RESEARCH ARTICLE

Improving Lovastatin Absorption using Supersaturated-Self Nanoemulsifying Drug Delivery System (SNEDDS)

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ABSTRACT
Lovastatin is a cholesterol-lowering agent has low bioavailability. Therefore, Supersaturated Self Nanoemulsifying Drug Delivery System (super-SNEDDS) can be used to increase bioavailability of lovastatin. This research aims to provide the formula for supersaturated-SNEDDS lovastatin. The selection of super-SNEDDS components is based on the solubility of lovastatin in various oils, surfactants, and cosurfactants. A pseudo-ternary phase diagram was created to determine the lower and upper limits of the oil (X1), surfactant (X2), and cosurfactant (X3). Super-SNEDDS optimization was carried out using a D-Optimal Mixture Design with response values in the form of percent transmittance and emulsification time. The optimum formula was confirmed and characterized including size of droplet, polydispersity index, zeta potential, physical stability, and *in vitro* absorption test using the small intestine-inverted method. The results showed the optimum formula consists of lovastatin (5,5 mg/gram SNEDDS preconcentrate), 5% oleic acid, 80% Tween 80, and 15% PEG 400. The optimum formula for super-SNEDDS had a droplet size of 18.07 nm, a polydispersity index of 0.34, and a zeta potential value of -228.9 mV. Lovastatin super-SNEDDS preparation was physically stable based on the stability test of the preparation at a temperature of 40°C and after freeze-thaw stability test cycles. The *in vitro* oral absorption using everted gut sac method showed that the cumulative amount of lovastatin absorbed in the lovastatin super-SNEDDS preparation was 1.6-fold higher than in the lovastatin SNEDDS preparation and 3.3-fold higher than in pure lovastatin.
Keywords: everted gut sac, lovastatin, D-Optimal Mixture Design, supersaturated SNEDDS.
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INTRODUCTION

Lovastatin is a lipophilic cholesterol-lowering agent with a log-P value of 4.3 and has low solubility. Lovastatin is categorized in Biopharmaceutical Classification System (BCS) class II with slow drug release and incomplete absorption.[1] Lovastatin has low bioavailability (less than 5%), due to a fairly large first-pass effect.[2] Attempts have been carried out in previous research to increase the solubility of lovastatin have including the creation of a Self Nanoemulsifying Drug Delivery System (SNEDDS).[3,4] SNEDDS is a nano emulsion system made from an isotropic mixture of oil, surfactant, and cosurfactant with drug content which is capable of forming nanoemulsions spontaneously and has a transparent dosage appearance.[5] SNEDDS allows the large scale manufacture to make an easy and economical manufacturing process that becomes the main attraction in the industry and thermodynamically stable.[6] The globule size in the SNEDSS system is 20-200 nm when dispersed in water[7], so it can increase drug dissolution in the gastrointestinal tract, and improve drug bioavailability through absorption, and permeation. The SNEDDS can be used orally to accelerates drug onset therefore it can reduce the dose of drug administration and increase the maximum concentration value (C_{max}).[8] Previous research conducted by Hashem *et al*. showed that SNEDDS containing atorvastatin with components of oleic acid, Tween 80, and propylene glycol had higher C_{max} and lower T_{max} values compared to commercial atorvastatin tablets.[7] Previous research related to lovastatin SNEDDS was carried out by Nurhabibah *et al* (2022) where an optimal formula was produced with a composition of oleic acid 5.2%, Tween 80 79.5% and PEG 15.8% and a drug content of 97%.[4] In SNEDDS preparations, the drug concentration is below the equilibrium solubility (Seq) ranging from 50-90% which often requires large amounts of preparation to reach a therapeutic dose.[9] To overcome this problem, a supersaturated-SNEDDS preparation was made which contains a drug with a concentration higher than the solubility equilibrium. Supersaturated-SNEDDS contains drugs with supersaturated concentrations, namely 150% to 250% above the equilibrium solubility.[10] Supersaturated-SNEDDS has better performance in term of

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Table 1: Lovastatin supersaturated-SNEDDS formula design and response

Formula	Run	Oleic Acid (X1)	Tween 80 (X2)	PEG 400 (X3)	Transmittance in phosphate buffer pH 7,2 (%) (Y1)	Transmittance in Aquadest (%) (Y2)	Emulsification time in phosphate buffer pH (second) 7.2 (Y3)	Emulsification time in Aquadest (second) (Y4)
1	1	5	67,5	27,5	100,2	99,4	15,13	11,78
2	2	14	45	41	64,6	84,7	29,89	20,9
	5	14	45	41	67,7	84,6	24,81	20,34
3	3	6	49	45	93	96,9	15,31	14,12
4	4	10	61	29	69,7	89,5	26,29	17,29
	6	10	61	29	66,7	88,9	23,44	19,15
	15	10	61	29	68,5	91,3	24,24	20,40
	16	10	61	29	67,4	90,1	24,17	18,8
5	7	10	80	10	76	90,6	18,15	15,32
	10	10	80	10	74	89,8	23,43	22,99
6	8	5	57	38	100	97,7	15,36	12,69
7	9	5	73	22	100,4	99,5	14,36	11,56
8	11	5	79	16	100,6	99,5	14,73	13,07
9	12	15	68,5	16,5	50,9	85,6	16,78	19,74
10	13	15	62	23	53,1	84,7	17,47	15,50
11	14	15	52	33	64	84,6	23,69	18,22

reducing the dose required compared to conventional SNEDDS.[11] Thomas *et al*., (2013) developed simvastatin supersaturated-SNEDDS and it was found that there was an increase in relative bioavailability values of 1.8-fold higher than conventional SNEDDS Simvastatin.[12] Supersaturated-SNEDDS reduce the drug amount in dosage form, therefore improve the effectiveness of drug administration. To determine the optimal phase ratio of oil, surfactant, and cosurfactant in preparing supersaturated-SNEDDS, the formula was optimized using the D-Optimal Mixture Design. The advantage of using the D-Optimal Mixture Design application is that it reduces the results of formula optimization and can provide statistical information between variables and responses.[13] The optimum formula then was characterized including percent transmittance, polydispersity index, globule size, zeta potential, and drug content in the supersaturated-SNEDDS system. Apart from that, it can provide scientific data regarding the permeability of lovastatin to supersaturated-SNEDDS in biological membranes *in vitro* using everted gut sac method.

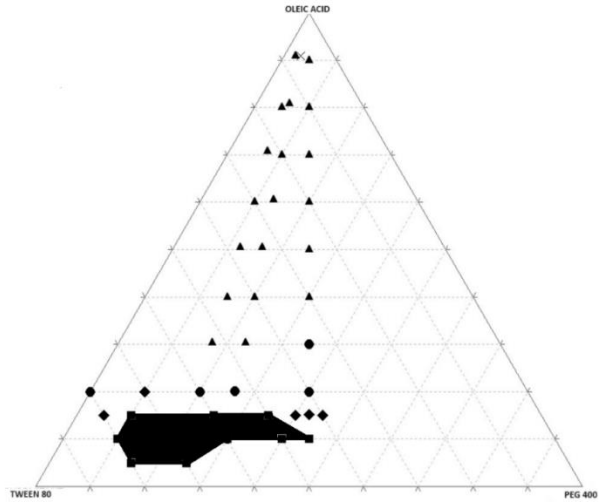

Figure 1: Pseudoternary phase diagram showed the emulsification area

MATERIALS AND METHODS

Materials

Lovastatin (Pharmaceutical grade) was obtained from PT. Sanbe Farma Indonesia, oleic acids, rice bran oil, virgin coconut oils, corn oil, palm oil, Tween 80, Tween 20, span 80, PEG 400, propylene glycol, and distilled water were provided in technical grade from PT. Citosa Kamila Prima. NaCl 0,9% (Otsuka®), phosphate buffer pH 7,2 (Neogen®), water for HPLC and acetonitrile (HPLC grade) was obtain from Merck®, filter paper PTFE 0,2 µm (Millipore, Billerica, MA, USA).

Selection of Oil, Surfactant, and Cosurfactant Phases

The selection of oil, surfactant, and cosurfactant phases was carried out by dissolving 50 mg of lovastatin in 5 mL of various oil phase (virgin coconut oil, oleic acid, corn oil, palm oil, rice bran oil), surfactant (Tween 80, span 80, Tween 20), cosurfactant (PEG 400 and propylene glycol).[14] Vials containing a mixture of solvent and lovastatin were placed in a shaking water bath (Memmert®) at a speed of 50 rpm for 24 hours at 37± 2ºC. The solvent was selected based on the ability of the excipient to dissolve lovastatin by observation of percent transmittance on 650 nm using a UV-Vis spectrophotometer (Shimadzu UV 1280®). The selected oil phase with the highest percent transmittance value was oleic acid with a percent transmittance value of 22.7 ± 0.24%, the surfactant used was Tween 80 with a percent transmittance value of 97.2 ± 0.39%, and the cosurfactant used was PEG 400 with a percent transmittance value of 83.2 ± 1.74%.

Determination of Equilibrium Solubility of Lovastatin in Oleic Acid

Determination of the solubility equilibrium was carried out by dissolving 50 mg of lovastatin in 5 grams of the oleic acid and placing it in a shaking water bath (Memmert®) at a speed of 50 rpm for 7 days at 37 ± 2ºC. On the 6th and 7th

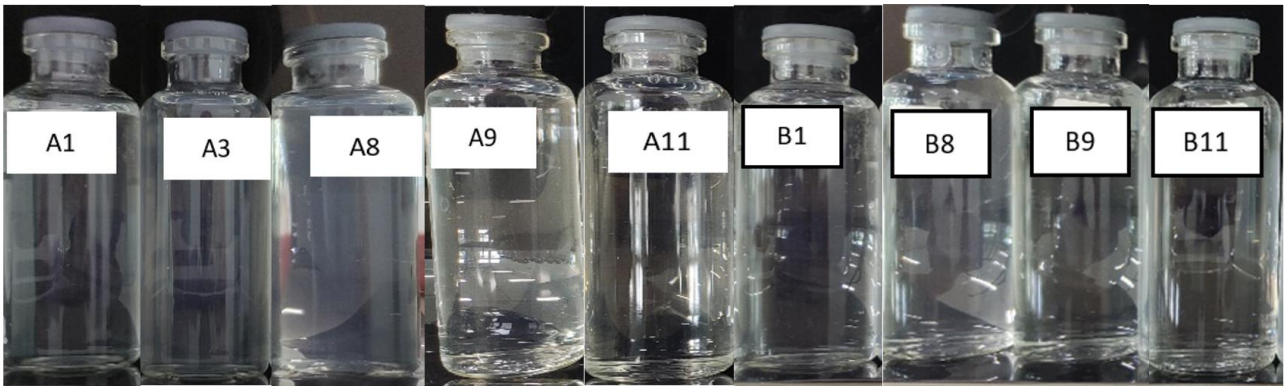

Figure 2: Nanoemulsions appearance of supersaturated-SNEDDS Lovastatin dispersed in water (A) and phosphate buffer (B). A clear visual was found in run A1, A3, A8, A9, A11, B1, B8, B9, B11

Table 2: Result of ANOVA dependent variable (response) design expert Lovastatin Supersaturated-SNEDDS

Response	Dispersed medium	P value	R2	Lack of fit
Percent Transmittance	Phophate buffer pH 7.2	<0,0001	0,9972	0,0839
	Water	<0,0001	0,9888	0,5917
Emulsification time	Phophate buffer pH 7.2	0,0001	0,8944	0,2701
	Water	0,006	0,7654	0,3264

days, samples were taken and the dissolved lovastatin levels were determined using HPLC (Waters e2695®) with Photodiode Array Detector (Waters 2998®) on 237 nm. The solution was assumed to be in a state of solubility equilibrium (Seq) when the concentration of lovastatin between days 6 and 7 has a difference of no more than 5%.[10]

Determination of Emulsification Efficiency using Pseudoternary Phase Diagrams

Pseudo-ternary phase diagram was plotted by mixing oleic acid, Tween 80, and PEG 400 with a total percentage of 100%. Oleic acid, Tween 80, and PEG 400 were mixed to form one clear phase called SNEDDS preconcentrate. The percent transmittance and emulsification time were evaluated for each preparation by dispersing the SNEDDS preconcentrate in distilled water with a ratio of 1:500. Percent transmittance was measured with a UV-Vis spectrophotometer (Shimadzu UV 1280®) at a wavelength of 650 nm.[15,6] Determining the emulsification time was carried out based on research conducted by Alothaid *et al*., (2021).[16] The emulsification efficiency assessment category is based on visual appearance, emulsification time, and percent transmittance based on the assessment categories from Devireddy and Jonnalagadda (2021).[17]

Optimization of Supersaturated-SNEDDS Lovastatin Formula using D-Optimal Mixture Design

Optimization of the supersaturated-SNEDDS formula was carried out using Design-Expert 13 software. The independent variables used were oleic acid, Tween 80, and PEG 400 concentrations. The concentration range obtained from the pseudo ternary phase diagram is 5 to 15% oleic acid, 45 to 80% Tween 80, and 10 to 45% PEG 400. There were 16 runs observed were the percent transmittance and emulsification time of lovastatin supersaturated-SNEDDS in phosphate buffer medium pH 7.2 and distilled water. The preparation of lovastatin supersaturated-SNEDDS was carried out by mixing the oleic acid, Tween 80, and PEG 400 according to the percentages in the Design-Expert. Lovastatin was added in an amount of 150% of the equilibrium solubility of lovastatin (5.5 mg/g preconcentrate). The SNEDDS preconcentrate to which lovastatin had been added was placed in an oven at 60ºC for 3 hours, then cooled overnight. The heating-cooling cycle was carried out 3 times to obtain a lovastatin supersaturated-SNEDDS preparation. Supersaturated-SNEDDS of lovastatin were then dispersed in phosphate buffer medium pH 7.2 and distilled water with a ratio of 1:500 at a temperature of 37± 2ºC. The responses observed were emulsification time and percentage of transmittance. Optimum formula was obtained by highest desirability value of 0.969, that is 5% oleic acid, 80% Tween 80, and 15% PEG 400. The optimum formula was evaluated on physical characterization, stability, and membrane absorption.

Physical Characterization Lovastatin Supersaturated-SNEDDS

The optimum formula containing 5% oleic acid, 80% Tween 80, and 15% PEG 400 was dispersed in phosphate buffer pH 7.2 at a temperature of 37± 2ºC. The size of droplets, polydispersity index, and zeta potential from the emulsion were measured using a Particle Size Analyzer (PSA-HORIBA SZ1000®).

Stability Evaluation of Lovastatin Supersaturated-SNEDDS

Evaluation of physical stability of lovastatin supersaturated-SNEDDS was carried out by storing the preparation at a temperature of 40 ± 2ºC for 28 days and testing with freeze-thaw methods for 5 cycles. Physical evaluation was carried out by measuring the percent transmittance and emulsification time. As a comparison, the same test was carried out for lovastatin SNEDDS.

In vitro Biological Membrane Absorption Test for Supersaturated-SNEDDS Lovastatin Formula

The *in vitro* testing was carried out using intestinal segments of male Wistar rats with the methods was approved by ethical clearance 41/EC/KEPK/XI/2023. The

rat intestinal segment used was 15 cm from the pylorus (jejunum segment). The intestines were cut and washed using 0.9% NaCl and turned over. The inverted rat intestinal segment was placed in a Crane and Wilson tube and filled with 2 mL of 0.9% NaCl solution as serosal fluid. The tube is inserted into a beaker that contains mucosal fluid, and phosphate buffer pH 7.2 containing the supersaturated preparation-SNEDDS lovastatin. The test temperature was controlled at 37 ± 2°C while stirring at 200 rpm. Serosal fluid sampling was carried out every 10 minutes for 1 hour. Lovastatin levels in serosal fluid were analyzed using HPLC acetonitrile: water (70:30) as a mobile phase in C18 column, 5µm, 4.6 mm x 150 mm (Merck®). The similar test also carried out for pure lovastatin and SNEDDS containing lovastatin.[18]

RESULTS AND DISCUSSION

Selection of Oil, Surfactant, and Cosurfactant Phases

Oleic acids was selected as an oil phase with a percent transmittance value of 22.7 ± 0.24%, while Tween 80 and PEG 400 were selected as surfactant and co-surfactant with transmittance value of 97.2 ± 0.39%, and 83.2 ± 1.74%, respectively. Percent transmittance was used as the indicator for clarity of the mixture. It assumed that the clearest mixture provided the best solvent for lovastatin. Oleic acid has often been used in the SNEDDS formula for lipophilic drugs that have poor solubility such as pentagamavunon-0[19], mefenamic acid[20], tetrandrine[21],

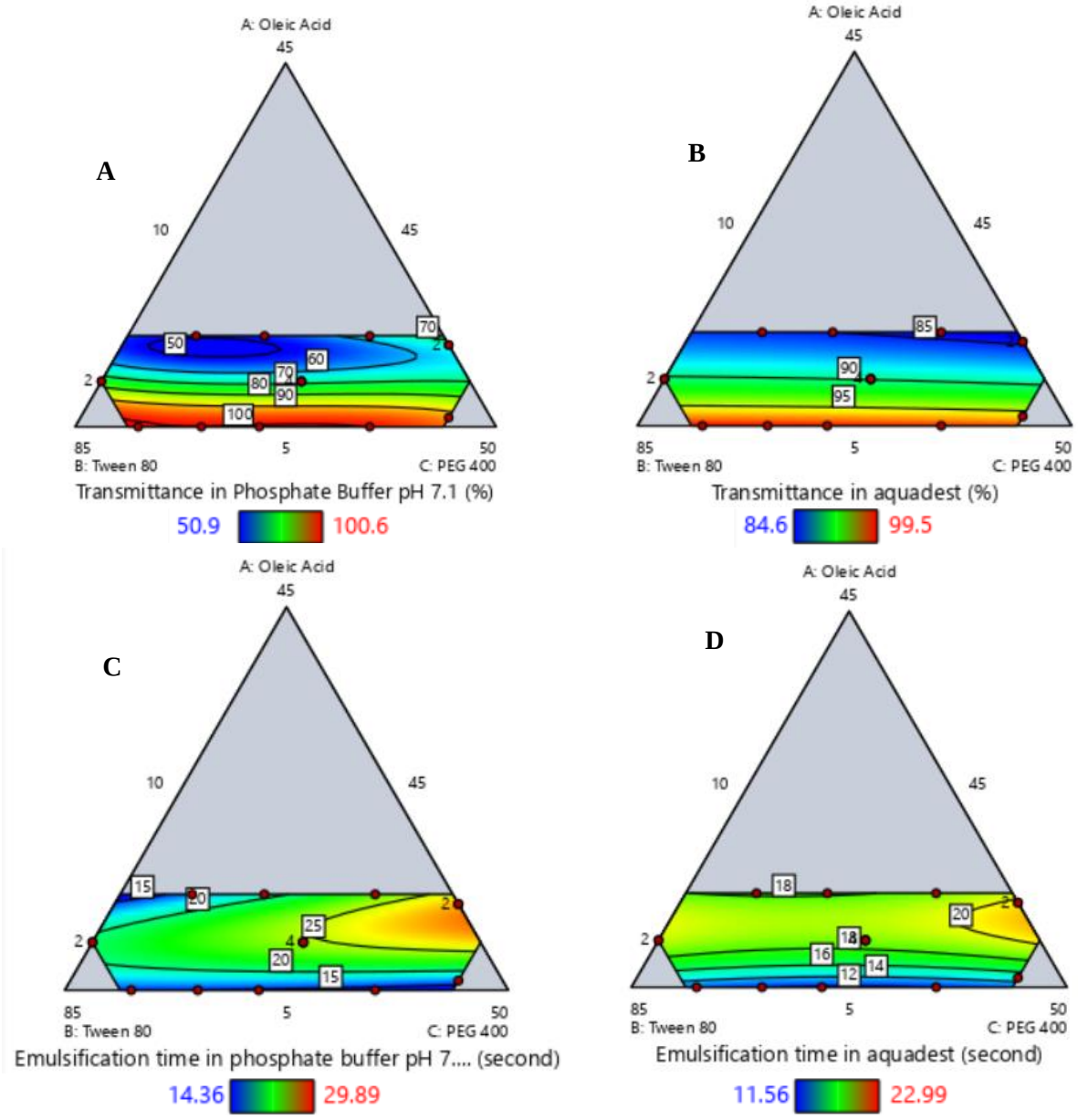

Figure 3: Contour plot of the relationship between independent variables (oleic acid, Tween 80, and PEG 400) on the response percentage transmittance in A) buffer phosphote pH 7,2 and B) distilled water, emulsification time in C) buffer phosphate pH 7,2 and D) distilled water

Table 3: The polynomial equation relationship between the independent variable and the response

Responses	Polynomial Equation		
	Oleic Acid	Tween 80	PEG 400
Percent transmittance in phosphate buffer pH 7.2	+1081.64	+0.25	-3.84
Percent transmittance in water	+6.19	+1.12	+1.07
Emulsification time in phosphate buffer pH 7.2	-21.48	-0.04	-0.43
Emulsification time in water	-11.69	+0.02	+0.14

Table 4: Predictive and observational response results for the lovastatin super-SNEDDS formula

Response	Predictive results	Experimental results	interpretation
Percent transmittance in phosphate buffer pH 7.2 (%)	96.1-105.77	100.4 ± 0.06	*In range*
Percent transmittance in water (%)	97.8-100.96	100.4 ± 0.13	*In range*
Emulsification time in phosphate buffer pH 7.2 (second)	11.0-19.33	15.1 ± 0.24	*In range*
Emulsification time in water (second)	8.2-17.20	13.1 ± 0.28	*In range*

furosemide[22] and glimepiride[23]. Tween 80 has a high hydrophilic-lipophilic balance (HLB) value of 15. A high surfactant HLB value (HLB>10) will produce a clear solution because it easily forms an oil-in-water (O/W) type nanoemulation.[24],[25] According to Miksusanti *et al*., (2023) Tween 80 and PEG 400 will be adsorbed on the surface of the nanoemulsion so that it can reduce the secondary surface tension, prevent aggregation, and produce good physical stability.[26]

Determination of Equilibrium Solubility (Seq) of Lovastatin

Determination of the equilibrium solubility of lovastatin in oleic acid was carried out for 7 days. The difference in lovastatin levels in oleic acid on days 6 and 7 is 1.12% (≤5%) which indicates that oleic acid has reached the saturation point and cannot dissolve lovastatin over again. According previous research by Thomas *et al*., (2012), the solubility test reached the equilibrium condition if the difference in dissolved drug levels on days 6 and 7 is no more than 5%.[9]

Determination of Emulsification Efficiency using Pseudoternary Phase Diagrams

The purpose of the pseudo-ternary phase diagram is to determine the emulsification area of a mixture of oleic acid, Tween 80, and PEG 400 with various concentration ratios to produce stable SNEDDS.[16] The parameters used to assess whether SNEDDS preconcentrate can produce good nanoemulsions are the percent transmittance value and emulsification time. Systems with a clear dispersion (percent transmittance value above or equal to 90%) and an emulsification time of less than 1 minute will be included in the grade A category and used as an emulsification area for a mixture of oleic acid, Tween 80, and PEG 400. Based on the 46 preparations that have been carried out, 11 preparations are included in the grade A category shown in Figure 1. Percent transmittance describes the clarity of the resulting nanoemulsion, according to Huda & Wahyuningsih (2016) a transmittance value close to 100% indicates that the size of the dispersion droplets produced by SNEDDS preconcentrate has reached nanometer sizes.[27] Emulsification time is the time required for SNEDDS preconcentrate to form a nanoemulsion in water media. Based on this area, it is known that the oleic acid concentration range that produces a clear dispersion and emulsification time that meets the requirements was 5% to 15%, the Tween 80 range was 45% to 80%, and the PEG 400 range was 10% to 45%.

Optimization of Supersaturated-SNEDDS Lovastatin Formula using D-Optimal Mixture Design

There were 16 experimental runs, the responses measured were percent transmittance and emulsification time in distilled water and phospate buffer pH 7.2 as shown in Table 1. The upper and lower limits for oleic acid, Tween 80, and PEG 400 were determined based on the results pseudo-ternary phase diagram. Optimization using D-optimal Mixture Design software gave percent transmittance response value of around 50.9-100.6% and 84.6-99.5% in phosphate buffer pH 7.2, and water, respectively . Emulsification time for all runs were less than 60 seconds in all dispersed medium (phosphate buffer pH 7.2 and water). Based on the percent transmittance, it shows that runs 1, 8, 9, and 11 had a percent transmittance 100-100.6%, while runs 1, 3, 8, 9, and 11 in distilled water showed a clear solution with a percent transmittance 96.9-99.5%. The clear appearance of the nanoemulsion in each run can be seen in Figure 2. Based on the response analysis for the optimization of the lovastatin supersaturated-SNEDDS formula, the most suitable models was cubic for the response of percent transmittance in phosphate buffer pH 7.2, while the quadratic model applied for percent transmittance in distilled water and emulsification time in both phosphate buffer pH 7.2 and water. The ANOVA results showed the significant p values ($\alpha<0.05$) as in Table 2. It was indicated that there was a statistically significant differences between experimental runs. The lack of fit value indicated the inaccuracy of the model, it is said to be good if the lack of fit value is not significant ($\alpha>0.05$).[13] The contour plot of the percent transmittance response showed the red area had a high percent transmittance close to 100% at the bottom of the diagram with a low oleic acid concentration (Figure 3A and 3B). The greater the amount of oleic acid added to the lovastatin supersaturated-SNEDDS formula produced the lower the percent transmittance value as showed in the blue area. The ternary diagram of the emulsification time response showed that the less Tween 80 added to the mixture would induce longer

Table 5: Stability of lovastatin super-SNEDDDS and SNEDDS at 40°C and freeze thaw 5 cycles

Formula	Response	Day 0	day 7th	day 14th	day 21th	day 28th	After 5 cylcle of freeze thaw
Supersaturated-SNEDDS Lovastatin	Percent transmittance (%)	100.56 ± 0.135	99.92± 1.178	100.18± 0.069	99.29± 0.08	98.72± 0,069	98.90 ± 0.265
	Emulsification time (second)	14.79± 0.945	15.29 ± 0.195	22.06± 2.48	27.39± 0.425	28.18± 0.716	22.08 ± 1.584
SNEDDS-Lovastatin	Percent transmittance (%)	100.46± 0.037	100.76 ±0.222	100.17± 0.153	98.56± 1.420	98.41± 0.444	99.36 ± 0.139
	Emulsification time (second)	14.31± 0.986	16.84± 1.279	19.44± 3.096	21.62± 0.384	22.60± 0.706	22.22 ± 0.868

emulsification time for lovastatin supersaturated-SNEDDS as showed in the red area (Figure 3C and 3D). Tween 80 as surfactants played a role in reducing the size of emulsion droplets and maintaining the active substance for a long period at the absorption site, as consequence there would be no deposition occured in the digestive tract. In the polynomial equation of Design-Experts showed that the oleic acid component most influences all responses, both percent transmittance and emulsification time, because has highest value as showed in Table 3. Optimization using D-Optimal Mixture Design showed that the optimum supersaturated-SNEDDS consists of 5% oleic acid, 80% Tween 80, and 15% PEG 400, with a desirability value of 0.969. Observation on the optimum formula for lovastatin supersaturated-SNEDDS was listed in Table 4. The results showed that all values were lies in the intervals of the predictive value range.

Physical Characterization of Lovastatin Supersaturated-SNEDDS Optimum Formula

The optimum formula for lovastatin supersaturated-SNEDDS had a droplet size of 18.07±1.550 nm, while the droplet size of lovastatin SNEDDS is 15.73 ± 0.503 nm. The increase in size in the lovastatin supersaturated-SNEDDS formula occurred due to an increase in the amount of drug contained in the SNEDDS preconcentrate due to reduced solubilization capacity.[29] Lovastatin supersaturated-SNEDDS had a significantly greater polydispersity index value (0.34 ±0.011) compared to the lovastatin SNEDSS preparation (0.31±0.015). Based on Nurismawati & Priani, (2021) the polydispersity index value that is well be accepted in the range of 0-0.5. The zeta potential values of lovastatin supersaturated-SNEDDS and lovastatin SNEDDS preparations after being dispersed in a phosphate buffer medium pH 7.2 were -228.90 ± 9.054 mV and -187.13 ± 17.632 mV, respectively. Zeta potential described the potential difference between the surface layers of strongly bonded ions on the surface of the solid and the electroneutral.[30] According to Nurismawati & Priani, (2021), a good zeta potential value is less than -30

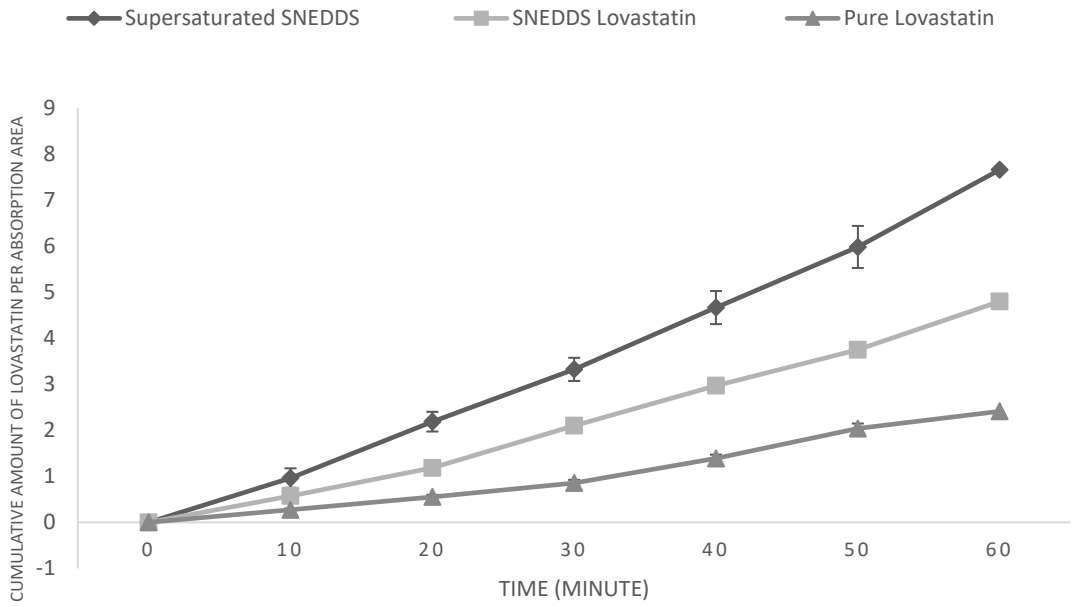

Figure 4: Cumulative amount of lovastatin absorbed per absorption area vs time. The highest amount was found in Supersaturated-SNEDDS

Table 6: Flux, permeability, and lag time of super-SNEDDS, SNEDDS, and pure lovastatin

Formula	Flux ($\mu g\ cm^{-2}\ min^{-1}$)	Kp (cm/min)	Lag Time (min)
Supersaturated-SNEDDS	0.13±0.007*	$4.88 \times 10^{-5} \pm 0.272 \times 10^{-5}$*	3.72±0.541*
SNEDDS	0.08±0.003*	$3.14 \times 10^{-5} \pm 0.119 \times 10^{-5}$*	4.78±0.503*
Pure Lovastatin	0.04±0.002*	$1.66 \times 10^{-5} \pm 0.067 \times 10^{-5}$*	7.03±0.393*

*mean value are significantly different from others ($\alpha<0.05$)

mV or more than +30 mV, the more negative or more positive the globule surface charge, the more stable the dispersion system formed.[31]

Physical Stability of Supersaturated-SNEDDS Lovastatin with heating temperature 40 ± 2ºC and Freeze-Thaw method

Stability testing by heating was carried out using a constant temperature oven of 40 ± 2ºC and freeze-thaw cycling test. The lovastatin supersaturated-SNEDDS and SNEDDS were stable after 28 days of storage in an oven at 40ºC and after freeze thaw cycles. According Huda & Wahyuningsih, (2018), the transmittance value was more than 90%, indicating that the size of the droplets was still nanometer.[27] The emulsification time of both lovastatin supersaturated-SNEDDS and SNEDDS showed no significant difference after being stored in the oven for 28 days and after freeze-thaw 5 cycles (Table 5). The percent transmittance value of lovastatin supersaturated-SNEDDS was maintained above 95% while emulsification time was still less than 60 seconds. It indicated that the lovastatin supersaturated-SNEDDS preparation is physically stable when dispersed in phosphate buffer pH 7.2

Oral Absorption Evaluation using Everted Gut Sac Methods

The cumulative amount of lovastatin absorbed into the serous fluid per absorption area was calculated as a flux. The cumulative amount of lovastatin absorbed was achieved the highest when lovastatin was in the form of supersaturated-SNEDDS (Figure 4). According previous research by Thomas *et al*., (2013) using *in vitro* lipolysis test, simvastatin supersaturated SNEDDS induced drug release 1.8 times compared SNEDDS preparations.[12] The apparent permeability value (Kp) of Supersaturated-SNEDDS Lovastatin have a significantly higher value compared to the SNEDDS Lovastatin and pure Lovastatin preparations (Table 6). The supersaturated-SNEDDS preparation containing lovastatin increased the permeability of lovastatin 1.55 times higher than SNEDDS preparation and 2.94 times higher compared to pure lovastatin. The average lag time value of the lovastatin supersaturated-SNEDDS was significantly faster (p-value <0.05) compared to the lovastatin SNEDDS and pure lovastatin preparations, namely 3.72± 0.541 minutes. The lag time of pure lovastatin was 7.03±0.393 minutes. Lovastatin supersaturated-SNEDDS has a faster lag time as nanoemulsion droplets contain supersaturated amounts of drug that carried a higher amount of drug compared to conventional SNEDDS preparations. The shorter lag time induced the rapid time for the drug to reach peak levels in plasma (Tmax).[10] Supersaturated-SNEDDS system contained more drugs with less carrier phase (supersaturated), therefore the distance for the drug to leave the carrier into the systemic circulation was shorter compared to the release of SNEDDS preparations with a smaller amount of drug and more carriers. Consequently, the flux value in the supersaturated-SNEDDS preparation was higher compared conventional SNEDDS. According to Binarjo *et al* (2018), lipophilic drugs experienced a rate-limiting step in the dissolution phase. Pure lovastatin has poor solubility in phosphate buffer media pH 7.2, thus it had a low apparent permeability value and a longer lag time.

CONCLUSION

The optimum formula for lovastatin supersaturated-SNEDDS contains 5.5 mg/g pre-concentrated lovastatin consisting of 5% oleic acid, 80% Tween 80, and 15% PEG 400. Lovastatin supersaturated-SNEDDS has a particle size in nanosize, low polydispersity index, and negative charged zeta potential. The lovastatin supersaturated-SNEDDS preparation was physically stable in the heating stability test at 40ºC and the freeze thaw method. The oral absorption test showed that the cumulative amount of lovastatin absorbed in the lovastatin supersaturated-SNEDDS preparation was 1.6-fold higher than the lovastatin SNEDDS preparation and 3.3-fold higher than pure lovastatin.

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