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PREPARATION AND EVALUATION OF OFLOXACIN MICROEMULSION GEL

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Abstract

Ofloxacin is a fluoroquinolone derivative used in topical products as an antibacterial agent. The objective of this work was to prepare and evaluate microemulsion (ME) systems with a view to increase percutaneous absorption of ofloxacin. Similar concentrations of ofloxacin were incorporated in a microemulsion base composed of isopropyl myristate, water and Tween 80: Propylene glycol in the ratio of 6:1 and 15:1. Pseudo-ternary phase diagrams were used to obtain the concentration ranges of the oil, surfactant (Tween-80) and co-surfactant (Propylene glycol) for microemulsion formation. Five different formulations were formulated with various amount of the oil, water and the mixture of surfactant and co-surfactant at the ratio of 15:1. The ME systems were evaluated by examining under cross-polarizing microscope, measuring of percent transmittance, pH measurement, determination of the specific gravity, assessment of rheological properties and *in vitro* permeability studies. Out of all the prepared formulations ME5 (Isopropyl myristate:amphiphilic mixture:water=26:55:19%w/v) was considered to be the best formulation. Hence the microemulsion system is also or one of the promising tool for percutaneous delivery of ofloxacin.

Key words: Microemulsions, Pseudo-ternary phase diagrams, Skin permeation, Percutaneous Delivery.

Introduction

Microemulsions (MEs) are clear, thermodynamically stable, optically isotropic systems. They are formed spontaneously upon mixing suitable oil, water, and an amphiphile blend (surfactants either alone or in combination with a cosurfactant^[1,2]). MEs offer advantages over traditional creams and lotions as topical drug delivery. They are used to solubilize drugs and to improve topical drug availability. They are able to increase the rate

and depth of moisturizing agents into skin. It has been suggested that MEs may dissolve the ordered structure of the stratum corneum lipids, leading to the loss of barrier properties of the skin^[3].

The transdermal delivery system enables controlled or sustained release of the active ingredients and an enhanced patient compliance^[4]. In this research, formulating topical microemulsions and *in vitro* permeation studies for ofloxacin was

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considered. The concept of microemulsion was first introduced by Hoar and Schulman in 1943 [5].

The existence of this theoretical structure of ME was later confirmed by use of various technologies, and we can today adopt the definition given by Attwood [6]. A microemulsion is a system of water, oil, and amphiphilic compounds (surfactant and co-surfactant) which is a transparent, single optically isotropic, and thermodynamically stable liquid. The main difference between emulsions and micro emulsions lies in the size and shape of the particles dispersed in the continuous phase [7].

Microemulsions are formed when (i) the interfacial tension at the oil/water interface is brought to a very low level and (ii) the interfacial layer is kept highly flexible and fluid. These two conditions are usually met by a careful and precise selection of the components and of their respective proportions, and by the use of a “co-surfactant” which brings flexibility to the oil/water interface. These conditions lead to a thermodynamically optimized structure, which is stable as opposed to conventional emulsions and does not require high input of energy (i.e. through agitation) to be prepared. The size of the particles in microemulsions is much smaller than the wavelength of visible light, and are transparent and their structure cannot be observed through an optical microscope.

Microemulsions have attracted increasing attention as potential drug delivery systems, either as vehicles for topical applications or as bioavailability enhancers for poorly water

soluble active pharmaceutical ingredients (API) because of their unique solubilization properties.

In topical formulations, microemulsions have been proved to increase the cutaneous absorption of both lipophilic and hydrophilic API's when compared to conventional vehicles (emulsions, pure oils, aqueous solutions, etc.). The role of penetration enhancers played by the amphiphilic components of the microemulsion and the internal mobility of the drug within the vehicle also contribute to the overall performance of microemulsions in dermal or transdermal drug delivery. [8]

Ofloxacin is a synthetic fluoroquinolone drug used as an antibacterial agent [9]. In the form of microemulsion produces controlled delivery of the drug.

1. After oral doses, peak plasma levels occur in 1 to 3 hours but in case of microemulsions the peak plasma level of drug occur in 7 h so therapy can be maintained.
2. By the formulation of the drug in the form of microemulsion the dose could be minimized.
3. Ofloxacin is poorly soluble in water so it shows better entrapment during the preparation of microemulsion. By considering the above points the ofloxacin might be a suitable candidate for the design of microemulsion.

Materials and methods

Materials

Ofloxacin was obtained as a gift sample from Waksman Selman pharmaceuticals Pvt. Ltd., India. Oleic acid, polyoxyethylene 20 sorbitan

mono-oleate (Tween 80), propylene glycol were purchased from Sigma labs ltd. All other reagents were of analytical grade.

Preparation of the Micro emulsion gel and Microemulsion Systems Containing Different Concentrations of Ofloxacin.

A microemulsion gel composed of isopropyl myristate, water and Tween 80: PG as surfactant/cosurfactant mixture (S/CoS) in ratio of 6:1 was prepared. Ofloxacin was added to the mixtures of oil, surfactant and co-surfactant with varying ratios as described in the table and then an appropriate amount of water was added to the mixture drop by drop with constant stirring on magnetic stirrer until the transparent region is obtained. Ofloxacin was mixed first with isopropyl myristate by a magnetic stirrer (Wheaton, Rc-2, Japan), then the S/CoS mixture was added, and the mixture was stirred till the drug completely dissolved, then water was added and stirring was continued for about further 10 min. Microemulsions containing Ofloxacin were obtained spontaneously on stirring the mixtures. The system was stored in a tightly closed glass container and left for 3 days to attain equilibrium before evaluation. All microemulsions were stored at 27^oc temperature .^[10]

Construction of pseudo-ternary phase diagrams

The pseudo-ternary phase diagrams were constructed using titration method to determine the microemulsion region and to detect the possibility of making microemulsions with different possible compositions of oil, surfactant/ co-surfactant, and water.

Construction of pseudo-ternary phase diagrams and microemulsion formulation

Phase diagrams were constructed to determine the microemulsion regions.

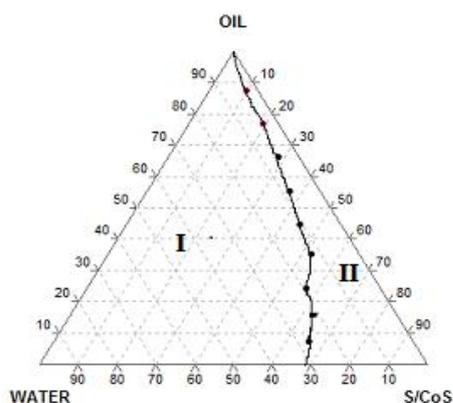


Figure 01: S/CoS (6:1)

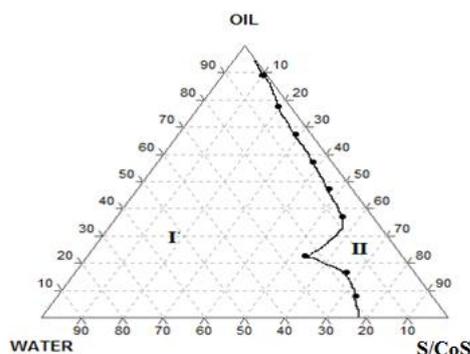


Figure 02: S/CoS (15:1)

Pseudo ternary phase diagrams

I-microemulsion formation zone

II-Non-microemulsion zone.

Preparation procedure

A microemulsion gel composed of isopropyl myristate, water and Tween80: propylene glycol as surfactant/cosurfactant mixture (S/CoS) in ratio 15:1 was prepared. Ofloxacin was added to the oil and amphiphilic mixture as described in the table1 and then an appropriate amount of water was added to the mixture drop by drop with constant stirring on magnetic stirrer until the transparent region is obtained. Microemulsions containing ofloxacin

were obtained spontaneously on stirring the mixtures. All microemulsions were stored at 27°C temperature.

Evaluation of the systems

The Microemulsion base and the five prepared ofloxacin ME systems were subjected to the following tests:

Visual Inspection

The systems were visually inspected for homogeneity, optical clarity, and fluidity.

Examination under Cross-polarizing

Microscope

The systems were examined under cross polarizing microscope HINDS PEM-ATC for the absence of birefringence to exclude liquid crystalline systems. ^[11, 12]

Limpidity Test (Percent Transmittance)

The limpidity of the systems was measured spectrophotometrically using spectrophotometer systronics UV Visible double beam spectrophotometer MODEL 2202. ^[13]

pH Measurements

The pH values of the samples were measured by using ELICO, LI 120, pH meter, INDIA. For Ofloxacin ME systems, it was measured by direct immersion of the electrode of the pH meter in the system. ^[14]

Determination of the Specific Gravity

The specific gravity of the systems was determined, at ambient conditions, using a specific gravity bottle of 10 ml capacity. ^[15]

Assessment of the Rheological Properties

The rheological properties of microemulsion systems were determined by using Brookfield digital Viscometer (LVDV II+CP). ^[16]

In vitro permeation study

Procedure

In this method microemulsion system is placed in the donor compartment of the diffusion cell. The microemulsion system faces the receptor compartment in which the 17.5ml of receptor fluid *i.e.*, buffer of pH 7.4 were placed. The agitation speed (50rpm) and temperature (37±2°C) are kept constant. The whole assembly is kept on magnetic stirrer and solution in the receiver compartment is constantly and continuously stirred throughout the experiment using magnetic beads. At predetermined time intervals, 2ml of the receptor fluid is removed for analysis and is replaced with an equal volume of fresh receptor fluid. The concentration of drug is determined spectrophotometrically at λ_{\max} of 254.0nm.

Result and discussion

Evaluation of the Systems

Visual inspection of the microemulsion showed homogeneous system of gel consistency. Examination under cross-polarizing microscope showed dark field indicating isotropic system. Assessment of rheological properties showed pseudo-plastic flow with thixotropic behaviour. The pH of micro-emulsion is slightly acidic in nature. Hence, formed product pH was slightly acidic in nature and they are regarded as microemulsions of Ofloxacin. The physical properties of the base are recorded in Table 2.

Table 01: Formulations of Ofloxacin microemulsion gel

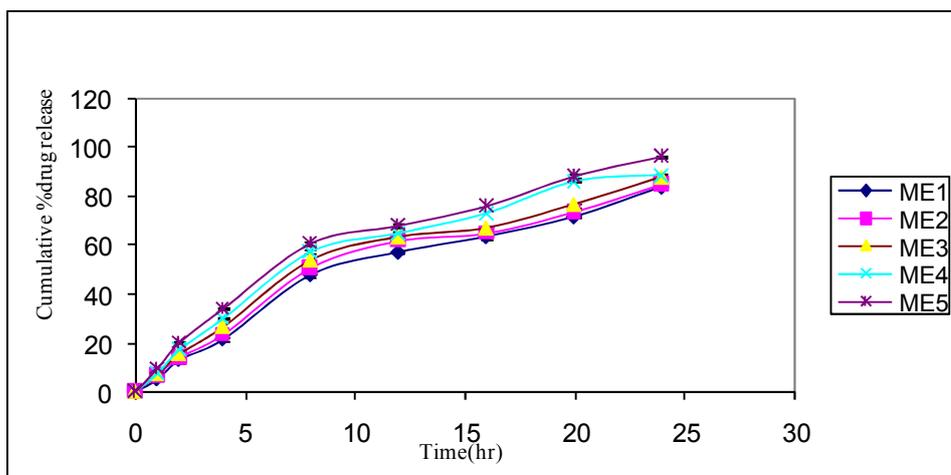
Formulation code	Isopropyl myristate (%w/v)	surfactant/cosurfactant ratio (15:1) (%w/v)	Water(%w/v)
ME1	22	55	23
ME2	23	55	22
ME3	24	55	21
ME4	25	55	20
ME5	26	55	19

Table 02: Physical Properties of Microemulsion Systems

Parameter	ME1	ME2	ME3	ME4	ME5
pH	5.92±0.34	5.96±0.55	5.95±0.47	5.94±0.78	5.9±0.56
Viscosity(CPS)	3045	2875	2590	2690	2480
Specific gravity	0.995	0.998	1.008	1.005	1.003
Percent Transmittance	76.88	79.43	81.23	83.45	84.12

Table 03: Diffusion profiles of Ofloxacin microemulsion

Time(hr)	Cumulative %drug releases ± SD				
	ME1	ME2	ME3	ME4	ME5
0	0	0	0	0	0
1	5.1±0.09	6.92±0.12	7.24±0.17	7.47±0.12	9.66±0.11
2	13.1±0.87	14.11±0.11	15.4±0.44	17.18±0.32	20.2±0.38
4	21.5±0.78	23.53±0.10	26.56±0.25	30.1±0.14	34.16±0.18
8	47.9±0.95	50.89±0.53	53.84±0.69	57.49±0.52	60.98±0.67
12	57.1±0.66	61.89±0.85	63.28±0.58	65.08±0.87	68.21±0.55
16	63.51±0.85	65.03±0.96	66.96±0.14	73.29±0.14	76.26±0.29
20	71.63±0.58	73.86±0.47	76.66±0.85	86.28±0.36	88.49±0.63
24	83.88±0.44	85.19±0.36	87.89±0.52	89.1±0.33	96.63±0.42

**Figure 03: Cumulative% release profiles of Ofloxacin microemulsion**

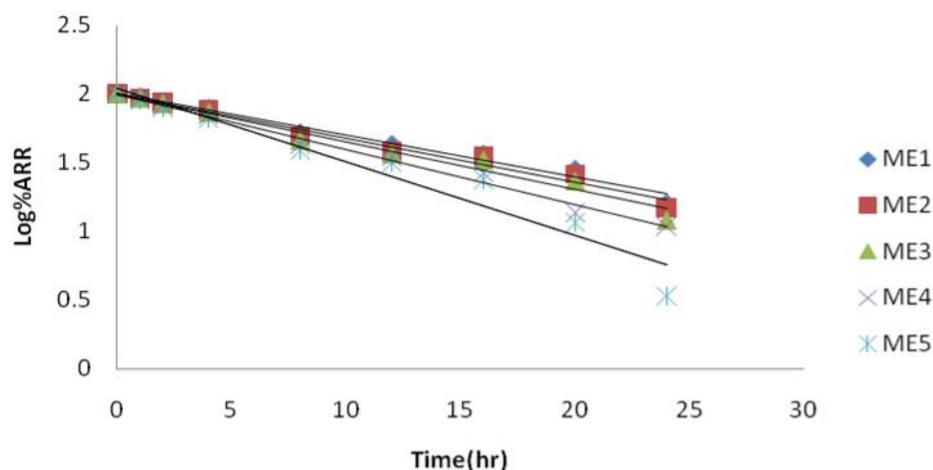


Figure 04: First order plots

Table 04: Release characteristics

Formulation code	First order equation	First order R ² value	K ₁ (hr ⁻¹)	t ₅₀ (hr)	t ₉₀ (hr)
ME1	Y=-0.030x+2.004	0.979	-0.06909	8.6	-
ME2	Y=-0.032x+1.996	0.978	-0.07369	7.8	-
ME3	Y=-0.034x+1.998	0.973	-0.07830	7.6	-
ME4	Y=-0.040x+2.000	0.988	-0.0921	6.6	-
ME5	Y=-0.053x+2.041	0.940	-0.12205	6.2	20.8

In vitro permeation study

The drug permeation rates from various microemulsion formulations are illustrated in Figure 3. The results were showed in table 3. Amongst the formulations tested, the batch ME-5 showed the highest permeation rate. The content of the surfactants mixture in microemulsions significantly affected the permeation rate of Ofloxacin.

By the results shown from the prepared formulations the ME-5 systems is having t₅₀ and t₉₀ more when compared with the other four formulations. Hence ME-5 is having better drug enhancing property.

Conclusion

For the formulation of microemulsions containing Ofloxacin, the proper components

and their optimum concentration ranges were obtained using pseudo ternary phase diagrams. In this study an attempt was made prepare microemulsion of ofloxacin for enhancing the drug permeation through the skin. By the visual inspection, percentage transmittance, t₅₀ and t₉₀ results suggested that micro-emulsion ME-5 is a better formulation. It could be a suitable vehicle for topical application of different concentrations of Ofloxacin. From this study it has been proved that the microemulsion technology enhances the topical delivery of ofloxacin.

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