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Iranian Association of Chemical Engineers, Unit 11, No. 13 (Block 3), Maad Building, Shahid Akbari Boulevard, Azadi Ave., Tehran - Iran.

Tel: +98 21 6604 2719 **Fax:** +98 21 6602 2196

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Transdermal Delivery of Desmopressin Acetate from Water-in-Oil Nano/Submicron Emulsion Systems

A. Soroushnia[1], F. Ganji[1*], S. M. Taghizadeh[2]

[1] Biomedical Engineering Group, Chemical Engineering Faculty, Tarbiat Modares University, Tehran, Iran
[2] Novel Drug Delivery Systems Department, Polymer Science Faculty, Iran Polymer and Petrochemical Institute, Tehran, Iran

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ABSTRACT

Desmopressin acetate is a potent synthetic peptide hormone that is administered via parenteral, intranasal and oral routes. A more acceptable route of administration with potentially good bioavailability could be offered by transdermal delivery. The present work reports on the development of water-in-oil (w/o) microemulsions for the transdermal administration of desmopressin acetate. A water-in-oil nano/submicron emulsion for transdermal administration of desmopressin was developed. Its skin penetration profiles were determined using Franz-diffusion cell. Pseudo-ternary phase diagrams for emulsion regions were constructed. Effects of hydrophilic-lipophilic balance (HLB), ratio of surfactants and co-surfactant mixture to oil phase (S_{mix}/oil) and ratio of surfactants to co-surfactant (S/Cs) on skin flux were also evaluated.

Skin flux was increased when S/Cs and S_{mix}/oil were decreased, and HLB was increased. Optimized formulation was obtained as: HLB=8, S/Cs=3 and S_{mix}/oil=5.4, with average particle size of 69 nm. The optimized nano/submicron emulsions increased desmopressin skin flux 4.45 fold relative to drug solution.

1. Introduction

Desmopressin acetate is a potent synthetic peptide hormone that is used chiefly to treat enuresis in young children as well as diabetes insipidus, hemophilia A and trauma-induced injuries. Desmopressin is administered via parenteral, intranasal and oral routes. When taken orally, the drug is destroyed in the gastrointestinal tract, and only 0.7–1 % of a given 100 or 200 μg dose may appear in the blood. The nasal application of desmopressin is accompanied by high variability in plasma pharmacokinetics. However, skin structure limits transdermal delivery only to lipophilic, low molecular weight potent drugs. The permeability of large hydrophilic molecules across the stratum corneum is extremely low [1, 2]. Microemulsions, which are thermodynamically stable mixtures of oil, water and surfactant, have great advantages in

*Corresponding author: fganji@modares.ac.ir

skin drug delivery [3-5]. First, a large amount of drug can be incorporated in the formulation due to their high solubilizing capacity, which increases thermodynamic activity towards the skin [6-8]. Second, the affinity of a drug to the interior phase of the microemulsion can be easily modified to favour partitioning into the stratum corneum using different compositions. Third, their ingredients may reduce the diffusional barrier of the stratum corneum and increase the permeation rate by acting as permeation enhancers. Also, the hydration effect of microemulsion on the stratum corneum may influence the permeation ability of formulations [9-12].

Although a sufficient number of studies have shown the potential of microemulsions in the transdermal delivery of drugs, little work has been done on how formulation components affect drug release and skin permeation in water-in-oil microemulsion containing desmopressin acetate. Getie et al. [13] compared the skin layer penetration profiles of desmopressin from a water-in-oil microemulsion and a conventional amphiphilic cream. They found that the drug can penetrate into deeper skin layers from microemulsions than from cream.

In this study, the effects of formulation components on the *in vitro* skin permeation of an emulsion drug delivery system containing nano-sized drops of desmopressin acetate were studied. The effect of surfactant hydrophilic-lipophilic balance (HLB), ratio of surfactants to co-surfactant (S/Cs), and ratio of surfactants and co-surfactant mixture to oil phase (S_{mix}/oil) were selected as independent variables and their effects on skin flux were evaluated by central composite design-response surface method. Response surface methodology (RSM) was used for experimental design, model development, evaluation of factors and optimization of conditions; central composite design (CCD) was used to make second order response surface models. The main objective of CCD is to determine the optimum operation conditions or an experimental design space that satisfies the operational specifications. Skin permeation studies, stability test and droplet size determination were performed to evaluate the advantages of transdermal emulsion delivery for desmopressin acetate.

2. Materials and methods

2.1. Materials

Desmopressin acetate was purchased from Biopharm, China; Tween80 and Span80 were purchased from Merck (Germany), and isopropyl myristate and 1-decanol were purchased from Sigma (USA). All other materials were HPLC grade.

2.2. Microemulsions preparation

Isopropyl myristate was used as the oil phase, mixture of Span80 and Tween80 as the surfactants, 1-decanol as the co-surfactant and distilled water as the aqueous phase. The microemulsion region was defined by constructing pseudo-ternary phase diagrams. Surfactants to co-surfactant ratios (S/Cs) were fixed between 2:1 and 5:1. These mixtures of surfactants and co-surfactant (S_{mix}) were mixed with the oil phase to give weight ratios of 9:1, 8:2, 7:3, 6:4, 5:5, 4:6, 3:7, 2:8 and 1:9. Water was added drop by drop and the mixture was stirred until a homogeneous dispersion/solution was obtained [14].

A central composite design (with $\alpha = 1.68$) using five levels of each of the three factors was designed to estimate the coefficient of a quadratic model (Table 1). The factors are HLB = 7 to 9, S_{mix}/oil = 5:5 to 7:3 and S/Cs = 2:1 to 5:1.

Table 1
Factors and their compositions for different runs in CCD (Desmopressin acetate 2 % w/w and Water 10.5 % w/w in all runs).

Run	HLB	Surfactants (w/w %)		S_{mix}/oil	Isopropyl myristate (w/w %)	S/Cs	1-decanol (w/w %)
		Span80	Tween80				
1	8.00 (0)	15.50	25.40	6.00/4.00 (0)	35.00	3.50(0)	11.60
2	8.59(+1)	11.30	23.20	5.41/4.59(-1)	40.00	2.61(-1)	13.00
3	7.41(-1)	18.40	23.30	6.59/3.41(+1)	29.80	2.61(-1)	16.00
4	8.00(0)	18.00	29.70	7.00/3.00(+1.68)	26.30	3.50 (0)	13.50
5	7.41(-1)	12.80	21.60	6.59/3.41(+1)	40.10	2.61(-1)	13.00
6	7.41(-1)	23.00	29.20	6.59/3.41(+1)	29.80	4.39(+1)	5.50
7	8.59(+1)	12.80	25.80	5.41/4.59(-1)	40.10	4.39(+1)	8.80
8	7.41(-1)	11.30	19.20	5.41/4.59(-1)	40.10	2.61(-1)	16.90
9	8.59(+1)	13.90	28.20	6.59/3.41(+1)	29.80	2.61(-1)	15.60
10	8.00(0)	15.50	25.40	6.00/4.00 (0)	35.00	3.50 (0)	11.60
11	8.00(0)	16.50	27.20	6.00/4.00 (0)	35.00	5.00(+1.68)	8.80
12	7.00(-1.68)	26.60	26.60	6.00/4.00 (0)	35.00	3.50 (0)	11.60
13	8.59(+1)	15.50	31.50	6.59/3.41(+1)	29.80	4.39(+1)	10.70
14	8.00 (0)	13.30	21.70	6.00/4.00 (0)	35.00	2.00(-1.68)	17.50
15	8.00 (0)	15.50	25.40	6.00/4.00 (0)	35.00	3.50 (0)	11.60
16	8.00 (0)	15.50	25.40	6.00/4.00 (0)	35.00	3.50 (0)	11.60
17	8.00 (0)	15.50	25.40	6.00/4.00 (0)	35.00	3.50 (0)	11.60
18	8.00 (0)	15.50	25.40	6.00/4.00 (0)	35.00	3.50 (0)	11.60
19	9.00(+1.68)	12.00	28.90	6.00/4.00 (0)	35.00	3.50 (0)	11.60
20	8.00 (0)	13.00	21.10	5.00/5.00(-1.68)	43.70	3.50 (0)	9.70

Desmopressin acetate was dissolved in distilled water, and the aqueous solution was added into the mixtures of oil, surfactants, and co-surfactant drop by drop with a varying component ratio as given in Table 1. Microemulsion formulations containing desmopressin acetate were obtained by stirring the mixtures.

For physical stability testing, centrifuge and heating–cooling cycle tests were done. Centrifuge tests (for 30 min at 10,000 rpm), were carried out to assess the physical stability of the microemulsions [15]. The heating–cooling cycle test was done by storing in the refrigerator at 4°C ±1°C for 48 h followed by hot air oven at 45°C ±1°C for 48 h as one cycle. Six cycles were performed. The clarity, phase separation, and precipitation of microemulsions were investigated.

For chemical stability test, accelerated stability studies were carried out on

microemulsions according to International Conference on Harmonization (ICH) guidelines. Sufficient replicates of the optimized microemulsion were kept at 40°C ± 2°C and 75 % ± 5 % RH. These were placed in a humidity chamber at 40°C ± 0.5°C and 75 % ± 5 % RH. Samples were withdrawn at 0 to 6 months. The concentrations of desmopressin acetate were determined by high-performance liquid chromatography (HPLC) method [16].

2.3. *In Vitro* skin penetration studies

The skin permeation of desmopressin acetate was determined using Male Sprague-Dawley rats, (150–170 g), obtained from Tarbiat Modares University. They were kept under standard laboratory conditions in 12 hours light/dark cycle at 25 ± 2°C and were nourished with a pellet diet and water *ad libitum*. The animals were received after the study was duly approved by the University Animal Ethics Committee. Animals were sacrificed using chloroform. Hair on the abdominal region (200-400 μm) was carefully removed, and a 5 cm × 5 cm patch of full-thickness skin was excised. The dermis side was wiped with isopropyl alcohol to remove any residual adhering fat. The skin was dipped and soaked in normal saline solution, washed with distilled water. The skins were clamped between the donor and the receptor chambers of the Franz-diffusion cells, with an effective diameter of 4.9 cm^2 mm and a receptor volume of 16 ml, filled with sodium phosphate buffer (pH 7.4) at 37±0.5°C. In all experiments, 240 mg of each formulation was placed on the membrane. 1.5 ml of the receptor phase was withdrawn at predetermined intervals from 0.5 h to 8 h, replaced by fresh buffer solution, and HPLC assayed (Younglin, SDV30) with a UV detector at 207 nm. The HPLC separation system consisted of a Perfect Sil Target (column 150×4.6 mm id, 5 μm) equipped with a guard column. The mobile phase consisted of MeCN: K_2HPO_4 10 mM, at (80:20), which was adjusted to a pH of 6.0 ± 0.1 with the addition of H_3PO_4. The flow rate of the mobile phase was 1 mL per minute. To prepare the standard curve, solutions of 0.5, 1, 2, 3, 4, 5, 6, 8 and 10 μg/mL of desmopressin acetate were prepared by spiking materials at these nine concentrations to draw a linear calibration curve (R^2=0.9998). The specificity for assay was established using 3 sequential replicates of the solution which was used in standard curve [17].

2.4. Statistical evaluation and formulation optimization

Design-Expert 7.0.0 was used for optimization study. Eight (2^3) factorial designs with six (2×3) axial points and six central points were selected. The following second-order polynomial empirical model explains the behavior of the system:

$$Y = \beta + \sum_{i=1}^{k} \beta_i X_i + \sum_{i=1}^{k} \sum_{j=1}^{k} \beta_{ij} X_i X_j \qquad (1)$$

where, Y is the measured response associated with each factor level combination response, β is an intercept representing the arithmetic average of all quantitative values obtained by 20 runs, β_i and β_{ij} are the model parameters, X_i and X_j are the coded factors and k is the number of factors under study. In this equation for three factors, β_1 to β_3 are the linear interaction coefficients, β_{11} to β_{33} are the quadratic interaction coefficients, β_{12} and β_{13} are the second-order interaction coefficients [18].

Design-Expert 7.0.0 software (Version

7.0.0, Stat-Ease Inc., Minneapolis, MN, USA) was used for data processing with Eq. (1) and the interaction between process variables and response was evaluated using analysis of variance (ANOVA). The coefficient of determination (R^2) expressed the quality of fit of the polynomial model and the F test in the same program was used to check its statistical significance.

Skin flux as a response was optimized numerically. Maximization for flux was selected in the same order to obtain an optimized formulation. One optimum check point was selected according to its desirability. The formulation was prepared for experimental evaluation. The optimized formulation predicted by the model was prepared experimentally and the response was determined. The results predicted by the model and the ones obtained experimentally were compared at a 95 % confidence level.

2.5. Characterization of optimized formulation

The average droplet size and polydispersity index of the microemulsions were checked at 25˚C by a dynamic light scattering zeta sizer instrument (Malvern, ZEN3600 UK).

3. Results and discussion

3.1. Microemulsions preparation

To obtain the appropriate components and their concentration ranges for the microemulsions, pseudo-ternary phase diagrams were constructed, so that w/o clear microemulsion regions could be identified (dark areas in Fig. 1). No distinct conversion from water-in-oil (w/o) to oil-in-water (o/w) microemulsion was observed. The rest of the region on the phase diagram represents the turbid and conventional emulsions. It is clear that, with an increase in the weight ratio of S/Cs and HLB, the microemulsion region is expanded. This behavior could be assigned to the interfacial tension reduction and the interface fluidity increase and thereby the entropy of the system is increased.

Visual observations revealed no evidence of phase separation, flocculation or precipitation. No sign of phase separation under stress was observed after centrifugation at 10000 rpm for 30 min and after six heating–cooling cycles. Chemical stability testing under accelerated conditions (40°C) for 6 months showed that desmopressin acetate content did not decrease in microemulsions.

3.2. *In Vitro* skin penetration study

The skin fluxes of desmopressin acetate in 20 experimental formulations were determined in Table 2.

The following reduced quadratic model explained the skin flux of the microemulsions:

$$\textbf{\textit{Skin flux}} = -1315.54+212.00\,(HLB)+156.88\,(S_{mix}/oil)+14.64\,(S/Cs)-10.77\,(HLB)^2-9.38\,(S_{mix}/oil)^2-3.47\,(S/Cs)^2 \quad (2)$$

The values of F tests for ANOVA analysis indicated that the model described above was statistically significant ($P<0.0001$). There was no lack of fit for this model. The adjusted R^2 was 0.94 and the prediction R^2 was 0.88. Table 3 indicates that HLB, S_{mix}/oil, S/Cs, HLB^2, S_{mix}/oil^2, and S/Cs^2 are the significant terms of the model ($p<0.05$).

In other words, HLB, S_{mix}/oil and S/Cs had significant impacts on skin flux and by increasing the amounts of HLB and decreasing S_{mix}/oil and S/Cs, skin flux increased.

Figure 2 represents predicted versus actual values of microemulsion skin fluxes. The clustering of points around the diagonal line

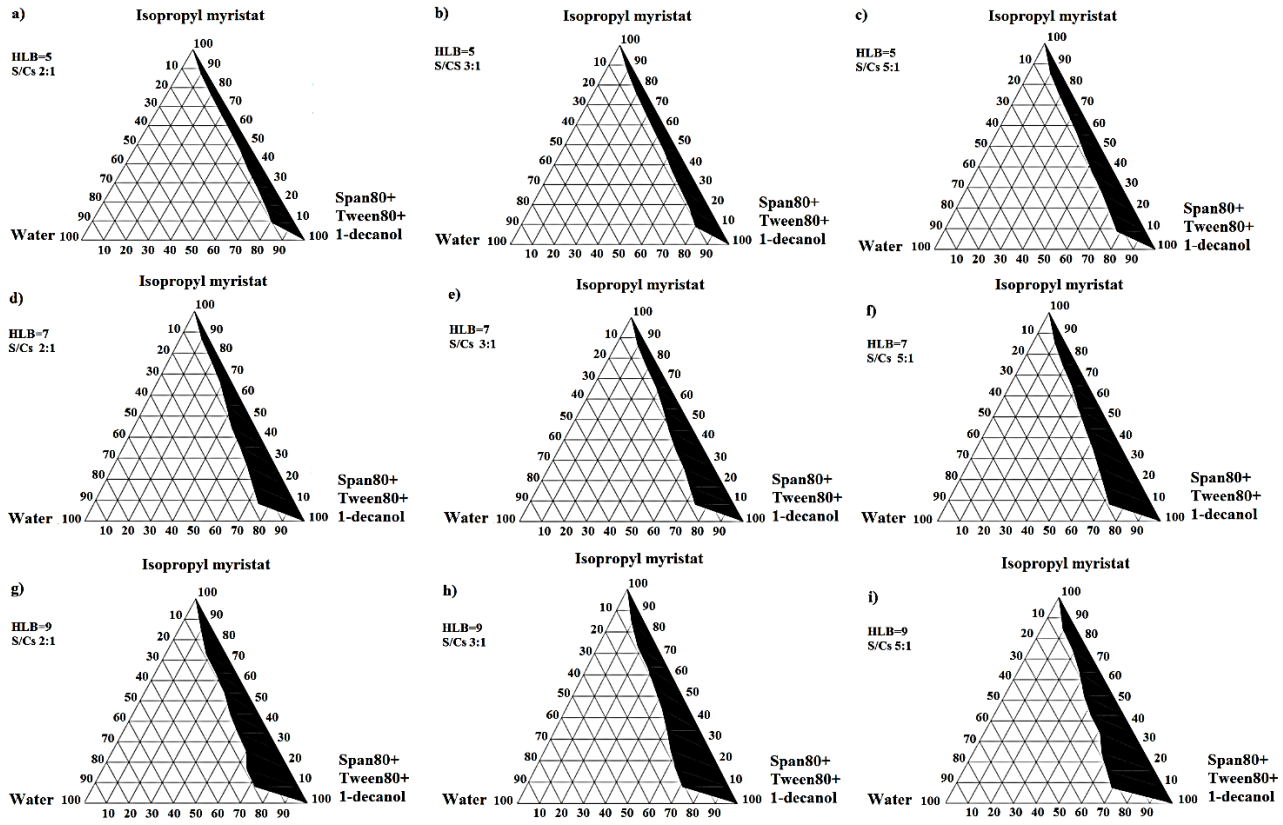

Figure 1. Phase behavior of microemulsions in: a) HLB=5, b) HLB=7 and c) HLB=9.

Table 2

Skin flux and lag time for different formulations in CCD.

Formulation	Skin flux (μg/(cm^2.h))	Lag time (h)
1	10.70±0.66	0.36±0.03
2	11.80±0.44	0.26±0.05
3	7.76±0.36	1.08±0.07
4	6.84±0.65	2.20±0.11
5	6.76±0.19	1.70±0.09
6	4.17±0.23	1.70±0.12
7	10.70±0.63	0.40±0.11
8	7.78±0.21	1.22±0.10
9	9.76±0.27	0.74±0.06
10	10.91±0.66	0.36±0.03
11	7.44±0.53	1.35±0.14
12	5.86±0.19	1.55±0.09
13	7.17±0.33	1.32±0.09
14	11.35±0.20	1.16±0.08
15	10.10±0.66	0.37±0.03
16	9.82±0.66	0.40±0.03
17	10.27±0.66	0.35±0.03
18	10.77±0.66	0.35±0.03
19	11.74±0.42	0.10±0.01
20	11.32±0.36	0.23±0.02

Table 3

Statistical significance test of skin flux for terms within the frame of generalized quadratic model: (a) before reducing and (b) after reducing the insignificant terms.

Terms in model	F value	P value
Before reducing		
A	110.04	< 0.0001
B	51.96	< 0.0001
C	46.71	< 0.0001
AB	3.15	0.1062
AC	0.30	0.5932
BC	5.97	0.0346
A^2	22.98	0.0007
B^2	17.20	0.0020
C^2	11.73	0.0065
After reducing		
A	73.63	< 0.0001
B	34.77	< 0.0001
C	31.25	< 0.0001
A^2	15.37	0.0018
B^2	11.51	0.0048
C^2	7.85	0.0150

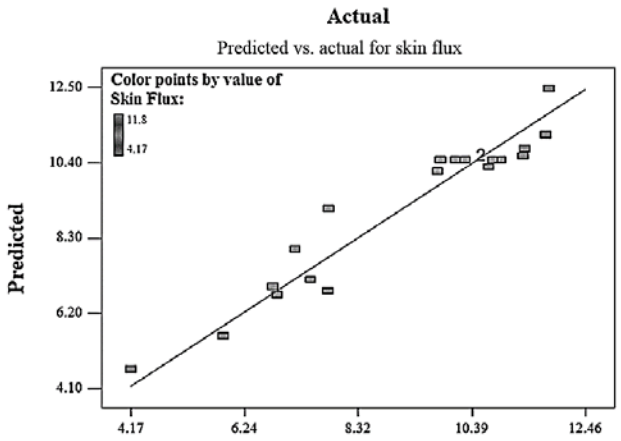

Figure 2. Predicted versus actual values for skin flux.

indicates a satisfactory correlation between the experimental data and the predicted values, confirming the robustness of the model.

The response surfaces and contour plots in Fig. 3 show the effects of HLB, S/C and S_{mix}/oil on microemulsion skin fluxes. It is evident that a higher degree of HLB results in higher skin flux due to the greater amount of Tween80 in the formulation (Fig. 3).

Skin barrier functions could be affected by non-ionic surfactants through different mechanisms. Tween80 is a polar component that enhances penetration by allowing the polar molecule to partition across the stratum corneum more easily. This modifies the composition of the membrane and favors permeation of the hydrophilic compounds [19], while Span80 affects the intercellular lipids of the stratum corneum by enhancing their fluidity. Therefore, the diffusion of lipophilic mixtures across the stratum corneum is enhanced.

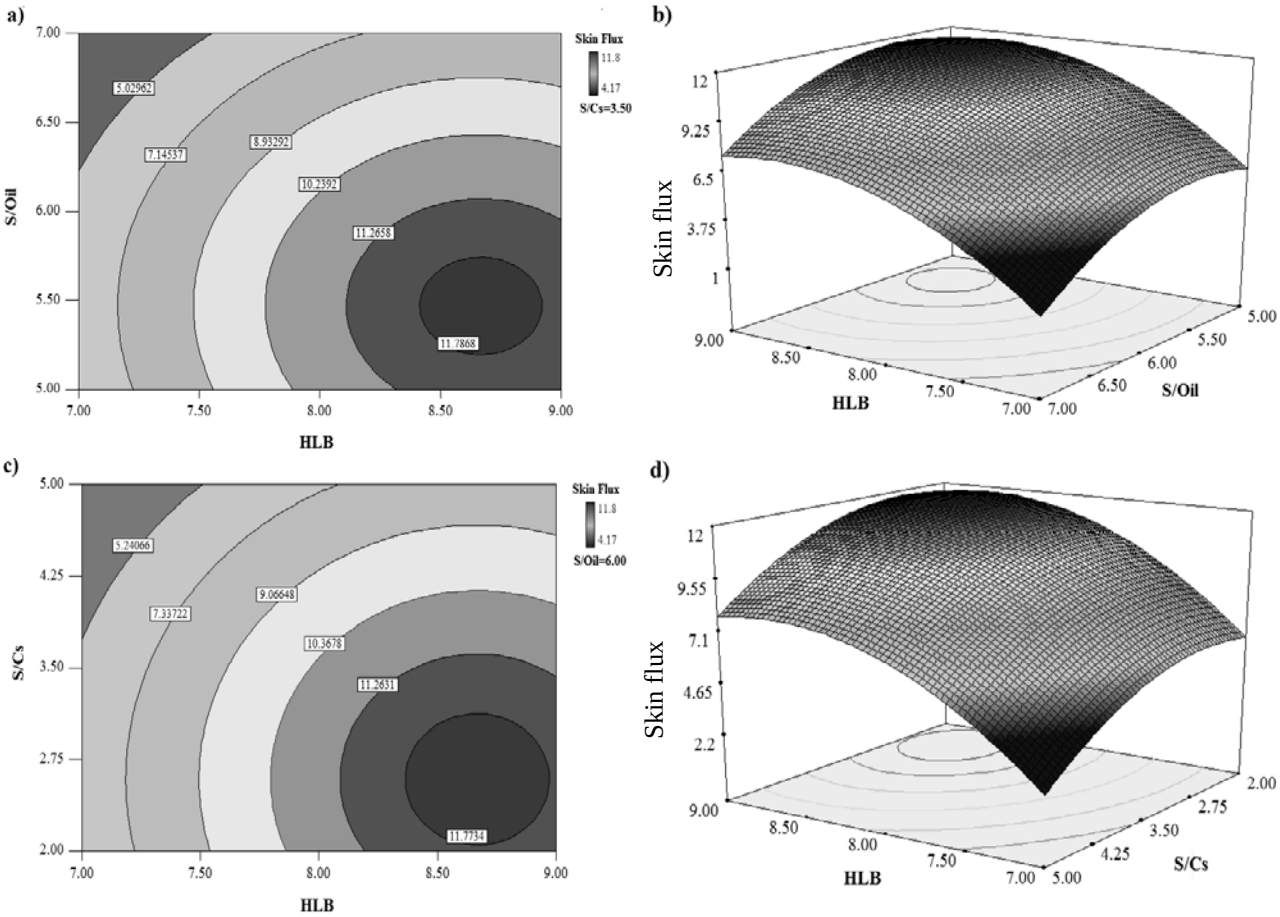

Figure 3. 2D contour plots (a & c) and 3D response surfaces (b & d) of skin flux.

Increasing the ratio of S/Cs results in a decrease of skin flux (Fig. 3 (a, b)). Higher Cs could lower the interfacial tension of the surfactant in microemulsions and make the interfacial layer more flexible and dynamic. Drug can diffuse across the flexible film between the internal and external phases; therefore, partitioning and diffusion into the stratum corneum increase. Moreover, the long chain hydrocarbon of hydrophobic alcohol structures can disrupt two layers of skin and facilitate penetration of drugs across the barrier.

According to Fig.3 (c, d) reducing the S_{mix}/oil ratio causes the skin flux to increase. Regarding the hydrophobic ester terminal of the stratum corneum, fatty acids can enter this dual structure and create a new region to penetrate. Isopropyl myristate is an aliphatic ester and its mechanism is related to its moderate polarity [20]. It affects polar and non-polar regions of the skin and has a permeation enhancement effect [21]. Moreover, isopropyl myristate reduces the viscosity of the formulation, providing the drug with more freedom and mobility.

3.3. Characterization of optimized formulation

The optimized formulations as well as the upper and lower levels of confidence of response evaluated by the software are shown in Table 4.

The drops of this formulation have a spherical shape with an average particle size of 69 nm and polydispersity index of 0.27 (Fig. 4). To test the validity of the optimized conditions given by the model, experiments were carried out using the optimal conditions determined by the parameters. It was observed that the experimentally obtained and the predicted responses were closely related; therefore, the model was successful in predicting the response.

Table 4
Model-predicted and observed values of skin flux in optimum condition.

Optimized Formulation				
HLB	S_{mix}/oil	S/Cs	Desirability	
8	5.46:4.54	3:1	0.985	
Response (Skin flux; µg/(cm^2.h))				
Prediction	SE Mean	95 % Cl Low	95 % Cl High	Observed Value
11.30	0.29	10.65	11.95	11.12

In order to evaluate the skin permeation of desmopressin acetate from optimized formulation versus drug solution, aqueous drug solution (4.8 mg desmopressin acetate in 240 µl water; the same drug concentration as in the optimized microemulsion formulation) was prepared, and its corresponding skin flux was evaluated. As shown in Fig. 5, it was observed that the skin flux of desmopressin acetate from optimized microemulsion was 4.45 fold higher than the above-mentioned drug solution.

4. Conclusions

This research investigated the effects of formulation components on the *in vitro* skin permeation of a microemulsion drug delivery system containing desmopressin acetate.

It was concluded that percutaneous absorption of desmopressin acetate from the microemulsion was enhanced by a decrease in S/Cs and S_{mix}/oil, and the higher level of HLB increased skin flux. The optimized formulation (HLB = 8, S/Cs = 3 and S_{mix}/oil = 5.4), with average particle size of 69 nm and good stability for a period of 6 months, could increase the skin flux of desmopressin acetate 4.45 fold relative to the drug solution. Skin permeation studies showed that developed

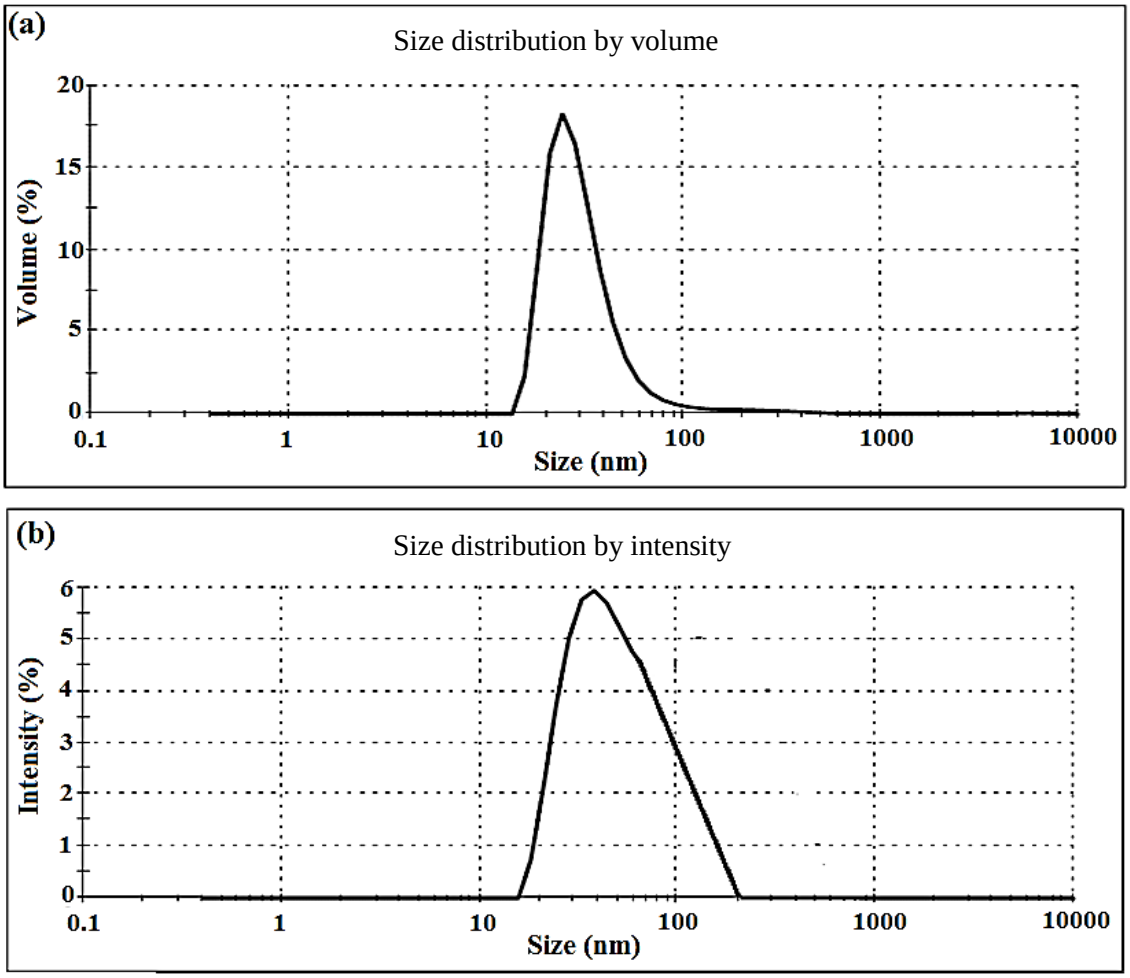

Figure 4. Average particle size of microemulsion by a) volume and b) intensity.

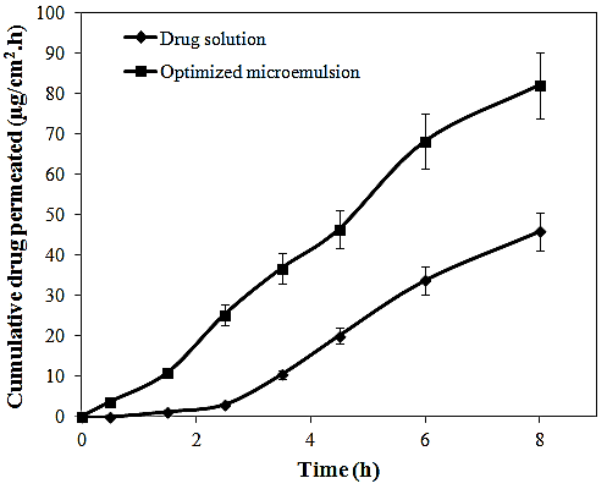

Figure 5. Cumulative skin permeation from ♦: drug solution and ■: optimized microemulsion.

desmopressin acetate microemulsion has better efficiency than the simple drug solution.

References

[1] Gaur, P.K., Mishra, S., Purohit S. and Dave K., "Transdermal drug delivery systems: A review", *Asian Pharm. Clin. Res.*, **2**, 14 (2009).

[2] Prausnitz, M.R. and Langer, R., "Transdermal drug delivery", *NBT.*, **26**, 1261 (2008).

[3] Fang, J.Y., Wu, P.C., Fang, C.L. and Chen, CH., "Intravesical delivery of 5-aminolevulinic acid from water-in-oil nano/submicronemulsion systems", *J. Pharm. Sci.*, **99**, 2375 (2010).

[4] Fouad, Sh.A., Basalious, E.B., El-Nabarawi, M.A. and Tayel, S.A., "Microemulsion and poloxamer microemulsion-based gel for sustained

transdermal delivery of diclofenac epolamine using in-skin drug depot: *In vitro*/*in vivo* evaluation", *Int. J .Pharm.*, **453**, 569 (2013).

[5] Gupta, S., "Biocompatible microemulsion systems for drug encapsulation and delivery", *Curr. Sci.*, **101**, 174 (2011).

[6] Al Abood, R.M., Talegaonkar, S., Tariq, M. and Ahmad, F.J., " Microemulsion as a tool for the transdermal delivery of ondansetron for the treatment of chemotherapy induced nausea and vomiting", *Colloid Surf. B.*, **101**, 143 (2013).

[7] Maghrabi, G.M.E., "Microemulsions as transdermal drug delivery systems", *Curr. Nanosci.*, **12**(6), 504 (2016).

[8] Mehta, D.P., "Microemulsions: A potential novel drug delivery system". *Int. J. Pharm. Sci*., **1**, 48 (2015).

[9] Hathout, R.M., Woodman, T.J., Mansour, S., Mortada, N.D., Geneidi, A.S. and Guy R.H., "Microemulsion formulations for the transdermal delivery of testosterone", *Eur. J. Pharm. Sci.*, **40**,188 (2010).

[10] Changez, M., "Effect of the composition of lecithin/n-propanol/isopropyl myristate/water microemulsions on barrier properties of mice skin for transdermal permeation of tetracaine hydrochloride", *Colloid Surf. B.*, **50**, 18 (2006).

[11] Jadhav, K.R., "Applications of microemulsion based drug delivery system", *Curr. Drug Deliv.*, **3**, 267 (2006).

[12] Kreilgaard, M., "Influence of microemulsions on cutaneous drug delivery", *Adv. Drug Deliv. Rev.*, **54**, 77 (2002).

[13] Getie, M., Wohlrab, J. and Neubert, H.H.R., "Dermal delivery of desmopressin acetate using colloidal carrier systems", *J. Pharm. Pharmacol.*, **57**, 423 (2005).

[14] Yang, J.H., Kim, Y.I. and Kim, K.M., "Preparation and evaluation of aceclofenac microemulsion for transdermal delivery system", *Arch. Pharmacal. Res.*, **25**, 534 (2002).

[15] Karasulu, H.Y., "Microemulsions as novel drug carriers: The formation, stability, applications and toxicity", *Expert Opin. Drug Deliv.*, **5**, 119 (2008).

[16] Panapisal, V., Charoensri, S. and Tantituvanont, A., "Formulation of microemulsion systems for dermal delivery of silymarin", *AAPS Pharm. Sci.Tech*., **13**(2), 389 (2012).

[17] Araújo, L.M.P.C., Lopez, R.F.V. and Thomazine, J.A., "Development of microemulsions to topically deliver 5-aminolevulinic acid in photodynamic therapy", *Eur. J. Pharm. Sci.*, **75**, 48 (2010).

[18] Taghizadeh, S.M., Soroushnia, A., Mirzadeh, H. and Barikani M., "Preparation and *in vitro* evaluation of a new fentanyl patch based on acrylic/silicone pressure-sensitive adhesive blends", *Drug Dev. Ind. Pharm*., **35**, 487 (2009).

[19] Kogan, A. and Garti, N., "Microemulsions as transdermal drug delivery vehicles", *Adv. Colloid Interface Sci.*, **123**, 369 (2006).

[20] Nahas, H., Fakhry, Gh., Ghamry, H. and Sabry, Sh., "Effect of various penetration enhancers concentrations on diclafenac sodium release from cellulose acetate phthalate polymeric film", *Asian J. Pharmacol*., **5** (1), 33 (2011).

[21] Tsai, Y.H., Fu, L.T., Huang, C.T. and Wu, P.C., "Formulation optimization of estradiol microemulsion using response surface methodology", *J. Pharm. Sci.*, **100**, 4383 (2011).