Fabrication and optimization of camptothecin loaded Eudragit S 100 nanoparticles by Taguchi L4 orthogonal array design

Manikandan Mahalingam, Kannan Krishnamoorthy
Department of Pharmacy, Annamalai University, Chidambaram, Tamil Nadu, India

Abstract

Introduction: The objective of this investigation was to design and optimize the experimental conditions for the fabrication of camptothecin (CPT) loaded Eudragit S 100. Nanoparticles, and to understand the effect of various process parameters on the average particles size, particle size uniformity and surface area of the prepared polymeric nanoparticles using Taguchi design. Materials and Methods: CPT loaded Eudragit S 100 nanoparticles were prepared by nanoprecipitation method and characterized by particles size analyzer. Taguchi orthogonal array design was implemented to study the influence of seven independent variables on three dependent variables. Eight experimental trials involving seven independent variables at higher and lower levels were generated by design expert. Results: Factorial design result has shown that (a) except, β-cyclodextrin concentration all other parameters do not significantly influenced the average particle size (R1); (b) except, sonication duration and aqueous phase volume, all other process parameters significantly influence the particle size uniformity; (c) all the process parameters does not significantly influence the surface area. Conclusion: The R1, particle size uniformity and surface area of the prepared drug-loaded polymeric nanoparticles were found to be 120 nm, 0.237 and 55.7 m2/g and the results were good correlated with the data generated by the Taguchi design method.

Key words: Camptothecin, Eudragit S 100, nanoprecipitation, polymeric nanoparticles, Taguchi orthogonal array design

INTRODUCTION

Camptothecin (CPT), a plant alkaloid isolated from Camptotheca acuminata[1] has been reported to possess promising anticancer activity that targets the nuclear enzyme topoisomerase I and inhibits the relegation of the cleaved DNA strand, resulting in tumor cell death.[2,3] Despite of the prominent antitumor activity toward a wide range of experimental tumor’s, poor solubility in water and in physiologically acceptable organic solvents presents a serious obstacle in the practical use of potent CPT.[4]

One way to improve the solubility of CPT is to change the lactone form to the carboxylate form, which leads to less activity and more unwanted toxicity.[5,6] Now a days, specifically designed techniques and dosage forms have been evaluated to overcome their hydrophobic and unstable characteristics of the CPT.[7] Therefore, to improve the solubility of CPT, the lactone form was incorporated into nanoparticles.[8]

Several approaches have been implemented to enhance the aqueous solubility of hydrophobic drugs, which including liposomes, micelles, nanoemulsions, co-crystallization, pH adjustment, polymeric nanoparticles, solid lipid nanoparticles (SLN), super-critical fluid process, dendrimers, carbon nanotubes and peptide-protein nanotubes are still under investigation for convenient drug deliver.[9,10] Among all the approaches, polymeric nanoparticles are one of the most popular method used due to its easy production and process diversity into the required characteristics for the design of suitable drug delivery systems.[11]

Nanomedicine formulation depends on the choice of suitable polymeric system. These drug nanof ormulations (nanodrug) are superior to traditional medicine with respect to control release, targeted delivery and therapeutic impact.[12] The size and size distributions of nanoparticles are important to determine their
interaction with the cell membrane and their penetration across the physiological drug barriers. The size of nanoparticles for crossing different biological barriers is dependent on the tissue, target site and circulation.\(^{[13]}\)

Polymeric nanoparticles possess some significant advantages over other approaches which includes:
a. significant size reduction leading to the improvement in the solubility,
b. Providing stability to the encapsulated drug,
c. Choice of various route of administration,
d. Reduced side-effect of the drug,
e. Ability to target the drug at the specific site.\(^{[14]}\)

However, there are various methods used for the preparation of polymeric nanoparticles such as desolvation, dialysis, ionic gelation, nanoprecipitation, solvent evaporation, salting out, spray drying and supercritical fluid.\(^{[15]}\) However, nanoprecipitation is the most convenient and economical technique to fabricate polymeric nanoparticles.\(^{[12,6,17]}\)

Though, it is a simple technique the quality of the prepared polymeric nanoparticles is influenced by many process and formulation parameters. The average particle size (R1), particle size uniformity and surface area are influenced by various parameters such as concentration of drug, concentration of Eudragit S 100, concentration of β-cyclodextrin (CD), concentration of poloxamer 188, volume of organic phase, volume of aqueous phase and sonication duration.

The interrelationships between the parameters are complex. For this reason, optimization of the factors, which influence particle size and particle distribution, is an expensive and time-consuming task. Hence, the analysis using conventional experimental methods is inefficient.\(^{[18]}\) Therefore, the Taguchi robust design method was used in this research. The Taguchi method is a combination of mathematical and statistical techniques used in an empirical study, which is economical for optimization of complicated processes.\(^{[19]}\)

Various studies have been carried out to enhance the solubility of CPT. Tong and Cheng prepared a CPT-polylactide conjugate nanoparticles using the nanoprecipitation method to improve the solubility of the CPT.\(^{[20]}\) Swaminathan et al., developed a CD-based nanosponges to increase the solubility of poorly soluble CPT and also to protect the labile groups and control the release.\(^{[21]}\) Fan et al., prepared an alpha, beta-poly ([N-carboxybutyl]-L-aspartamide)-CPT conjugated nanomicelle in order to enhance the solubility of CPT.\(^{[22]}\) Minelli et al., demonstrated β-CD nanosponge to increase the solubility of CPT and to protect from degradation.\(^{[23]}\)

The objective of this investigation was to design and optimize the experimental conditions for the fabrication of CPT loaded Eudragit S 100 nanoparticles and to understand the effect of various parameters on the average particles size, particles size uniformity and surface area of the prepared nanoparticles applying Taguchi orthogonal array (OA) design with an L4 (2^4) OA.

**MATERIALS AND METHODS**

Camptothecin was commercially purchased from S. M Herbals, India. β-CD and Poloxamer (Grade 188) were procured from sigma Aldrich, India. Eudragit S 100 was obtained from Evonik Industries, India. All other chemicals and reagents used were of analytical grade.

**Development of camptothecin loaded Eudragit S 100 nanoparticles**

Camptothecin loaded Eudragit S 100 nanoparticles were prepared by nanoprecipitation method. Briefly, a specified quantity of CPT and Eudragit S 100 (anionic polymer) were dissolved in specified quantity of dimethyl sulfoxide (DMSO) and sonicated (40 kHz, Lark, India) for 5 min to ensure complete dissolution. Prepared organic phase was then emulsified with specific volume of aqueous phase containing poloxamer 188 (nonionic surfactant) and β-CD (Stabilizer) under sonicator (Lark, India) at 40 kHz for specified duration and polymeric nanoparticles are formed spontaneously.\(^{[24]}\)

Taguchi OA design was implemented to study the influence of independent variable such as concentration of drug (a), concentration of Eudragit S 100, (b) β-CD (c) and poloxamer 188, (d) volume of organic phase (e) and aqueous phase (f) and sonication duration (g) on the dependent variables such as R1, particle size uniformity (R2) and surface area (R3) of the prepared nanoparticles. Hence, Taguchi factorial design was used to optimize the process parameter at lower and higher level [Table 1]. The particle size is widely recognized as a critical attribute in determining the overall performance of the formulations. The role of particle size has become increasingly important in the case of poorly water soluble drugs. The surface area analysis was carried out to check the particles for agglomeration. The R1, particle size uniformity and surface area of the particles can affect the product performance, stability and appearance of end product. Considering these parameters, the dependent variables were selected.\(^{[25]}\) Eight experimental trials [Table 2] involving seven independent variables at higher and

| Factors | Process parameters | Levels |
|---------|--------------------|--------|
| \(X_1\) | Drug concentration | 10 mg 12.5 mg |
| \(X_2\) | Eudragit S 100 concentration | 100 mg 125 mg |
| \(X_3\) | β-CD concentration | 50 mg 62.5 mg |
| \(X_4\) | Poloxamer 188 concentration | 100 mg 125 mg |
| \(X_5\) | Organic phase volume | 10 ml 12.5 ml |
| \(X_6\) | Aqueous phase volume | 20 ml 25 ml |
| \(X_7\) | Sonication duration | 50 min 70 min |

\(β\)-CD: β-cyclodextrin
lower levels were generated using Design-Expert® Version 9 (Stat-Ease, Inc., Minneapolis, USA).

**Experimental design by Taguchi method**

The Taguchi experimental design was selected to investigate the effect of different parameters on the mean and variance of the process performance and to obtain an optimal, well-functioning process. The parameter of the Taguchi design generally includes the following steps:

1. Identifying the objective of the experiment,
2. Identifying the quality characteristic (performance measure) and its measurement systems,
3. Determining the factors that may influence the quality characteristic and their levels,
4. Selecting the appropriate OAs and assigning the factors at their levels to the OA,
5. Conducting the test described by the trials in the OA,
6. Analyzing the experimental data using the analysis of variance (ANOVA) to evaluate, which factors are statistically significant and finding the optimum levels of factors and
7. Verifying the optimal design parameters through confirmation experiment.[19,26]

The Taguchi method uses a statistical measure of performance called signal-to-noise (S/N) ratio, which was used in this work to evaluate the quality of results. Both mean and variability are called signal-to-noise (S/N) ratio, which was used in this work. The Taguchi experimental design was selected to investigate the effect of different parameters on the mean and variance of the process performance and to obtain an optimal, well-functioning process. The parameter of the Taguchi design generally includes the following steps:

The experimental design by Taguchi method, the standard OA, namely L4 that reduces the number of experiments to 8 was used. The designed L4 is an array of 8 experiments with the specified combination of levels.[28] This number of experiments to 8 was used. The designed L4 is an array, OA: Orthogonal array.

**Fabrication of camptothecin loaded Eudragit S 100 nanoparticles**

Camptothecin loaded Eudragit S 100 nanoparticles were prepared by nanoprecipitation method as per the scheme and the observed responses of Taguchi design are shown in Table 3. About, 10 mg of drug along with 100 mg of Eudragit S 100 were dissolved in 10 ml of DMSO. The prepared organic phase was transferred at once into 500 ml beaker containing 62.5 mg of β-CD, 125 mg of poloxamer 188 and 25 ml of distilled water under sonication (Lark, India) at 40 kHz for 50 min.

The average particles size, particle size uniformity and surface area of the prepared polymeric nanoparticles were measured based on laser light scattering principle using Mastersizer (Malvern Instruments, UK). Briefly, prepared CPT loaded Eudragit S 100 nanoparticles formulation was added drop-wise in to the water maintained in the sample dispersion unit of particle size analyzer, where the nanoparticles scattered using single shaft pump and stirrer and re-circulated continuously around the measurement zone of the particle size analyzer. The surface morphology of the optimized trial was determined by transmission electron microscopy (TEM). TEM is an excellent tool for characterizing the size of nanoparticles.[29] The prepared CPT loaded polymeric nanoparticles were dropped onto formvar-coated copper grids and air dried. The samples were then negatively stained with 1% uranyl acetate for 10 min and air dried again. The samples were then imaged using TEM (Hitachi H7500, India) at 20,000 magnifications.[30]

Average particle size and surface area determine the performance including solubility, dissolution, stability, circulation half-life, cellular uptake, drug release and bio-distribution. Hence, R1 <200 nm and surface area above 50 m²/g are required for maximum performance of the prepared polymeric nanoparticles. Similarly, particle size uniformity determines the consistency of performance of the prepared polymeric nanoparticles. Particle size uniformity between 0.1 and 0.25 indicates narrow distribution and value above 0.5 indicates a broad distribution.[31,32]

**RESULTS AND DISCUSSION**

**Development of camptothecin loaded Eudragit S 100 nanoparticles**

Camptothecin loaded Eudragit S 100 nanoparticles were developed using the nanoprecipitation method. During nanoprecipitation method, addition of organic phase in to the aqueous phase leads to rapid miscibility of DMSO with water results in spontaneous growth of nanoparticles, which
is initially controlled by sonication, followed by adsorption of Eudragit S 100, which acts as the barrier and inhibits the further growth of nanoparticles. Prepared polymeric nanoparticles were characterized for R1, particle size uniformity and surface area [Table 4]. Regardless of its simplicity, nanoprecipitation method involves many processes, which influence the quality of nanoparticles. Hence, we have implemented Taguchi factorial design with an L4 OA to optimize the process parameters.

**Effect of process parameters on the average particle size**

It is essential to fabricate polymeric nanoparticles with least R1 in view of the fact that the R1 of the prepared polymeric nanoparticles decides the recital such as solubility, dissolution, drug release and cellular uptake.[31,32] ANOVA has shown that the process parameters have a significant effect (Prob. F, 0.0430) on the R1 [Table 5]. Except, β-CD concentration all other parameters do not significantly influence the average particle size [Figure 1]. In figure, orange color indicates the parameter has a positive effect and blue color column indicates the negative effect on the average particle size. The white column inside the orange columns indicates that the parameters have a significant effect on the average particle size.

Cyclodextrins are a family of cyclic oligosaccharides with a hydrophilic outer surface and a lipophilic central cavity. In the pharmaceutical industry, they are used as complexing agents to increase the aqueous solubility of poorly soluble drugs and to increase their bioavailability and stability.[34] Camptothecin nanoparticles were formulated with and without β-CD, the formulation without β-CD showed increased particles size of nanoparticles when compared with that of the formulation with β-CD. During the formulation process, the β-CD forms a complex with the CPT, which in turn decreases the particle size of the nanoparticles.

The mechanism of the process is as follows, when the drug is added to the aqueous solution containing a polymer without β-CD, crystallization starts due to the insoluble nature of the drug. When the β-CD is added to the drug, it prevents the crystallization of the drug by forming a complex with the drug, thereby decreasing the size of the particles.

Process parameters such as β-CD concentration has favorable effect on the average particle size whereas drug concentration, Eudragit S 100 concentration, poloxamer 188 concentration, aqueous phase volume, organic phase volume and sonication duration have inverse relationship with the average particle size [Figure 1]. Moreover, the observed average particle size was comparable with predicted values of Taguchi factorial design [Table 6].

Cirpanli *et al.*, developed a Nanoparticulate delivery systems with either amphiphilic CDs, poly (lactide-co-glycolide) or poly-E-caprolactone in order to maintain the active lactone form and prevent the drug from hydrolysis with nanoprecipitation technique and the mean particle sizes obtained was 130-280 nm.[34] Martins *et al.*, formulated CPT-loaded SLN, by hot, high-pressure homogenization and the mean particle sizes was ≤200 nm.[35] As per the reported observation, the particle size was in the range of 130-200 nm, the results of our study showed

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**Table 4: Characterization of prepared Eudragit S 100 based polymeric nanoparticles**

| Trials | Average particle size (nm ± SD) | Particle size uniformity (± SD) | Surface area (m²/g ± SD) |
|--------|---------------------------------|---------------------------------|--------------------------|
| 1      | 161±0.73                        | 0.566±0.01                      | 51.6±0.52                |
| 2      | 148±0.57                        | 0.441±0.01                      | 51±0.45                  |
| 3      | 190±0.95                        | 0.761±0.02                      | 52.5±0.56                |
| 4      | 387±1.25                        | 2.34±0.04                       | 50.6±0.47                |
| 5      | 8788±1.89                       | 19.1±0.03                       | 21.5±0.57                |
| 6      | 120±0.37                        | 0.273±0.01                      | 55.7±0.49                |
| 7      | 142±0.50                        | 0.439±0.01                      | 53.6±0.54                |
| 8      | 168±0.74                        | 0.629±0.02                      | 51.6±0.48                |

**Table 5: ANOVA of average particle size, particle size uniformity and surface area**

| Variables         | Source         | Sum of square | df | Mean of square | F ratio | Prob. >F* |
|-------------------|----------------|---------------|----|----------------|---------|-----------|
| Average particle size | Model          | 0.00002603    | 1  | 0.00002603    | 6.55    | 0.0430    |
|                   | Residual       | 0.00002386    | 6  |                |         |           |
|                   | C. Total       | 0.00004989    | 7  |                |         |           |
| Particle size uniformity | Model          | 1.95          | 5  | 0.39           | 79.23   | 0.0125    |
|                   | Residual       | 0.009863      | 2  | 0.004932       |         |           |
|                   | C. Total       | 1.96          | 7  |                |         |           |
| Surface area      | Model          | 0.000         | 0  |                |         |           |
|                   | Residual       | 0.0006592     | 7  | 0.0009418      |         |           |
|                   | C. Total       | 0.0006592     | 7  |                |         |           |

ANOVA: Analysis of variance
Effect of process parameters on the particle size uniformity

It is essential to fabricate polymeric nanoparticles with the particle size uniformity between 0.1 and 0.25 in view of the fact that the particle size uniformity determines the consistency of the prepared polymeric nanoparticles.\cite{11,12} Hence, ANOVA has shown that the process parameters have a significant effect (Prob. F, 0.0125) on the particle size uniformity [Table 5]. Except, sonication duration and aqueous phase volume, all other process parameters significantly influenced the particle size uniformity [Figure 2]. In figure, orange color indicates the parameter has a positive effect and blue color column indicates the negative effect on the particle size uniformity. The white column inside the orange columns and the blue color indicates that the parameters have a significant effect on the particle size uniformity.

Process parameters such as drug concentration, Eudragit S 100 concentration, β-CD, poloxamer 188 concentration and organic phase volume concentration has favorable effect on the particle size uniformity whereas sonication duration and aqueous phase volume have inverse relationship with the particle size uniformity [Figure 2]. Moreover, the observed particle size uniformity was comparable with predicted values of Taguchi factorial design [Table 6].

Effect on process parameters on the surface area

It is essential to fabricate polymeric nanoparticles with the surface area above 50 m²/g in view of the fact that the surface area is responsible for the biological effect of the prepared polymeric nanoparticles.\cite{11,12} All the process parameters does not significantly influence the surface area [Figure 3]. In the figure, orange color indicates the parameter has a positive effect and blue color column indicates the negative effect on the surface area. The white column inside the orange columns and the blue color indicates that the parameters have a significant effect on the surface area.

Process parameters such as drug concentration, Eudragit S 100 concentration, β-CD, poloxamer 188 concentration, organic phase volume, aqueous phase volume concentration and sonication duration have inverse relationship with the surface area [Figure 3]. Moreover, the observed surface area was comparable with predicted values of Taguchi factorial design [Table 6]. The optimized formula (with desirability: 0.968) for the fabrication of CPT loaded Eudragit S 100 nanoparticles was displayed in RAMPS format [Figure 4]. CPT loaded Eudragit S 100 nanoparticles were prepared using

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**Figure 2**: Taguchi plot for the particle size uniformity

**Figure 3**: Taguchi plot for the surface area

**Figure 4**: RAMPS format of optimized formula for the fabrication of Eudragit S 100 based nanoparticulate drug delivery system
the final optimized formula form Taguchi factorial design. About, 10 mg of drug along with 100 mg of Eudragit S 100 were dissolved in 10 ml of DMSO. The prepared organic phase was transferred at once into 500 ml beaker containing 62.5 mg of β-CD, 125 mg of poloxamer 188 and 25 ml of distilled water under sonication (Lark, India) at 40 kHz for 50 min. Prepared drug loaded polymeric nanoparticles were characterized for average particle size, particle size uniformity and surface area. The characterized results were summarized in Table 7 and Figure 5.

The average particle size, particle size uniformity and surface area of prepared polymeric nanoparticles formulations ranged from 120 nm to 8788 nm, 0.273-19.1 to 55.7-21.5 m²/g, respectively. The increase in particle size was observed with the decrease in surface area. This is due to the droplet solidification resulting in the aggregation of particles with increased particle size.[36] The particles were found to have a uniform size when the average particle size is low and vice versa.

Surface morphology decides the basic function of particles, degradation, release of drug from the polymer matrix, transport of particles in the body, internalization of drug. Prepared drug loaded polymeric nanoparticles were imaged using TEM and found to be spherical in shape [Figure 6].

The finding shows that β-CD concentration had influence the particles size of the nanoparticles as drug concentration [Figure 1], Eudragit S 100 concentration, β-CD concentration, poloxamer concentration and organic phase volume had produced significant effect on the particle size uniformity [Figure 2]. This may be due to the difference in the formulation process, where we have formulated using sonicator.

**CONCLUSION**

In the present investigation, Taguchi L4 OA design method was used to optimize the experimental conditions for the fabrication of CPT loaded Eudragit S 100 nanoparticles using nanoprecipitation method. Eight experimental trials involving seven independent variables at higher and lower levels were generated by design expert. Effect of seven process parameters on average particle size, particle size uniformity and surface area were studied. Average particle size <200 nm, particle size uniformity between 0.1 and 0.25 and surface area above 50 m²/g were used to evaluate the quality of the prepared nanoparticles. The optimized formula comprising 10 mg of drug along with 100 mg of Eudragit S 100, 10 ml of DMSO, 62.5 mg of β-CD, 125 mg of poloxamer 188 and 25 ml of distilled water under sonication duration of 50 min were implemented for the fabrication of the CPT loaded polymeric nanoparticles. The prepared nanoparticles were characterized for the average particle size, particle size uniformity and surface area and the experimental results were found to be in good agreement with the predicted data analyzed by the Taguchi design method.

| F | Average particle size | Particle size uniformity | Surface area |
|---|---|---|---|
| 1 | 0.006211 | 0.007086 | 1.33 | 1.37 | 0.019 | 0.023 | -12.35 | -2.92 | -17.39 |
| 2 | 0.006757 | 0.007086 | 1.51 | 1.54 | 0.020 | 0.023 | -4.64 | -1.95 | -13.04 |
| 3 | 0.005263 | 0.003478 | 1.15 | 1.18 | 0.019 | 0.023 | -51.32 | -2.54 | -17.39 |
| 4 | 0.002584 | 0.003478 | 0.65 | 0.69 | 0.020 | 0.023 | -25.70 | -5.80 | -13.04 |
| 5 | 0.0001138 | 0.003478 | 0.23 | 0.19 | 0.047 | 0.023 | -96.73 | 21.05 | 104.34 |
| 6 | 0.008333 | 0.007086 | 1.91 | 1.87 | 0.019 | 0.023 | 17.60 | 2.14 | -21.74 |
| 7 | 0.007042 | 0.007086 | 1.51 | 1.48 | 0.019 | 0.023 | -0.63 | 2.03 | -17.39 |
| 8 | 0.005952 | 0.003478 | 1.26 | 1.23 | 0.019 | 0.023 | 71.13 | 2.44 | -17.39 |

F: Formulation, %RE: % Relative error
Table 7: Average particle size, particle size uniformity and surface area of prepared drug loaded polymeric nanoparticles

| Trials | Average particle size (nm ± SD) | Particle size uniformity (± SD) | Surface area (m²/g ± SD) |
|--------|--------------------------------|--------------------------------|--------------------------|
| 1      | 150±0.19                       | 0.210±0.01                      | 51.7±0.42                |

SD: Standard deviation

REFERENCES

1. Wall ME, Wani MC, Cook CE, Palmer KH, Mcphail AT, Sim GA. Plant antitumor agents. I. Isolation and structure of camptothecin a novel alkaloidal leukemia and tumour inhibitor from Camptotheca acuminata. J Am Chem Soc 1966;88:3888-90.

2. Jaxel C, Kohn KW, Wani MC, Wall ME, Pommier Y. Structure-activity study of the actions of camptothecin derivatives on mammalian topoisomerase I: Evidence for a specific receptor site and a relation to antitumor activity. Cancer Res 1989;49:1465-9.

3. Rothenberg ML. Topoisomerase I inhibitors: Review and update. Ann Oncol 1997;8:837-39.

4. Kuni R, Onishi H, Machida Y. Preparation and antitumor characteristics of PLA/(PEG-PPG-PEG) nanoparticles loaded with camptothecin. Eur J Pharm Biopharm 2007;67:9-17.

5. Ding XQ, Wang AX, Kong QY, Chen HZ, Chen Y. Anticancer effect of hydroxycamptothecin on oral squamous carcinoma cell line. Ai Zheng 2002;21:388-91.

6. Kim TE, Park SY, Hsu CH, Dutschman GE, Cheng YC. Synergistic antitumor activity of taxotax and camptothecin in selected human cancer cell lines. Mol Pharmacol 2004;66:285-92.

7. Zhao YX, Hua HY, Chang M, Liu WJ, Zhao Y, Liu HM. Preparation and cytotoxic activity of hydroxycamptothecin nanosuspensions. Int J Pharm 2010;392:64-71.

8. Williams J, Lansdown R, Sweltert R, Romanowski M, LaBell R, Ramaswami R, et al. Nanoparticle drug delivery system for intravenous delivery of topoisomerase inhibitors. J Control Release 2003;91:167-72.

9. Ajazuddin RT, Giri TK, Tripathi DK, Jain V, Alexander A. An exhaustive review on solubility enhancement for hydrophobic compound by possible application of novels techniques. Trends Appl Sci Res 2012;7:596-619.

10. Chaudhary A, Nagaich U, Gulati N, Sharma VK, Khosa RL, Partapur MU. Enhancement of solubilization and bioavailability of poorly soluble drugs by physical and chemical modifications: A recent review. J Adv Pharm Educ Res 2012;2:32-67.

11. Kilicyay E, Demirbilek M, Türk M, Güven E, Hazer B, Denkbas EB. Preparation and characterization of poly (3-hydroxybutyrate-co-3-hydroxyhexanoate) (PHBHHX) based nanoparticles for cancer therapy. J Nanobiotechnology 2010;8:18.

12. Kumari A, Yadav SK, Yadav SC. Biodegradable polymeric nanoparticles based drug delivery systems. Colloids Surf B Biointerfaces 2010;75:1-18.

13. Brannon-Peppas L, Blanchette JO. Nanoparticle and targeted systems for cancer therapy. Adv Drug Deliv Rev 2004;56:1649-59.

14. Moorthi C, Kathiresan K. Application of Plackett-Burman factorial design in the development of curcumin loaded Eudragit EL 100 nanoparticles. Nano Biomed Eng 2013;5:28-33.

15. Sailaja A, Amareeshwar P, Chakravarty P. Different techniques used for the preparation of nanoparticles using natural polymers and their application. Int J Pharm Sci Res 2011;3:45-50.

16. Khayata N, Abdelwaheed W, Chehna MF, Charcosset C, Fessi H. Preparation of vitamin E loaded nanocapsules by the nanoprecipitation method: From laboratory scale to large scale using a membrane contactor. Int J Pharm 2012;423:419-27.

17. Mahalingam M, Krishnamoorthy K. Selection of a suitable method for the preparation of polymeric nanoparticles: Multi-criteria decision making approach. Adv Pharm Bull 2015;5:57-67.

18. Kim KD, Choi DW, Choa YH, Kim HT. Optimization of parameters for the synthesis of zinc oxide nanoparticles by Taguchi robust design method. Colloids Surf A Physicochem Eng Asp 2007;311:170-3.

19. Torkaman R, Soltanieh M, Kazemian H. Optimization of parameters for synthesis of MFI nanoparticles by Taguchi robust design. Chem Eng Technol 2010;33:902-10.

20. Tong R, Cheng J. Controlled synthesis of camptothecin-polyacrylate conjugates and nanoconjugates. Bioconjug Chem 2010;21:111-21.

21. Swaminnathan S, Pastero L, Serpe L, Troffa F, Vavia P, Aquilano D, et al. Cyclodextrin-based nanosponges encapsulating camptothecin: Physicochemical characterization, stability and cytotoxicity. Eur J Pharm Biopharm 2010;74:193-201.

22. Fan N, Duan K, Wang C, Liu S, Luo S, Yu J, et al. Fabrication of nanomicelle with enhanced solubility and stability of camptothecin based on alpha,beta-poly[(N-carboxybutyl)-L-aspartamide]-camptothecin conjugate. Colloids Surf B Biointerfaces 2010;75:543-9.

23. Minelli R, Cavalli R, Ellis L, Pettazzoni P, Troffa F, Ciamporcer E, et al. Nanosponge-encapsulated camptothecin exerts antitumor activity in human prostate cancer cells. Eur J Pharm Sci 2012;47:686-94.

24. Chidambaram M, Krishnasamy K. Modifications to the conventional nanoprecipitation technique: An approach to fabricate narrow sized polymeric nanoparticles. Adv Pharm Bull 2014;4:205-8.

25. Rohrs BR, Amidon GE, Meury RH, Secreast PJ, King HM, Skoug CJ. Particle size limits to meet USP content uniformity criteria for tablets and capsules. J Pharm Sci 2006;95:1049-59.

26. Bendell A, Disney J, Pridmore WA. Taguchi Methods: Applications in World Industry. London: IFS Publications, Springer-Verlag; 1989. p. 19-26.

27. Taguchi G. Introduction to Quality Engineering. Tokyo: Asian Productivity Organization; 1990. p. 103-11.

28. Kamyabi-Gol A, Zebanjarad SM, Sajjadi SA. Fabrication of NiO/SiO2 nanocomposites using sol-gel method and optimization of gelation time using Taguchi robust design method. Colloids Surf A Physicochem Eng Asp 2009;336:69-74.

29. Jung KY, Park BC, Song WY, B.H.O, Eom TB. Measurement of 100-nm polystyrene sphere by transmission electron microscope. Powder Technol 2002;126:255.

30. Champion JA, Katare YK, Mitragotri S. Particle shape: A new design parameter for micro- and nanoscale drug delivery carriers. J Control Release 2007;121:3-9.

31. Xie H, Smith JW. Fabrication of PLGA nanoparticles with a fluidic nanoprecipitation system. J Nanobiotechnology 2010;8:18.

32. Lakshmi P, Ashwini KG. Nanosuspension technology: A review. J Int Pharm Sci 2010;2:35-40.

33. Brannon-Peppas L, Blanchette JO. Cyclodextrins and their pharmaceutical applications. Int J Pharm 2007;329:1-11.

34. Cirpanli Y, Allard E, Passirani C, Bilensoy E, Lemaire L, Calis S, et al. Antitumoral activity of camptothecin-loaded nanoparticles in 9L rat glioma model. Int J Pharm 2011;403:201-6.

35. Martins S, Tho I, Reinold I, Fricker G, Souto E, Ferreira D, et al. Brain delivery of camptothecin by means of solid lipid nanoparticles.
36. Rahman Z, Zidan AS, Habib MJ, Khan MA. Understanding the quality of protein loaded PLGA nanoparticles variability by Plackett-Burman design. Int J Pharm 2010;389:186-94.

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