- a size decrease in size of microemulsion drops using 1-buthanol in comparison with those obtained using ethanol;
- the DS microemulsion drops are smaller than those without DS.

As a result of using 1-buthanol as CoS which lipophilic properties are certainly greater, the size of microemulsions drops obtained with 1- buthanol is smaller than those prepared with ethanol as CoS.

Furthermore, there are many solid microparticles of DS which are not dissolved yet in the oil phase and which are absorbed on O/W interface. Their presence is the explanation of DS microemulsion drop size which is smaller, as well as the fact that the microemulsion can be stabilized using the Pickering stabilization method.

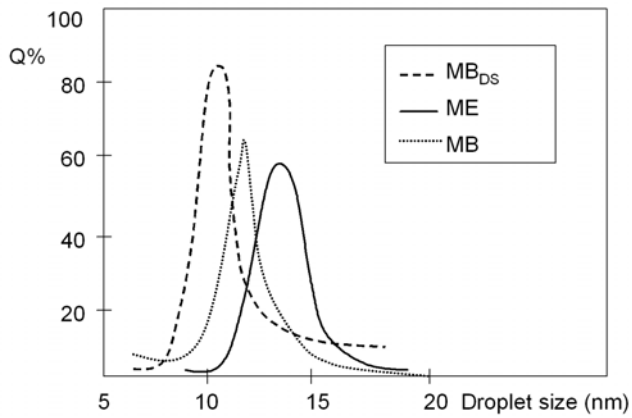

Figure 2. Particle size distributions of W/O microemulsions with/without DS
MB_{DS} – microemulsion with DS prepared with ethanol as cosurfactant
ME, MB – microemulsions without DS prepared with ethanol and 1-buthanol respectively

The electrical conductivity study of microemulsions shows a steady increase of conductivity with the water amount until a certain value; after that, the conductivity remains constant, which explains the stability of microemulsions (Fig. 3).

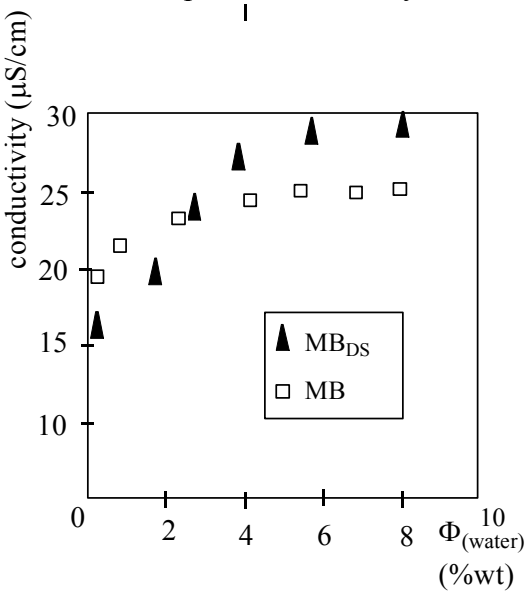

Figure 3
The conductivity of W/O microemulsions

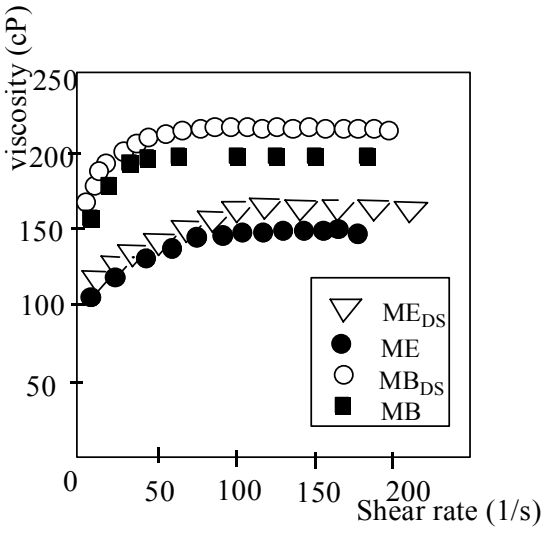

Figure 4
The viscosity of W/O microemulsions

Furthermore, the results show an increase of microemulsions conductivity with DS rather than those without DS, grace to the ionic character of the drug.

Viscosity measurements were examined as a function of sheer rate and Newtonian fluid was observed both for drug loaded microemulsions and unloaded microemulsions (Fig.4).

The results show an increased viscosity at MB microemulsions in comparison with ME microemulsions and the same aspect at DS microemulsions in comparison with those microemulsions without DS.

These results can be explained by:

a) the decrease of drops size leads to an increase of viscosity;
b) the lipophilic properties of 1-buthanol that increase the oil phase viscosity.

The DS solubility in distilled water was determined and its value is 19.7 mg/mL, in accordance with the literature [8, 9].

The partition coefficient of DS in n-octanol/water and soybean oil/water systems can be calculated using this equation (1):

$$P = \frac{C_{DS-oil\ phaze}}{C_{DS-water}} \qquad (1)$$

Where: $C_{DS\ oil\ phase}$ is DS concentration in oil phase (g/L);

$C_{DS\ water\ phase}$ is the DS concentration in water (g/L).

It was obtained the following values: $P_{noctanol/water} = 5.5$ and $P_{soybeanoil/water} = 1.5$

There was 1g of microemulsion added in the donor compartment which was already containing 0.01g of DS. According to DS solubility, there were almost 10% of DS dissolved into the MB microemulsion aqueous phase and the rest of it is diffused in its oil phase.

DS across the synthetic membrane obeys Fick's first law (2):

$$J = D\frac{C_0 P}{h} \qquad (2)$$

where: J is the steady-state flux (mg/cm^2/h)
D is the diffusion coefficient (cm^2/h)
C_0 is the applied drug concentration (% w)
P is the soybean/water partition coefficient
h is the membrane thickness (μm)

Table 2 shows the permeation parameters of DS from both microemulsions and commercial gels with DS through synthetic membrane.

Table 2. Permeation parameters of DS from microemulsions and commercial gel through CMC synthetic membrane

Formulation	Flux J (mg/cm^2/h)	Partition coefficient P
ME_{DS}	$5{,}6\cdot10^{-2}$	$5{,}6\cdot10^{-3}$
MB_{DS}	$6{,}2\cdot10^{-2}$	$6{,}2\cdot10^{-3}$
MB_{DS} + farnesol	$6{,}8\cdot10^{-2}$	$6{,}8\cdot10^{-3}$
MB_{DS} + menthol	$6{,}6\cdot10^{-2}$	$6{,}6\cdot10^{-3}$
Commercial gel	$4{,}8\cdot10^{-2}$	-

Analyzing the results from table 2, we can conclude:

- the amount of DS that penetrates the synthetic membrane and is released from microemulsion is greater than the amount of DS which penetrates the same synthetic membrane but is released from commercial gels [9].
- the release and diffusion obtained more easily from MB microemulsions rather than from ME ones due to 1-buthanol which is acting as an enhancer.
- the amount of DS delivered in the presence of various terpenes, such as menthol, farnesol 1.5%, is two time bigger than the systems without terpenes(farnesol acts as an enhancer better than menthol) [10].

Conclusions

The results show the possibility of using microemulsions as transdermal drug delivery vehicles.

According to the results of this study, the release and the diffusion of DS from both microemulsions and commercial gels through a synthetic membrane establish the advantages of using microemulsions instead of commercial gels.

Acknowledgement

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