2017 년 8 월

석사학위 논문

A study on the development of platform technology to enhance bioavailability of drug

조선대학교 대학원

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약물의 생체 이용률 개선을 위한 경구투여용
플랫폼 기술 개발에 관한 연구

2017 년 8 월 25 일

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이 논문을 약학석사학위 신청 논문으로 제출함

2017 년 4 월

조선대학교 대학원

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2017 년 5 월

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약물의 생체 이용률 개선을 위한 경구투여용 플랫폼 기술 개발에 관한 연구

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본 연구의 목적은 경구 투여시 지질류 흡수 경로로 흡수되어 간 초회 효과를 회피하고 생체 이용률을 높일 수 있는 고형 지질 나노입자 (solid lipid nanoparticles, SLNs) 기술 기반 제형을 개발하여 현재 항 파킨슨 약물로 개발되어 경피 흡수 패취제로 시판, 사용중인 로티고틴을 경구 투여용 의약품으로 개발하고자 함이다.

파킨슨병은 중뇌 흑질에 존재하는 도파민 분비 신경세포의 소실로 나타나는 질환으로서 1 차적 치료제로 도파민 작용약물 (Dopamine agonists)이 고려된다. 로티고틴은 도파민 수용체 특히 D_3 수용체에 선택적으로 작용하므로 항 파킨슨 약물로 각광받고 있다. 하지만 로티고틴은 난용성인 약물이며, 경구로 투여시 간 초회 효과에 의한 대사로 인해 약물의 체내 흡수율이 낮은 약물로 알려져 있다. 현재 시판중인 경피 흡수 패취제-Neoupro® 의 경우 부착 부위의 피부 반응이 흔한 부작용으로 보고 되었으며 1 일 1 회 매번 다른 부위에 부착해야 하는 불편함이 존재한다. 따라서, 생체이용률을 높일 수 있는 경구 제형으로 개발한다면 상기의 단점을 극복하고 환자의 복약 순응도를 높일 수 있다. 이에 경구 투여시 로티고틴의

생체 이용률을 높이기 위해 소장 내에서 지질 물질의 흡수와 이동에 관여하는 Peyer's patch 경로를 통해 림프계로 약물을 이행시켜 간 초회 통과를 회피할 수 있는 고형 지질 나노 입자 기술을 기반으로 하는 제제를 개발하고자 하였다.

고형 지질로는 Precirol® ATO 5 를 사용하였는데 이는 림프계로의 uptake 를 촉진시키기 위해서는 긴 탄소 사슬의 지질이 유리하기 때문이며, 계면활성제와 유화제로는 TPGS 와 Solutol® HS 15 를 사용했다. 이들은 약물의 체내 흡수시 P-gp 단백질에 의한 efflux 를 억제하여 난용성이며 지용성인 물질의 림프계로의 이동을 용이하게 해 생체 이용률을 높이는 효과가 있다.

이들을 이용하여 제형을 설계시 최적의 조성을 알아내기 위해 실험 설계법 (Design of Experiment)을 시행하였다. 본 연구에서는 실험 설계법의 여러 방법들 중에서 혼합물 설계법 (Mixture designs)을 사용하였고, 꼭지점 설계법 (Extreme vertices design)을 통해 최적화 과정을 거쳤다. 실험 설계법에서 제시된 조성으로 제조된 로티고틴 고형 지질 나노 입자는 입자경, 다분산 지수, 봉입률 그리고 제타 전위 등을 측정함으로써 물리 화학적 성질을 확인하고, in vitro release 평가로 약물의 방출 특성을 평가하였다.

본 연구는 실험 설계법을 적용하여 보다 과학적인 방법으로 조성 성분들간 영향을 분석하여 최적의 제형을 설계하였고, 이를 바탕으로 장기간 안정적으로 높은 농도의 로티고틴을 함유하며, 경구 투여시 생체 이용률 개선의 잠재성을 갖는 제형을 개발하였다. 또한, 로티고틴 외에도 난용성이며 생체 이용률이 낮아 경구외의 투여경로로 개발되던 약물들에도 범용적으로 응용할 수 있으리라 생각된다.

2. Introduction

Parkinson's disease (PD) is a common neurolodegenerative disease that affects about 1 to 1.5% of population of 60 years or older. The global trend of aging and the prolonged life expectancy are expected to increase the incidence of PD [1].

The most commonly used drug for PD is levodopa, which relieves various symptoms and improves the quality of life of the patient. However, resistance to long-term levodopa treatment is expressed, and levodopa-induced dyskinesia (LID), as well as side effects [2].

Rotigotine, first developed as an adjuvant treatment for PD, is a non-ergorline dopamine agonist that acts selectively on D_3 dopamine receptors, which play an important role in PD. However, rotigotine is a poorly soluble drug, and it belongs to drugs that have a low bioavailability when administered orally by the first pass effect of the liver [3].

Many studies have been reported to develop various drug delivery systems to enhance the efficiency of delivery of rotigotine into the body. An example is the transdermal patches marketed under the trade name Neupro® for indications for Restless legs syndrome (RLS), a characteristic symptom of PD. But in the case of transdermal patches, skin reactions such as erythema and rash on the attachment site appear as common side effects, and there is a hassle to attach them to different sites every 24 hours [4].

The oral route is still considered the best way to administer drug with higher patient compliance, lesser complications, and lower cost, in comparison with other routes [5,10].

The use of solid lipid nanoparticles (SLNs) can be suitable when considering a new oral platform with two factors in mind: sustained release of rotigotine to relieve symptoms of PD and relief of discomforts in using transdermal patches. SLNs have been actively studied as carriers of targeted drug delivery with the solubilization of particularly poorly soluble drugs as vehicles more suitable for lipid-soluble drugs than for water-soluble drugs. When the drug is encapsulated into the SLN and administered orally, migration into the lymphatic system in the small intestine is facilitated, and drug-loaded SLNs are able to avoid first-pass metabolism. Thus, a bioavailability of drug can be improved [5, 11, 13, 14].

But various factors such as particle diameter, surface charge, type of lipid and concentration of the surfactant or emulsifier are very important because that affect to regulation the lymphatic delivery of drugs. Lymphatic system is known to play an important role in uptake of lipids and lipophilic drugs through intestine. Thus formulations composed of long chain triglycerides (LCT) and a proper surfactants favor transfer into the lymphatic system [7, 9].

In this study, we developed rotigotine loaded SLNs (RG-SLNs) with Precirol® ATO 5 as solid lipid, and TPGS and Solutol® HS 15 as surfactants to deliver rotigotine through oral administration. Precirol® ATO 5 is a long chain triglyceride (LCT), which is known to be easy to uptake into the lymphatic system and was used as a solid lipid in this study. TPGS was used as a surfactant because of the advantage of avoiding P-gp efflux. There're reports that the intestinal absorption and oral bioavailability of poorly soluble drugs were improved in TPGS emulsified SLNs, maybe due to inhibition of drug efflux by TPGS, along with intestinal lymphatic uptake [15,16,17]. Therefore, it is valuable noting that SLNs may perform as efficient oral delivery systems for rotigotine. The optimal composition of SLNs were attained by Minitab® program for design of experiment (DoE) that predicted the best parameters by investigating the combined effect of various factors [6, 8, 12]. It was conducted after carrying out the preliminary study with different ratio of lipids, surfactants and water. The developed RG-SLNs were prepared with optimized composition and characterized for physiochemical properties such as particle diameter (PD), polydispersity index (PDI) and entrapment efficiency (EE). In vitro release studies were done to evaluate releasing characteristics of RG-SLNs [18].

3. Materials and Methods

3-1. Materials

Rotigotine was purchased from Sigma–Aldrich Chemical (St. Louis, MO, USA). The solid lipid, Precirol® ATO 5 (Glyceryl Distearate) was supplied by Gattefosse SAS (Saint Priest Cedex, France). Kolliphor® TPGS (Vitamin E Polyethylene Glycol Succinate) and Solutol® HS 15 (Polyoxyl 15 hydroxystearate) were obtained from BASF (Ludwigshafen, Germany). Acetonitrile (ACN) and methanol of all HPLC grade were purchased from Sigma–Aldrich Chemical (St. Louis, MO, USA). All other reagents used in this study were laboratory grade.

3-2. Preparation of SLNs

Hot melting-sonication methods was used to prepare RG-SLNs and blank SLNs. In brief, accurately weighed Precirol® ATO 5, Kolliphor® TPGS, Solutol® HS 15 and rotigotine were mixed and heated at 70℃ for 45 min in a water bath. Hot purified water (over 70℃) was added into the melted mixtures and mixed again with stirring at 70℃ for 30 min. Then the crude SLN suspension was homogenized using a probe sonicator at 70℃ for 10 min in a water bath. The obtained SLN suspension was filtered through syringe filter (0.8 μm pore size) and cooled down at 4℃ or room temperature during 6 hrs. Blank SLNs were prepared in a same process without rotigotine.

3-3. Experimental design (DoE)

The optimal ratio of components was attained by design of experiment (DoE) using Minitab®16 [6,8,19]. To put it in detail, extreme vertices design was used on preliminary experimental data. Extreme vertices design is used when the upper and lower limits are present or constraints are included in the experiment. This is particularly useful when setting up a relationship to optimize response values.

Independent variables were the amount of Precirol® ATO 5 as a solid lipid (X_1), the amount of Kolliphor® TPGS as a surfactant (X_2), the amount of Solutol® HS 15 as a co-surfactant (X_3) and the amount of water (X_4). The established dependent variables were: particle diameter (Y_1) and EE (Y_2). The optimization model was selected considering the main effects, interaction effects, and quadratic effects of the major independent variables, and the optimal conditions were selected. The equation generated from the DoE is given below:

Equation 1:

$$Y_n=a_1X_1+a_2X_2+a_3X_3+a_4X_4+a_{12}X_1X_2+a_{13}X_1X_3+a_{14}X_1X_4+a_{23}X_2X_3$$

Y_n : the dependent variable

X_1, X_2, X_3 and X_4 : independent variables

3-4. Optimization and validation of SLNs

In order to verify the reliability of the developed model, optimized variables were used to prepare a checkpoint solid lipid nanoparticle formulation. The results were compared with predicted values to evaluate the predicted error. The graphical and numerical analyses were executed by Minitab®16 to obtain the optimum values of the variables (Table 2).

3-5. Quantification of rotigotine by HPLC analysis

The analysis of rotigotine was performed by HPLC. The HPLC system were consisted of pumps (LC-10ADVP, LC-20AD), an autosampler (SIL-10ADP) and UV detector (SPD-10VP) (Shimadzu, Tokyo, Japan). C18 column (Luna C18, 4.6 mm × 150 mm, 5 μm; Phenomenex, Torrance, CA, USA) was used and was heated at 40℃. Rotigotine was eluted with a mobile phase consisting of 30 mM dipotassium hydrogen orothophosphate: MeOH (30:70, v/v%) and detection wavelength was fixed at 225 nm. Injection volume was 20 μl and flow rate was 1.0 ml/min.

3-6. Characterization of SLNs

The particle diameter (PD), polydipersity index (PDI), and zeta potential (ZP) of the prepared SLNs were analyzed using Zeta-potential & Particle size Analyzer ELSZ-2000 series (Otsuka Electronics Co., Ltd, Japan). Samples were diluted with distilled water to reach a proper concentration before measurement. All analyses were performed at room temperature.

The entrapment efficiency of rotigotine in SLNs was determined by the HPLC assay. The aliquot of SLNs (100 μl) was diluted 10 times with 900 μl of ACN and then subjected to centrifugation. The supernatant was directly injected to the HPLC system. The encapsulation efficiency (EE) was presented according to the equation:

Equation 2:

$$\mathrm{EE}(\%) = \frac{\text{Encapsulated drug } (\mu g/ml)}{\text{Total drug } (\mu g/ml)} \mathrm{X100}$$

In vitro release of rotigotine from RG-SLNs was evaluated using the dialysis bag diffusion method. Phosphate buffered saline (PBS; pH 7.4) containing 1% (v/v) tween 80 was selected as the release medium. The dialysis bag with a 6–8 kDa molecular weight cut-off was soaked in the release medium for 10 min before use. RG-SLNs containing 3 mg of rotigotine were taken into the bag, then both ends were tightly sealed and immersed into 200 mL medium in a glass vessel. The vessel was placed onto a hot plate with a speed of 100 rpm and maintained at a temperature of 37°C. At predetermined time intervals (0.25, 0.50, 1, 2, 4, 6, 8, 12 and 24), 0.5 mL of aliquots were taken and immediately replaced with an equal volume of fresh medium and rotigotine concentration was analyzed using HPLC. The experiments were performed in triplicate.

4. Results and discussion

4-1. Preliminary experiment for experimental design

The preliminary experiment is important to find out the critical factors that affect the parameters of evaluation, such as particle diameter, PDI, and EE (%), during development of SLNs. The purpose of the preliminary experiment was to determine the appropriate level of the factors to be input to DoE. As shown in Table 1, several SLNs were prepared with the fixed amount of solid lipids 10% (w/w) and changing the composition of the surfactant mixture (TPGS and Solutol® HS 15) and water amount. To choose appropriate factors, the particle diameter of the SLNs were evaluated (Figure 1). The amount of surfactant mixture (SM) and the composition ratio of SM were determined as critical factors. We set the target range of the particle diameter from 100 to 300 nm and selected the compositions of SLNs. The SLNs with from 5 to 7.5% (w/w) of SM satisfied the target range. Based on the results, the factors and the range of factors were determined for DoE. The cooling temperature did not effect on the initial particle diameter of SLNs, however, the particle diameter of SLNs prepared at RT increased after 1 week. The SLNs prepared at 4°C were stable for 1 week. The re-crystallization of solid lipid in cooling step of the preparation of SLNs was affected by the cooling process, especially, temperature, and the crystal form of solid lipid at 4°C was seemed to be more stable. Moreover, SLN 11-1 and SLN 12-1 that were prepared at RT changed into gel during cooling process. Therefore, the cooling temperature was fixed at 4°C for further study.

4-2. Experimental design (DoE)

The application of experimental design using with Minitab®16 program enables to not only reduce the number of experiments and the time consumed but also manage the risk of experiment failure [6,8,19]. Optimization of the preparation of SLNs was performed using the extreme vertices experimental design. Among the various methods, extreme vertices design is used when experimenting in a limited area, not all areas. This

experimental design is to conduct experiments by appropriately choosing several points consisting of all the vertices of the restricted region and the linear combination of these vertices. The extreme vertices design proposed us to obtain 17 different formulations, with a triplication of the center point, resulting in a total of 19 formulations. The triplication of the center point enables to value the experimental error. The minimum, medium, and maximum values of the variables used in the extreme vertices design were 8, 10 and 12 for Precirol® ATO 5, 1, 3.5 and 6 for Kolliphor® TPGS, 1, 3.5 and 6 for Solutol® HS 15 and 76, 83 and 90 for water, respectively. In the case of the dependent variables, the lower value, the upper value and the target value were designated as 50, 250, and 130 nm in the particle diameter and 90, 100, and 100 in the EE, respectively (Table 2).

As presented in Table 3, the models of polynomial equations implying an effects and interaction factors for particle diameter and EE were statistically analyzed. A particle diameter and EE ranged from 58.7-1116 nm and 54.67-96.37%. Average of particle diameter and EE were 309.447nm and 86.05%, respectively.

Independent variables were calculated by substituting the constants listed in Table 4 using the equation (1) mentioned in the methods section above.

Equation 3:

$$Y_1=5560.64X_1+114.082X_2+254.952X_3+52.6339X_4-77.9392X_1X_2-101.549X_1X_3-66.2916X_1X_4+16.9219X_2X_3$$

$$Y_2=-156.961X_1-2.02885X_2-0.419367X_3-0.552567X_4+2.55453X_1X_2+2.30194X_1X_3+1.87453X_1X_4-0.963576X_2X_3$$

Y_1, Y_2: dependent variables

X_1, X_2, X_3, X_4: independent variables

The R^2 means the amount of variation of observed response values as described in the model. The variation values of the particle diameter and EE explained in this experimental model are 95.7 and 86.55%, respectively. Lack of fit means one of the residual errors of the whole model. If this value is less than 0.05, it means that higher order equations are needed because it cannot be explained by the current term [19]. As shown in Table 4, the R^2 value and the lack of fit value are appropriate, thus, DoE can be judged

to be significant. In the case of particle diameters presented in DoE, the mixture contour plots with fixed center value of one of four variables showed that the dotted line inside the triangle is the target design space (Figure 2). As presented in Figure 3, similar results were obtained for EE. In the same way, when a mixture contour plot was performed on the remaining variables while only one variable is fixed, dotted spaces appeared in the triangle similar to the case of the above-mentioned particle diameter. By integrating these results, a design space was set up (Figure 4). To construct the design space, we used the upper limit value, the lower limit value, and the target value of the predetermined dependent variables in the design space setting. As shown in the overlaid contour plots of Figure 4, the areas satisfying all of the predetermined values of particle diameter and EE were indicated by white areas. Choosing any point in the area indicated by the white area is very useful because it determines the composition condition that can prepare the optimum SLNs and predicts the properties of the SLNs.

4-3. Validation of the DoE model

On the basis of the polynomial models, the results of response surface analysis were represented the effect of independent factors on each observed responses. In order to validate the models, an arbitrary point in the section presented in the white area of the overlaid contour plots (Figure 4) was selected and experimented with the composition. The selected composition values of solid lipid (Precirol® ATO 5), surfactant (Kolliphor® TPGS), co-surfactant (Solutol® HS 15) and water were 10, 2.5, 2.5 and 85 (%), respectively. The predictive values and evaluated values of particle diameter and EE were presented in Table 5. The evaluated values were similar to the predictive values obtained through the established models and the design space with the accuracy of 104.9% and 101.6% for the particle diameter and EE, respectively. Furthermore, another SLNs was prepared with the center value composition- Precirol® ATO 5, Kolliphor® TPGS, Solutol® HS 15 and water were 10, 3.5, 3.5 and 83 (%), respectively. The accuracy for the particle diameter and EE were 103.0% and 99.9%, respectively. The results suggest that the optimized model accurately predicted the particle diameter and EE of SLNs.

4-4. In vitro release study

The composition of RG-SLN was prepared as a center value of ATO 5 : TPGS : Solutol : water = 10 : 3.5 : 3.5 : 83. This composition is in the design space containing the optimal composition as shown in DoE overlaid plots (Figure. 4) and was prepared with this composition because it showed good predictability in the previous DoE evaluation. The release experiment was performed under sink conditions. Figure 5 represented in-vitro drug release profiles of optimized RG-SLNs and free rotigotine. Free rotigotine was released over 90% during 24 hr. Rotigotine was released from RG-SLNs without initial burst release, then 40% of rotigotine was released during 24 hr. The release pattern seemed like zero order release rate. Based on the curve obtained from in vitro release data, drug release from orally administered RG-SLNs occurs mostly via diffusion process through the solid lipid matrix degradation in the gut.

5. Conclusion

RG-SLNs were developed by hot melting-sonication method by employing the extreme vertices experimental design, after the preliminary study was performed with different ratio of mixtures. Through Minitab®16 software, it was possible to obtain RG-SLNs with optimum particle diameter and high EE. The optimized formulations were characterized. Rotigotine was successfully loaded in SLNs with high EE and desirable particle diameter range. Between the predicted values of SLNs by the DoE model and their evaluated values from actual experiments, linearity was found. Furthermore, a low predicted error was observed in the responses, suggesting a good predictive ability of the design. Finally, our results demonstrated that RG-SLNs could be a promising oral administration formulation of rotigotine for Parkinson's disease and Restless legs syndrome.

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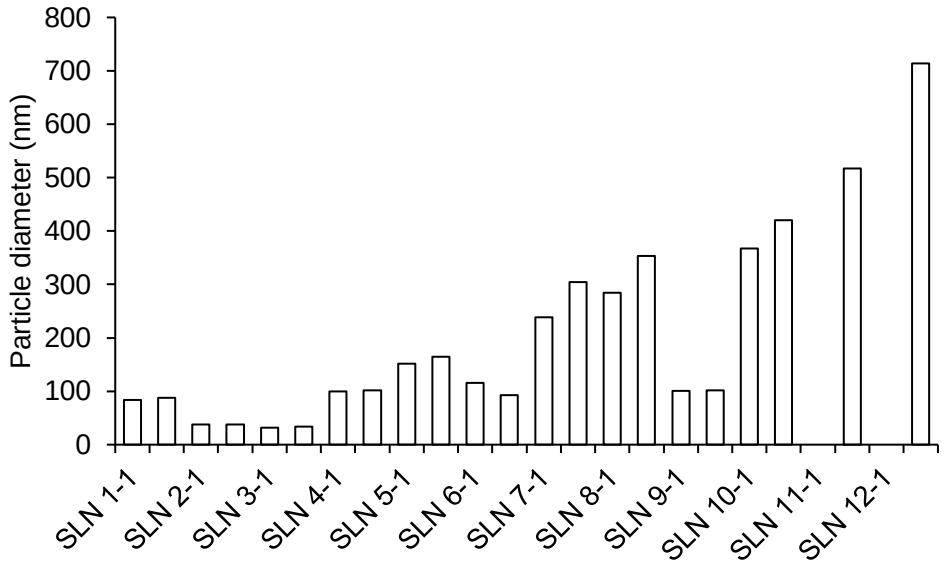

Figure 1. Particle diameter of SLNs from preliminary study

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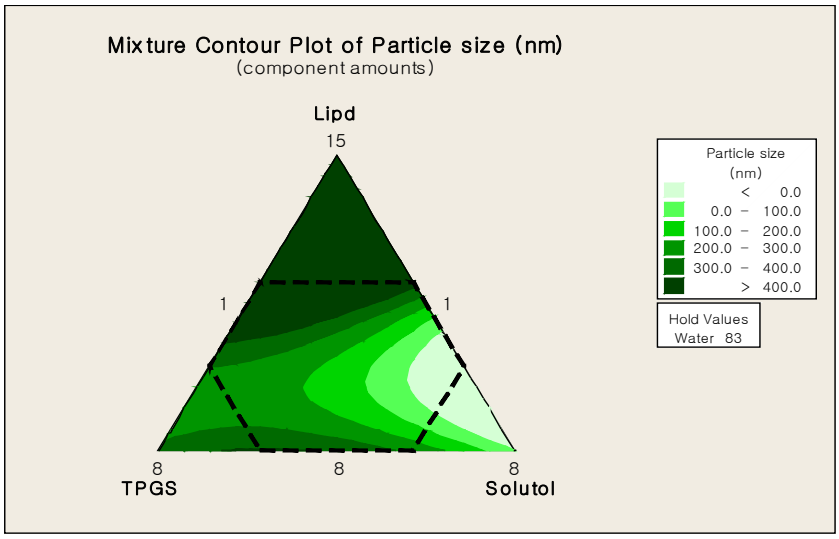

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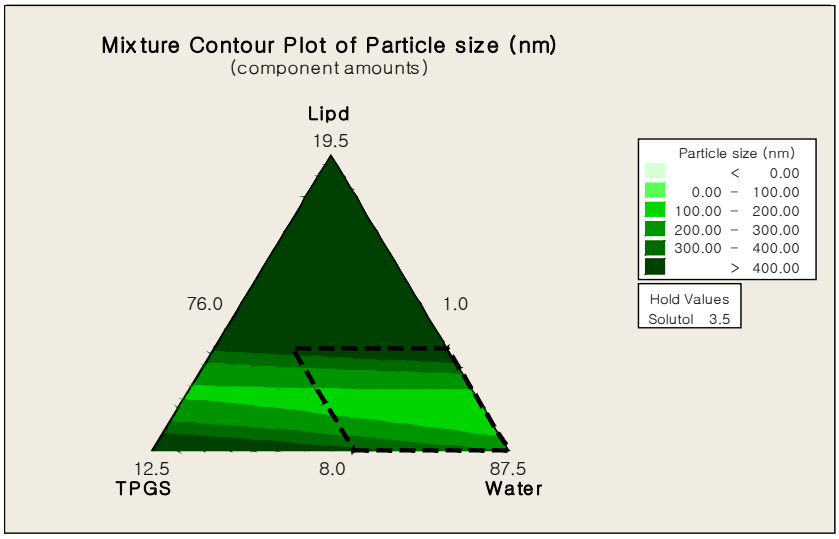

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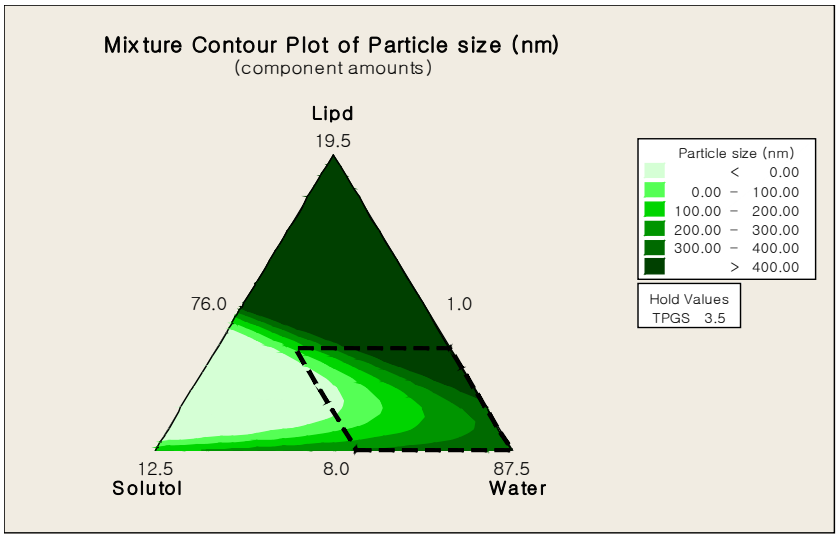


(d)

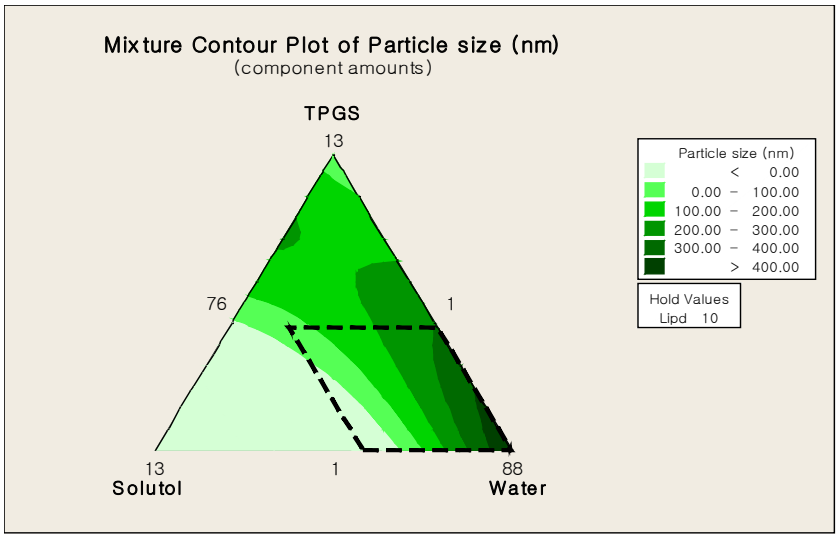
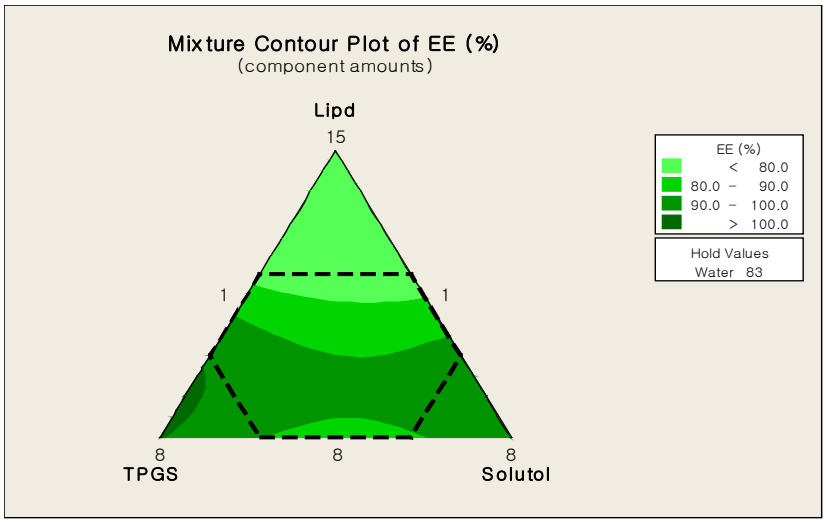
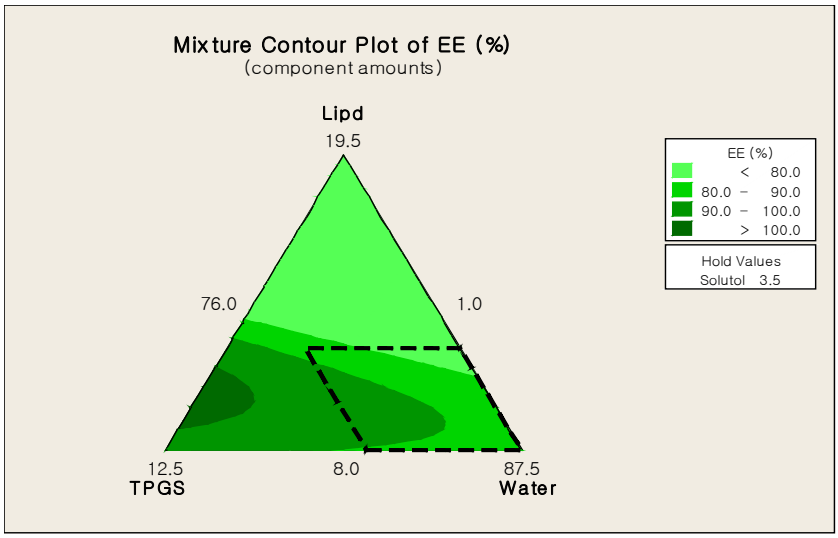


Figure 2. Mixture contour plots of particle diameter (nm) from DoE (a) when the hold value is water, (b) when the hold value is Solutol HS 15, (c) when the hold value is TPGS and (d) when the hold value is Precirol® ATO 5

(a)



(b)



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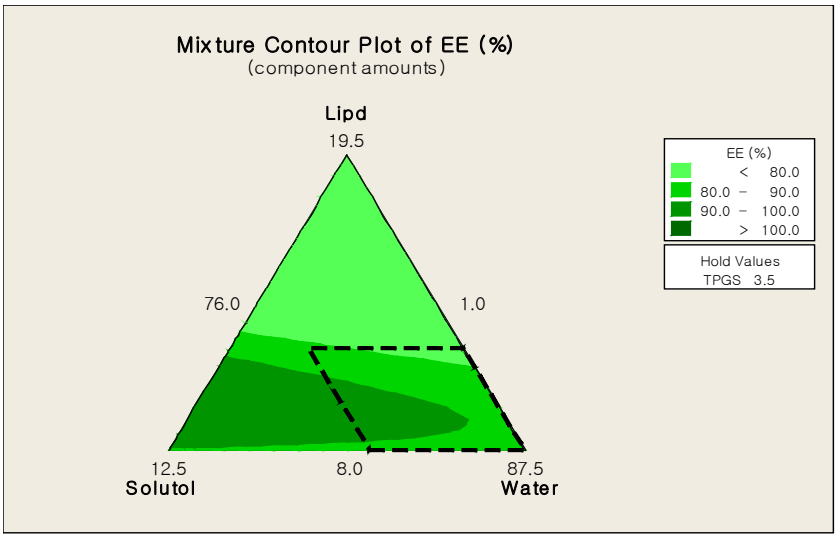

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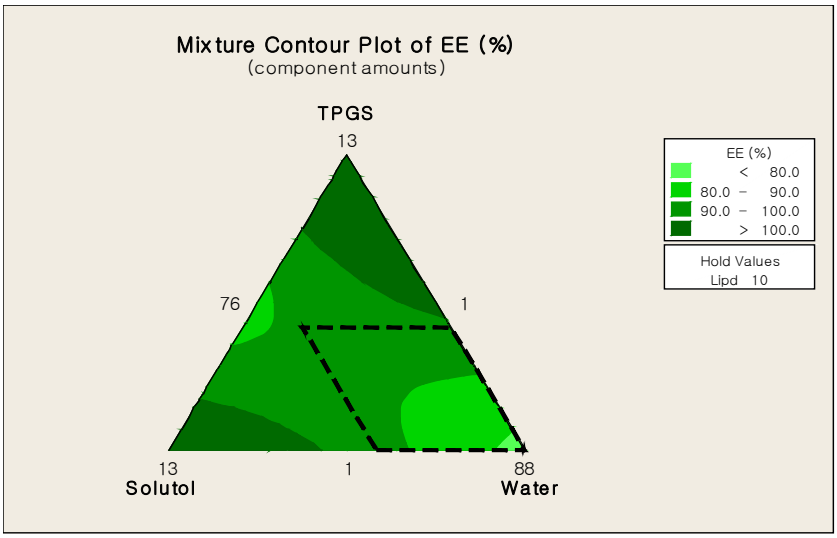
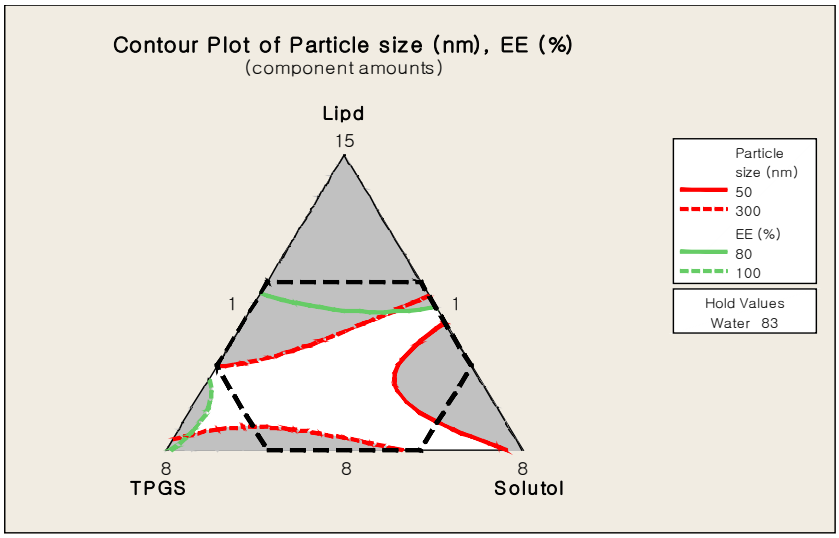
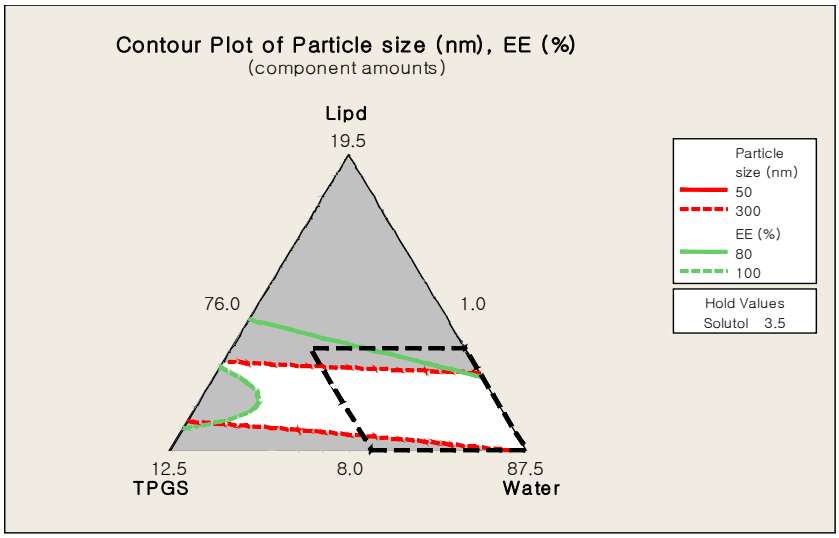


Figure 3. Mixture contour plots of EE (%) from DoE (a) when the hold value is water, (b) when the hold value is Solutol HS 15, (c) when the hold value is TPGS and (d) when the hold value is Precirol® ATO 5.

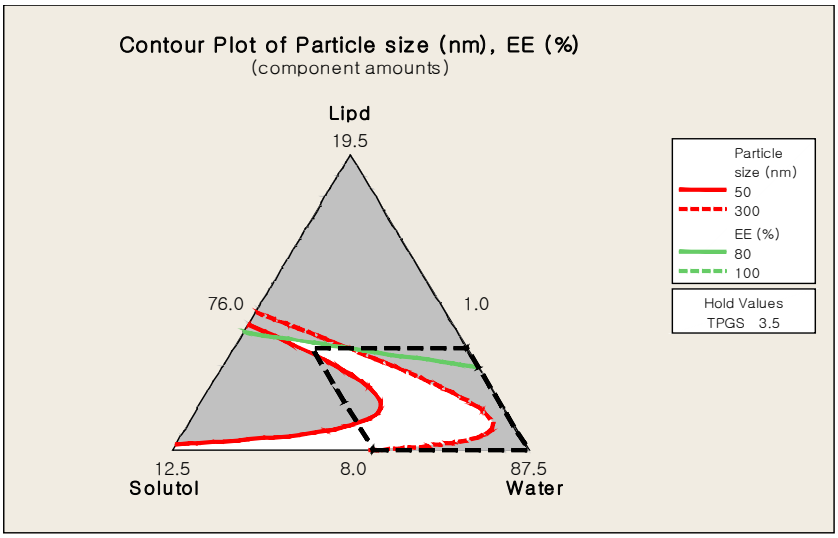
(a)



(b)



(c)



(d)

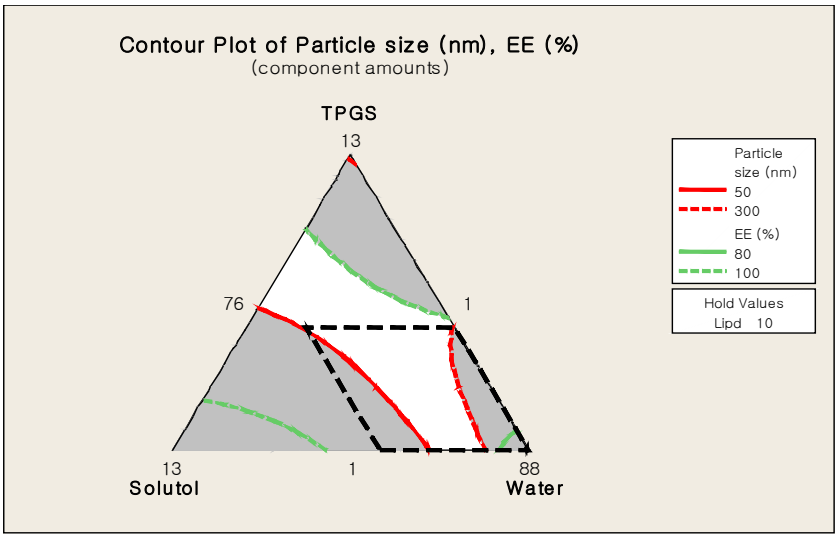


Figure 4. Overlaid contour plots of particle diameter (nm) and EE (%) from DoE (a) when the hold value is water, (b) when the hold value is Solutol HS 15, (c) when the hold value is TPGS and (d) when the hold value is Precirol® ATO 5.

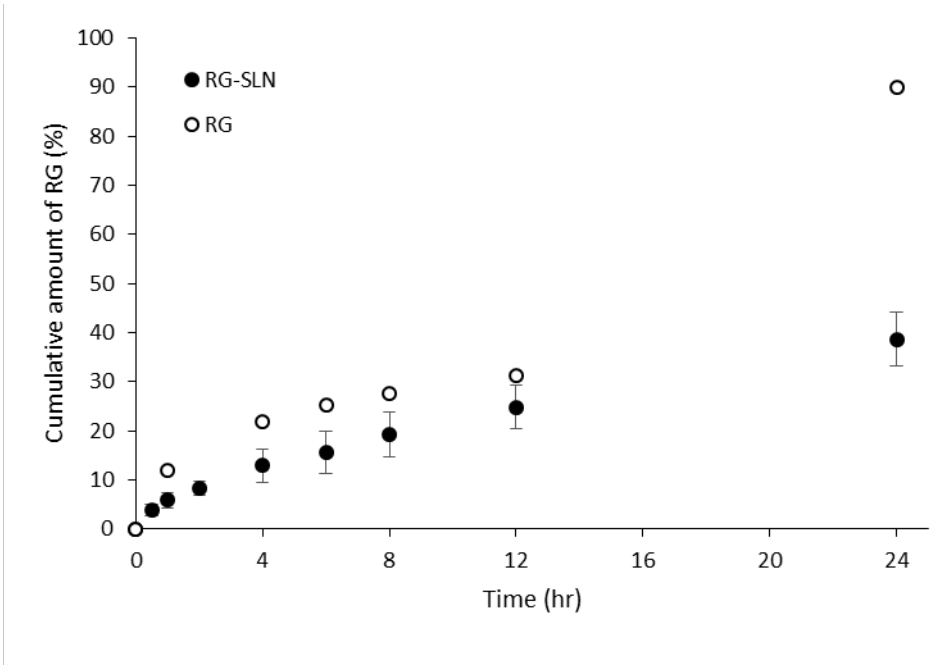


Figure 5. In-vitro drug release profile of free rotigotine and rotigotine solid lipid nanoparticles in phosphate buffer pH 7.4.

Table 1.The composition of SLNs for the preliminary study.

Formulation	Lipid (%)	TPGS (%)	Solutol (%)	Water (%)	Cooling Temp. (℃)
SLN 1-1	10	7.5	2.5	80	RT
SLN 1-2	10	7.5	2.5	80	4
SLN 2-1	10	5	5	80	RT
SLN 2-2	10	5	5	80	4
SLN 3-1	10	2.5	7.5	80	RT
SLN 3-2	10	2.5	7.5	80	4
SLN 4-1	10	5.625	1.875	82.5	RT
SLN 4-2	10	5.625	1.875	82.5	4
SLN 5-1	10	3.75	3.75	82.5	RT
SLN 5-2	10	3.75	3.75	82.5	4
SLN 6-1	10	1.875	5.625	82.5	RT
SLN 6-2	10	1.875	5.625	82.5	4
SLN 7-1	10	3.75	1.25	85	RT
SLN 7-2	10	3.75	1.25	85	4
SLN 8-1	10	2.5	2.5	85	RT
SLN 8-2	10	2.5	2.5	85	4
SLN 9-1	10	1.25	3.75	85	RT
SLN 9-2	10	1.25	3.75	85	4
SLN 10-1	10	1.875	0.625	87.5	RT
SLN 10-2	10	1.875	0.625	87.5	4
SLN 11-1	10	1.25	1.25	87.5	RT
SLN 11-2	10	1.25	1.25	87.5	4
SLN 12-1	10	0.625	1.875	87.5	RT
SLN 12-2	10	0.625	1.875	87.5	4

Table 2. Values of variables in Extreme vertices design.

Independent variable	Symbol	Minimum	Medium	Maximum
Precirol® ATO5	X_1	8	10	12
TPGS	X_2	1	3.5	6
Solutol HS 15	X_3	1	3.5	6
Water	X_4	76	83	90
Dependent variable	**Symbol**	**Lower value**	**Target value**	**Upper value**
Particle diameter	Y_1	50	130	250
EE	Y_2	90	100	100

Table 3. Design and results in Extreme vertices design.

Run Order	Pt Type	Blocks	Lipid (X_1, %)	TPGS (X_2, %)	Solutol (X_3, %)	Water (X_4, %)	Particle diameter (Y_1, nm)	EE (Y_2, %)
1	1	1	8	1	6	85	138.2	92.73
2	-1	1	11	2.25	2.25	84.5	346.2	83.1
3	-1	1	9	4.75	2.25	84	357.7	88.31
4	0	1	10	3.5	3.5	83	188.6	92.83
5	-1	1	11	4.75	4.75	79.5	78.9	84.87
6	-1	1	9	4.75	4.75	81.5	80.2	89.36
7	-1	1	9	2.25	2.25	86.5	280.2	87.64
8	1	1	8	1	1	90	424.6	80.68
9	-1	1	11	2.25	4.75	82	58.7	86.02
10	-1	1	11	4.75	2.25	82	405.1	86.04
11	1	1	12	6	6	76	200.2	84.22
12	1	1	12	1	1	86	1116	54.67
13	1	1	8	6	1	85	333.3	94.68
14	1	1	8	6	6	80	488.7	86.71
15	-1	1	9	2.25	4.75	84	75.3	85.4
16	1	1	12	1	6	81	75.2	80.91
17	0	1	10	3.5	3.5	83	249.9	96.37
18	1	1	12	6	1	81	756	88.95
19	0	1	10	3.5	3.5	83	226.5	91.47

Table 4. Statistical analysis results of the optimal models in Particle diameter (Y_1) and EE (Y_2)

Term	Particle diameter (Y_1)		EE (Y_2)	
	coefficient	*p*	coefficient	*p*
X_1	5560.64	*	-156.961	*
X_2	114.082	*	-2.02885	*
X_3	254.952	*	-0.419367	*
X_4	52.6339	*	-0.552567	*
X_1X_2	-77.9392	0.000	2.55453	0.002
X_1X_3	-101.549	0.000	2.30194	0.004
X_1X_4	-66.2916	0.000	1.87453	0.007
X_2X_3	16.9219	0.001	-0.963576	0.001
R^2(%)	95.7		86.55	
Lack of fit	0.150		0.275	

Table 5. Evaluation of DoE.

Term	Particle diameter (nm)	EE (%)	Accuracy of diameter (%)	Accuracy of EE (%)
Formula 1 (measured)	285.3	87.43	104.9	101.6
Formula 1 (predicted)	271.9	86.06		
Formula 2 (measured)	177.7	89.67	103.0	99.9
Formula 2 (predicted)	172.5	89.78		

ABSTRACT

A Study on the development of platform technology to Enhance bioavailability of drug

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The aim of this study is to develop a drug platform for oral administration of rotigotine, that marketed and used as an antiparkinsonian transdermal patch, by develop the SLNs (Solid lipid nanoparticles) technology based formulation that can avoid the first-pass effect and increase the bioavailability through lipid-based absorption pathway.

Parkinson's disease (PD) is a disease caused by loss of dopaminergic neurons in the substantia nigra. As a primary treatment, dopamine agonists are considered. Since rotigotine selectively acts on dopamine receptors, especially D_3 receptors, it is being seen as a promising antiparkinsonian drug.

But rotigotine is a poorly soluble drug and is known to be a drug that has a low bioavailabilty due to metabolism caused by the hepatic first-pass effect when administered orally.

In addition, in the case of the presently-marketed the transdermal patch - Neupro®, skin reaction at the attachment site is reported to be a common side effect, and there is inconvenience because it must be applied to another site once a day. Therefore, if it is developed as an oral formulation which can increase the bioavailability, the above disadvantages can be overcome and the patient's compliance with the medicines can be improved. In order to improve the bioavailability of rotigotine, the SLN technology - based drug delivery system, which can avoid the first-pass effect by transferring drugs to

the lymphatic system through the pathway such as Peyer's patch that involved in the absorption and migration in lipid-soluble compounds in the small intestine.

As a solid lipid, Precirol® ATO 5 was used because lipid such as the long chained triglyceride was advantageous to promote uptake to the lymphatic system, and TPGS and Solutol HS 15 were used as surfactants and emulsifiers. They have the effect of inhibiting the efflux by P-gp protein upon absorption of the drug and enhancing the bioavailability by facilitating the migration of poorly soluble and lipophilic substances to the lymphatic system.

The design of experiment (DoE) was applicated to find out the optimum composition. In this study, mixture designs were used among various methods of DoE and optimization process was performed through the use of an Extreme vertices design.

Physicochemical properties of rotigotine-loaded SLN prepared with optimal composition were confirmed by measuring particle diameter, PDI, EE and zeta potential. And in vitro release experiments were performed to determine if sustained release was properly controlled.

In this study, the optimal formulation was designed by analyzing the effects of the components of the formulation in a more scientific way by applying the experimental design method. Based on these results, we have developed a formulation that can maintain the concentration of rotigotine at a high level for a long time and improve the bioavailability when administered orally. In addition, it is thought to be universally applicable to other drugs which have been developed only by the route of administration other than orally because of poor solubility and low bioavailability.

Keywords: Solid lipid nanoparticles (SLNs), Rotigotine, Parkinson’s disease, Design of experiments (DoE), Extreme vertices design, Optimization , Drug delivery, Platform, Oral route, Particle diameter, EE