

SMEDDS of Olmesartan Medoxomil: Formulation, Development and Evaluation

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Abstract: - The objective of the work was to develop and evaluate self-microemulsifying drug delivery system (SMEDDS) for improving the delivery of BCS class II antihypertensive agent, Olmesartanmedoxomil (OLM). The solubility of OLM in oils, cosurfactants, and surfactants was evaluated to identify the components of the microemulsion. The pseudoternary phase diagram was plotted to identify the area of microemulsion existence at different Smix ratios (1:1, 2:1, 3:1) based on the results of solubility and emulsification studies. Smix ratio of 1:1 was selected for formulation of SMEDDS as the flowability of the formulation with Smix ratio of 1:1 was retained and it also gave a fairly larger microemulsion existence region. The *in vitro* drug release studies were carried out in phosphate buffer of pH 6.8 using USP XXIII dissolution testing apparatus type II at a paddle speed of 75 rpm. The optimized OLM liquid SMEDDS exhibited mean globule size of 59.91 nm, while experimental value was found to be 51.11 nm and polydispersity index of 0.42. The stability of OLM in SMEDDS was determined as per the International Conference on Harmonisation guidelines.

Keywords:- Olmesartanmedoxomil, SMEDDS, optimization, lyophilization

I. INTRODUCTION

Olmesartanmedoxomil (OLM) is a low dose, BCS Class II drug. It is an extensively used antihypertensive drug [1]. By the action of aryl esterases, situated in both intestine and plasma, it gets quickly de-esterified upon oral administration into an active metabolite, i.e., olmesartan [1]. However, the oral bioavailability (BA) of OLM is only 26% in healthy humans due to low solubility in gastrointestinal fluids and unfavorable breakage of the ester drug to a poorly permeable parent molecule in the gastrointestinal fluids. OLM is highly lipophilic with a log P value of 4.31, which attributes to its poor aqueous solubility.

BCS Class II, which has low solubility and high permeability require solubility enhancement as an integral part of the formulation strategies [2]. Thus, SMEDDS are beneficial since it is a simple process and the drugs are in a pre-dissolved state and the energy input associated with a solid-liquid phase transition is avoided, thus overcoming the slow dissolution process after oral intake [2].

Thus, OLM becomes an ideal candidate to formulate into SMEDDS to enhance the solubility and dissolution rate of the formulation, which may further increase the overall bioavailability of drug [3]. Thus, an attempt was made to formulate a SMEDDS formulation for oral drug delivery of

OLM and the liquid formulation was converted into solid for filling into capsule by lyophilization and adsorption onto a solid carrier.

II. MATERIALS AND METHODS

Materials

OLM was kindly provided by as a gift sample by Lupin Research Park, Pune, India. Capmul MCM C8 EP (Abitec corporation, USA), Capryol 90, Labrafac, Labrafil M 1944 CS, Transcutol HP (Gattefosse, France), Cremophor EL, Cremophor RH 40 (BASF, Germany) were received as a gift samples. Ethyl oleate, Isopropylmyristate, Isopropylpalmitate, Propylene glycol, ACN HPLC grade, orthophosphoric acid HPLC grade, methanol, toluene, ethyl acetate, glacial acetic acid (all AR grade) were purchased from S.D. Fine Chemicals, Mumbai, India. Tween 80 and oleic acid were purchased from Sigma Aldrich, Mumbai, India.

Solubility studies

The solubility of OLM was determined in different lipids (oils), cosurfactants and surfactants using shake flask method. The fixed volume (2 ml) of different oils was taken in 5 ml glass vials separately. An excess amount of drug was added to each vial, mixed using cyclone mixture and agitated on mechanical shaker for 72 h at room temperature. The vials containing mixture of drug and oil were centrifuged at 10,000 RPM for 30 minutes (Remi instruments, India) to separate the un-dissolved drug. The supernatant thus obtained was then filtered through membrane filter (0.45 μ). The filtrate was diluted suitably with methanol and saturation solubility of OLM (mg/ml) in oils was determined by recording absorbance using UV spectroscopy at 257 nm [4, 5].

HPLC Analysis of OLM

Highly sensitive analytical method is required for *in vitro* release and for stability studies. Therefore, a sensitive HPLC method for analysis was developed. The HPLC of Agilent Technologies 1200 series was used. It consisted of UV-VWD (Variable wavelength detector) equipped with Hypersil C18 (4.6 X 25 cm, 5 μ m) column. The mobile phase consisted of ACN: DDW with pH 3.5 (60:40) at a flow rate of 1 ml/min leading to retention time of 5.23 min. Peak area against concentration when subjected to linear regression showed a correlation coefficient of 0.9993 in the concentration range of

2-20 µg/ml. The method was validated with respect to accuracy and interday and intraday precision and sensitivity as per the International Conference on Harmonisation (ICH) guidelines [6].

Phase diagrams

Pseudo ternary phase diagrams were constructed for the selected combination of lipid (Capmul MCM C8 EP) and Smix (Cremophor RH 40 + Transcutol HP), at different Smix ratios. A series of oil/Smix mixtures were prepared at all nine combinations (1:9, 2:8, 3:7, 4:6, 5:5, 6:4, 7:3, 8:2 and 9:1). Pseudo-ternary phase diagrams were constructed for selected lipid and Smix at different Smix ratios (1:1, 2:1, 3:1). Pseudo-ternary phase diagrams were developed using the aqueous titration method. Slow titration with the aqueous phase was performed for each combination of oil and Smix separately to identify the microemulsion region. The total water consumed was noted and observations were made for phase clarity. The concentration of water at which turbidity-to-transparency and transparency-to-turbidity transitions occur were derived from the weight measurements. These values were used to determine the boundaries of the microemulsion region corresponding to the selected value of oil and Smix ratio [7].

Formulation of liquid SMEDDS

SMEDDS of OLM were prepared by microemulsion template method. In all the formulations the level of drug was kept constant at 1.66 %. Briefly, oil and Smix were accurately weighed into glass vials and the mixture was vortexed for 5 min on vortex mixer. The amount of SMEDDS should be such that it should solubilize the entire drug (single dose) completely. Hence the entire dose of 10 mg was added into the mixture of lipid and Smix. The mixture was mixed by gentle stirring on magnetic stirrer and heated at 40°C. The mixture was stirred until a clear, transparent solution was obtained. The homogenous SMEDDS were characterized for globule size, clarity (% transmittance) and number of inversions required to form clear microemulsion [8,9].

Optimization

A 3² randomized full factorial design was employed for carrying out the optimization of liquid OLM-SMEDDS. Based on the results obtained in preliminary studies, concentration of lipid [X1] and concentration of surfactant [X2] were found to be the major variables in determining the drug release (% DR) and mean globule size (MGS). So, these variables were selected as independent variables to obtain an optimized formula for maximum % DR and minimum MGS using 3² factorial design. The effect of these independent variables was investigated on two dependent variables, namely % DR [Y1] and MGS [Y2] to evaluate the responses. The optimization was done by using the Design-Expert® software (Version 10.0.4.0; Stat-Ease; Inc.; MN; USA).

Suitable mathematical models of the mixture design such as linear, quadratic, and special cubic models were analyzed by the software [10, 11, 12].

Characterization of optimized liquid OLM-SMEDDS (OLMopt)

Droplet size

Globule size (z-average diameter) and poly dispersity index (PDI) of the SMEDDS were measured by dynamic light scattering technique using NAOPHOX particle size analyzer (Sympatec, GmbH, Germany). Liquid SMEDDS was analyzed using Nanoprox software. All measurements were done in triplicate [8].

In vitro drug release study:

The SMEDDS formulation was put in to hard gelatin capsules 'size 0' and used for drug release studies as per following dissolution testing protocol. In vitro release profiles of SMEDDS of OLM were studied using USP XXIII dissolution testing apparatus type II at 37°C ± 2°C with rotating speed of 75rpm in dissolution medium of Phosphate Buffer pH 6.8. During the study, 5 ml volume of sample was removed at intervals of 5, 10, 15, 30, 45, 60 minutes. The sample removed was analyzed using UV spectrophotometer. By determining the amount of OLM released at various time intervals, the % release versus time graphs was plotted for optimized liquid SMEDDS.

Formulation of solidified OLM-SMEDDS

The optimized liquid SMEDDS formulation was solidified by two techniques, which are namely adsorption onto suitable carrier and freeze drying.

Solidification by Freeze drying

OLMopt SMEDDS was diluted with minimum quantity of deionized distilled water and thoroughly mixed with different solid carriers such as trehalose dehydrate, aerosil 200, microcrystalline cellulose (MCC), mannitol, maltodextrin. The resultant mixtures were allowed to stand for 15 min and consecutively freeze dried in a step-wise freeze-drying cycle using freeze dryer (Christ, Alpha 1-2 LD plus, Germany). Different percentages of cryo-protectant were tried such as 1%, 1.5% and 3%.

Solidification by Adsorption technique

Solidification was also carried out by adsorbing the OLMopt onto a suitable solid carrier. Solidification ability of carriers was tested by using different carriers such as aerosil 200, MCC, mannitol, maltodextrin, sucrose. The amounts of adsorbent tried were 100, 200, 300, 400, 500 and 600 mg.

Stability Studies[13]

Optimized batches of OLM-loaded SMEDDS-based capsules were subjected to stability testing as per conditions recommended by ICH Q1A (R2) guidelines for a period of 3 months.

The stability studies were conducted as per the condition recommended in ICH guidelines for a period of 3 months at the following storage conditions.

1. $30 \pm 2^\circ\text{C}$ / $65 \pm 5\%$ RH

2. $40 \pm 2^\circ\text{C}$ / $75 \pm 5\%$ RH

The samples were taken after 0 days, 30 days, 60 days and 90 days.

III. RESULTS AND DISCUSSIONS

Solubility studies

The results of solubility studies of OLM in various oils, cosurfactants, and surfactants are shown in Figs. 1, 2 and 3. Among various oils screened, it was clear that OLM has maximum solubility in Capmul MCM C8 EP as it accommodated maximum amount of drug which is greater as compared to other lipids. It was found that the combination of Capmul MCM C8 EP along with Smix of Cremophor RH 40 and Transcutol HP gave the best results for emulsification studies with the combination requiring minimum possible number of inversions and highest % transmittance even after 24 h. Thus this particular combination of lipid and Smix was used for further studies.

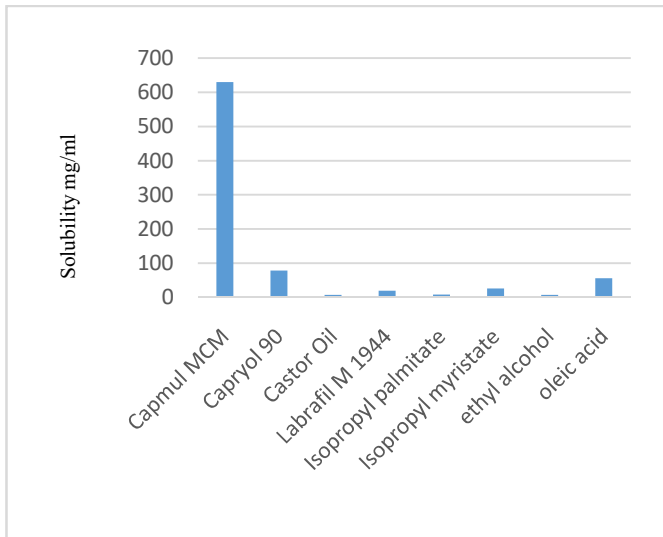


Figure 1: Solubility of OLM in various oils

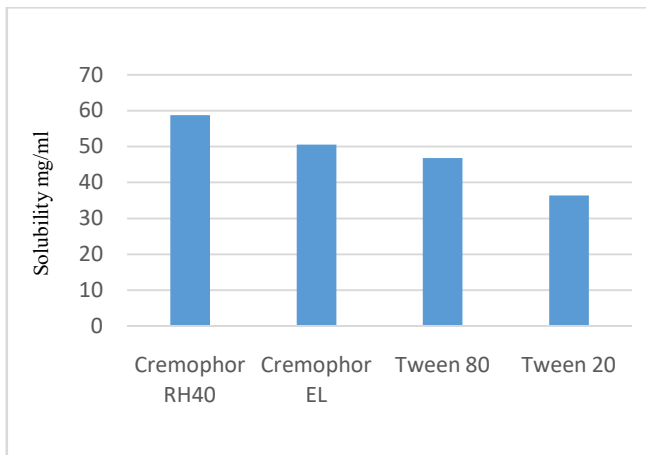


Figure 2: Solubility of OLM in various surfactants

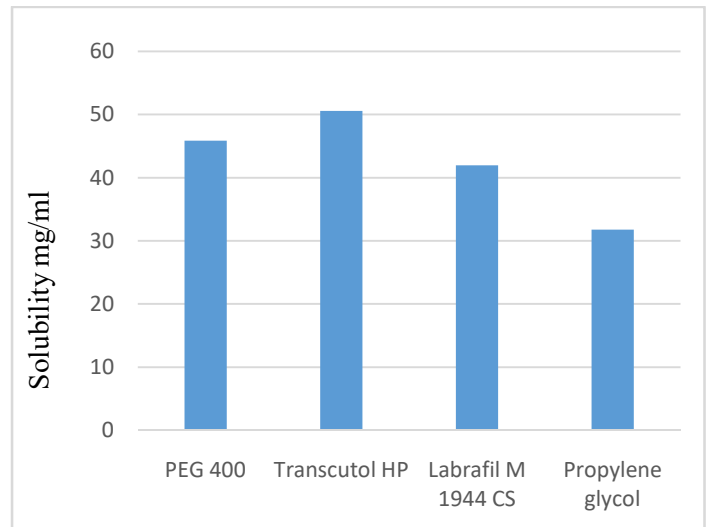


Figure 3: Solubility of OLM in various Co-surfactants

Phase diagram and Globule Size Analysis

Pseudo-ternary phase diagrams were constructed for selected lipid and Smix at different Smix ratios (1:1, 2:1, 3:1) based on the results of solubility and emulsification studies. In **Figure 4a** with Smix ratio of 1:1, the microemulsion existence region corresponds to 88%. Similarly, in **Figure 4b** with Smix ratio 2:1 and in **Figure 4c** with Smix ratio of 3:1, the microemulsion existence region was found to be 85% and 89% respectively. It was observed that there was no significant difference between the percentages of microemulsion existence region in three of the pseudo-ternary phase diagrams. However, it was seen during the aqueous titration that as the ratio of surfactant to co-surfactant (Smix ratio) was increased from 1:1 to 2:1 and 3:1, the mixture resulted in the loss of flowability. Thus, Smix ratio of 1:1 was selected for formulation of SMEDDS as the flowability of the formulation with Smix ratio of 1:1 was retained and it also gave a fairly larger microemulsion existence region. The value of mean globule size (MPS) for optimized OLM liquid SMEDDS as predicted by software was 59.91 nm, while experimental value was found to be 51.11 nm with polydispersity index 0.42.

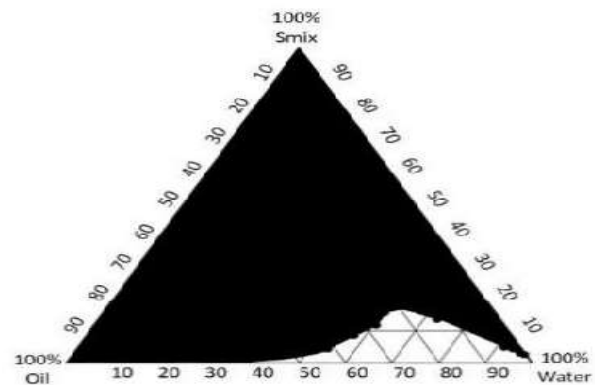


Figure 4a: Pseudo-ternary phase diagram for Smix ratio 1:1

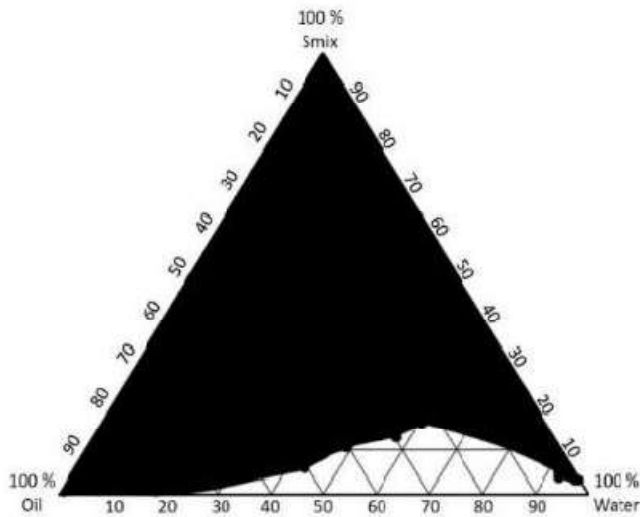


Figure 4b: Pseudo-ternary phase diagram for Smix ratio 2:1

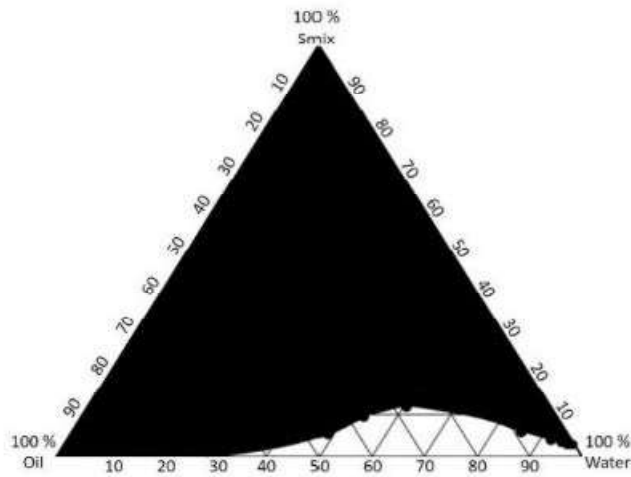


Figure 4c: Pseudo-ternary phase diagram for Smix ratio 3:1

Formulation of liquid SMEDDS

SMEDDS formulation were prepared using Capmul MCM C8 EP as oil, Cremophor RH 40 as surfactant and Transcutol HP as co-surfactant. 10 mg of the dose of drug was added in all batches. Based on the results of phase diagram, the ratio of Cremophor RH 40 to Transcutol HP was kept as 1:1. Batches which showed lowest number of inversions and gave lower globule were further characterized for % drug release.

Optimization of liquid SMEDDS

A 3² factorial design was used for optimization of liquid SMEDDS. The goal was set within the desired range, for % drug release and mean globule size as obtained with the levels selected as shown in **Table 1**. Two responses viz. Response-1 (% DR) and Response-2 (MGS) were recorded for the batches of SMEDDS and processed by Design-Expert software.

Table 1: Levels of independent variables for experimental run

Independent variables	Symbol	Levels		
		-1	0	1
Lipid concentration (mg)	X ₁	70	120	170
Surfactant concentration (mg)	X ₂	430	480	530

1) % Drug release (% DR):

The first step towards an optimal statistical analysis was to select the model that best fits the data. Results were analyzed by the sequential model comparison. Results of the sequential model comparison, which indicate whether a model could describe a response, are as given in **Table 2**.

Table 2: Sequential model comparison for Y1

Source	Model p-value	R-squared	Adjusted R-squared	Predicted R-squared	
Linear	0.000143	0.947647	0.930195	0.869185	Suggested
2FI	0.275638	0.95971	0.935536	0.823055	
Quadratic	0.078266	0.992628	0.980341	0.935607	
Cubic	0.852802	0.994639	0.957109	0.022881	Aliased

From the model comparison data as shown in **Table 2**, for this response linear model was found to be statistically significant with a p-value of 0.000143.

Contour and response surface plots:

Contour plot and three dimensional RSP showing the effect of concentration of lipid and Smix on % drug release is shown in **Figure 5 and Figure 6**. It is seen from the figures that as the concentration of Smix was increased, the % drug release decreased. However, as the concentration of lipid was increased, the % drug release increased. This could be attributed to the fact that more amount of lipid was needed to incorporate the entire dose of drug. This tendency could be attributed to limited water solubility and high lipophilicity.

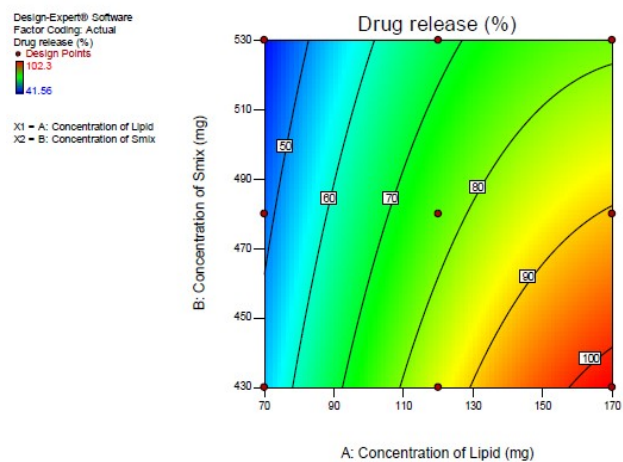


Figure 5: Contour plot showing effect of lipid and Smix concentration on % drug release

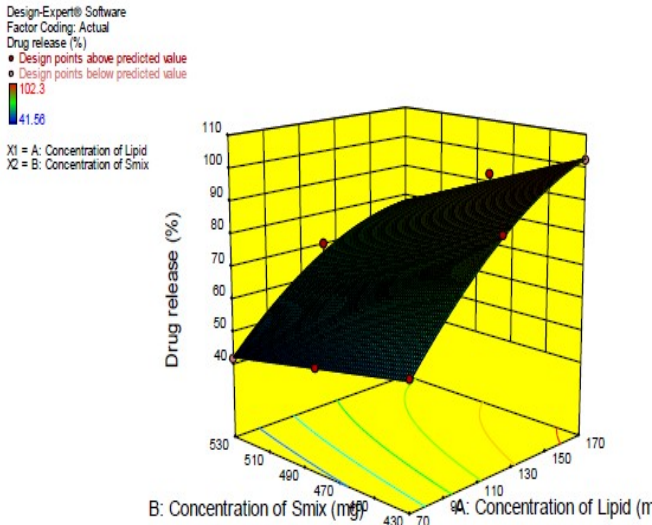


Figure 6: 3D response surface plot showing effect of lipid and Smix concentration on % drug release

2) Mean globule size (MGS):

Sequential model comparison shows that 2FI model was found to be statistically significant with a significant p-value of 0.03 as shown in Table 3.

Table 3: Sequential model comparison for Y2

Source	Std. Dev.	R-squared	Adjusted R-squared	Predicted R-squared	
Linear	50.7336	0.761434	0.681912	0.341581	
2FI	36.60912	0.896482	0.834372	0.621084	Suggested
Quadratic	38.73354	0.930472	0.814591	0.337124	
Cubic	50.16667	0.961123	0.688981	-6.0854	Aliased

From the model comparison data as shown in Table 3, for this response 2FI model was found to be statistically significant with a p-value of 0.621084.

Contour and response surface plots (RSP):

Contour plot and three dimensional RSP showing the effect of concentration of lipid and Smix on mean globule size is shown in Figure 7 and Figure 8. As the lipid concentration was increased, the globule size decreased. However, as the concentration of Smix was increased, the globule size decreased but until a limit was reached. After a particular point, further increase in the Smix concentration increased the mean globule size. This could be attributed to the fact that, too high of a concentration of Smix increases the penetration of water inside the oil droplets leading to ejection of oil droplets into the continuous aqueous phase and thus, the mean globule size increases.

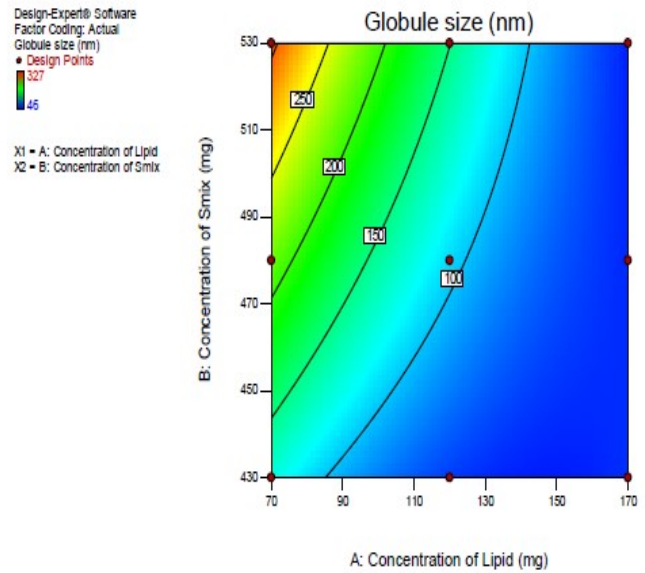


Figure 7: Contour plot showing effect of lipid and Smix concentration on globule size

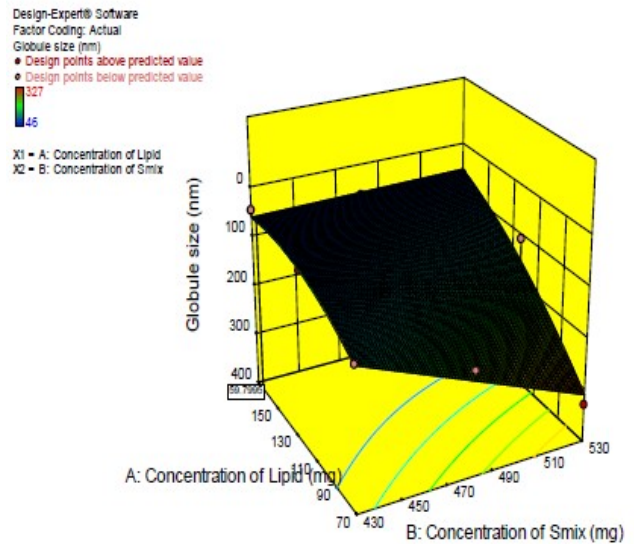


Figure 8: 3D response surface plot showing effect of lipid and Smix concentration on globule size

Overlay plot:

An overlay plot was used to determine quantity of both the independent variables i.e. concentration of lipid and concentration of Smix to obtain SMEDDS having % DR as close to 100 % as possible and mean globule size below 100 nm. Figure 9 shows the overlay plot of dependent variables i.e. the % DR (Y1) and mean globule size (Y2) at different values of independent variable i.e. lipid concentration (X1) and Smix concentration (X2). From the overlay plot, it can be concluded that optimum level of lipid and Smix favors the desired % DR and mean globule size of formulation.

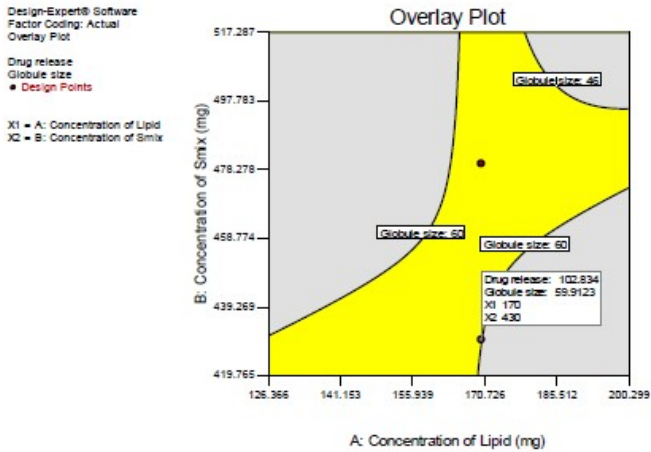


Figure 9: Overlay plot

In vitro drug release study

Figure 10 shows the % cumulative drug released v/s time (min). It can be clearly seen from the graph that more than 75 % of the drug released in 30 minutes of dissolution time. However, the total % cumulative drug release at the end of 60 min was found to be 99.40%.

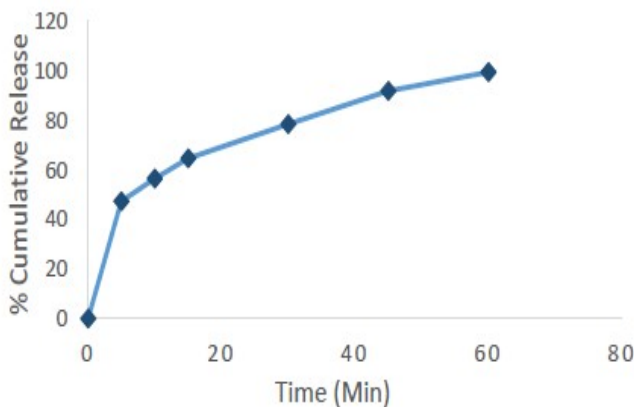


Figure 10: % cumulative drug released v/s time (min) of OLMopt

Formulation of solidified OLM-SMEDDS

1. Solidification by adsorption:

a) Selection of adsorbent:

OLMopt was added drop-wise onto solid carriers. Following results were obtained (Table 4).

Table 4: Selection of adsorbent

Name of adsorbent	Physical appearance	Flowability	Emulsification ability
Aerosil 200	White powder	Good	Good
MCC	White mass	Poor	-
Maltodextrin	White sticky mass	Very poor	-
Mannitol	White sticky mass	Very poor	-
Sucrose	White sticky mass	Very poor	-

Based on the above results, it can be seen that Aerosil 200 was able to form white powder that had good flowability and was able to emulsify when added to water. Thus Aerosil 200 was selected as the adsorbent for solidification by adsorption technique.

b) Selection of amount of Aerosil 200:

Different amounts of Aerosil 200 were tried to optimize the final amount of adsorbent required to form a free flowing powder of OLMopt. The selection of the amount was based on the appearance, emulsification time and globule size.

Solidification by Freeze drying:

a) Selection of adsorbent:

OLMopt batch was subjected to freeze drying. In order to obtain a free-flowing drug freeze dried powder. Equal concentration (1.5%) of different cryo-protectants such as Trehalosedihydrate, Aerosil 200, microcrystalline cellulose (MCC), Maltodextrin, Mannitol were tried and the adsorbent was selected based on the physical appearance, flowability and emulsification ability. Following results were obtained (Table 5).

Name of adsorbent	Physical appearance	Flowability	Emulsification ability
Aerosil 200	Very sticky mass	Very poor	-
MCC	Very sticky mass	Very poor	-
Maltodextrin	Slightly sticky	Poor	-
Mannitol	Sticky mass	Very poor	-
Trehalosedihydrate	White dry powder	Good	Good

Based on the above results, it can be seen that Trehalosedihydrate was able to form white powder that had good flowability and was able to emulsify. Thus, Trehalosedihydrate was selected as the adsorbent for solidification by Freeze drying technique.

b) Selection of concentration of Trehalosedihydrate:

Different concentrations of Trehalosedihydrate (1, 1.5, 3 and 5%) were tried to optimize the final amount of cryo-protectant required to form a free flowing freeze dried powder of OLMopt. The selection of the concentration was based on the appearance, emulsification time and globule size.

Stability

The results of stability studies of OLM SMEDDS are shown in Table 6 and Table 7. Initially, the cumulative drug release from solid SMEDDS formed by adsorption (Adr-OLMopt) and solid SMEDDS formed by freeze drying (FD-OLMopt) was found to be 98.90 % and 94.30 %, respectively, at the end of 60 minutes. Similar drug release profile was seen for the capsules of Adr-OLMopt and FD-OLMopt at the end of 30, 60 and 90 days as shown in the Table 6 and Table 7, respectively, at both the storage conditions.

Table 6: *In vitro* drug release of Adr-OLMopt based capsule subjected to stability study

Sample	Storage conditions	% Cumulative Drug release at 60min			
		0 day	30 day	60 day	90 day
Capsule of Adr-OLMopt	30°C/ 65% RH	98.90	98.83	98.79	98.78
	40°C/ 75% RH	98.90	98.85	98.81	98.77

Table 7: *In vitro* drug release of FD-OLMopt based capsule subjected to stability study

Sample	Storage conditions	% Cumulative Drug release at 60min			
		0 day	30 day	60 day	90 day
Capsule of FD-OLMopt	30°C/ 65% RH	94.30	94.22	94.10	93.98
	40°C/ 75% RH	94.30	94.28	94.19	94.11

Thus sample subjected to stability study at 30°C/ 65 %RH and 40°C/ 75 %RH were found to be stable with respect to *in vitro* drug release.

IV. CONCLUSION

This study attained a successful formulation of liquid SMEDDS, optimization and evaluation of liquid SMEDDS, solidification and characterization of optimized liquid SMEDDS by adsorption and freeze drying and both the formulations, Adr-OLMopt and FD-OLMopt, were found to be stable with respect to globule size and % drug release when subjected to stability conditions of 30°C/ 65 % RH and 40°C/ 75 % RH.

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