

Optimal Fabrication of Nano Menthol/PEG Particles by Electro spraying

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Abstract

Background: L-menthol [(1R,3R,4S)-(-)-menthol] is a flavoring that is the main component of mint herb essential oils, especially of the *Mentha piperita* and *Mentha arvensis* species. Its low solubility in aqueous systems makes precise formulation necessary in the final products. Of the methods available for fabrication of nanoparticles for use in pharmaceuticals, electro spraying is easy and requires only one step.

Objective: Electro spraying was used to fabricate menthol/PEG micro/nanoparticles. The experiments used menthol concentrations of 10%, 15% and 20% (wt) and PEG concentrations of 5%, 10% and 15% (wt).

Methods: The effect of menthol and PEG concentration on the morphology of the fabricated particles was investigated using scanning electron microscopy (SEM). Response surface methodology (RSM) was used to determine the best levels for each parameter under optimal conditions.

Results: SEM results revealed that an increase in PEG and menthol concentrations in solution, increased the particle diameters. RSM showed that particle diameter should be calculated as the square root of a function of the first order and cubic forms of menthol and PEG. Optimization results show that the optimal menthol concentration is 10.7% (wt) and PEG concentration is 7.31% (wt). The optimal modeled particle diameter of 1219 nm approached the real test particle diameters (1136 nm). The results indicate that the modeled conditions were appropriate for menthol/PEG electro spray particles.

Conclusion: The results showed that the maximum PEG concentration effects particle diameter because of its polymeric structure. At high menthol concentrations, the percentage of menthol in a droplet was greater than the PEG concentration and some menthol sublimated during drop formation. At low menthol concentrations, PEG covered the menthol and prevented sublimation, decreasing the effect of menthol concentration.

Keywords: Electro spray, Menthol, Micro/nanoparticles, Optimization



Introduction

Electrohydrodynamic atomization is a nanofabrication method with applications in biomedical and pharmaceutical research [1]. Electrospaying is used extensively for fabrication of micro/nano particulate systems used as drug delivery systems. In this process, high electric force is used to atomize a polymer solution containing a drug. Repulsive forces overcome the surface tension and atomize the solution from cannula tip [2]. Application of this technique of fabrication for hydrophobic active pharmaceutical ingredients (API) is a promising field of research for increasing the bioavailability of these drugs.

L-menthol is an herbal-based fragrance primarily used as a cooling agent in the pharmaceutical, food, and cosmetics industry [3]. Its exceptional properties make it attractive for use; however this compound is hydrophobic in nature and its solubility in water is limited by its aromatic structure. Increasing the solubility is a major marketing consideration for this product [7].

Electrospray technique improves the solubility of active compounds. In most cases, its polymeric structure is employed for encapsulation of active compounds. PEG is a hydrophilic/hydrophobic polymer used in the cosmetic industry. Mu et al. [6] used a PLGA/TPGS/PEG tertiary system as an excipient for spray-drying of paxitaxol compound. They showed that the dominant compound on particle surface was PLGA, which possesses hydrophobic characteristics, but that the presence of PEG in the structure of the particles increased solubility and noncrystallinity of the drug particles.

Kawakami et al. [4] used coaxial electrospaying to encapsulate the poorly soluble drug fenofibrate. They used poly (methyl methacrylate) as the shell material for encapsulation. The results showed that fabrication of decreased the particle size and increased the solubility of the compound. In addition, electrospaying decreased crystalline compound formation, which improved solubility and bioavailability.

Researchers also studied optimization of particle production during electrospaying. Zarchi et al. [8] used response surface methodology (RSM) to fabricate encapsulated N-acetylcysteine compound with PLGA. They demonstrated that the operational parameters of flow rate and tip to collector distance had a strong effect on particle diameter and polydispersity. The optimal conditions allowed fabrication of the smallest particle at 122 nm in size. The present study used RSM to fabricate menthol/PEG micro/nanoparticles and investigated the relationship of the response equation and operational solution concentrations.

Materials and Methods

Materials

Menthol crystals having 99.63% purity were prepared in the lab. PEG with a molecular weight of 4000 was purchased from Sepidaj Pharmaceuticals (Iran). Analysis grade ethanol (99% purity) and dichloromethane (DCM) were purchased from Merck (Germany).

Methods

Solution preparation

The clear homogeneous solutions were prepared by dissolving menthol and PEG in a

binary solvent of 1:1 ethanol/DCM. The ethanol/DCM solvent was used because of the ease of evaporation of the solvent and its appropriate electrical conductivity, which was necessary for stable electro spraying. The mixture was stirred at 200 rpm for 12 h at room temperature.

Particle preparation

Each electro spray sample was created using a 5 ml plastic syringe that delivered the solution at a 1 ml/h flow rate at ambient temperatures. The flow rate of the solution was controlled by a syringe pump (JMS SP-500; Japan). A distance from cannula tip to collector of 15 cm was used for particle deposition. The voltage used was 17 kV for charging the sample solutions. Grounded aluminum foil was used for particle collection and was subsequently detached and used for morphological analysis of the particles.

Particle characterization

The particles were sputter-coated with a thin layer of platinum and then their morphology was examined by scanning electron microscopy (SEM; KYKYEM3200; SN: 0056; China) at an accelerating voltage of 10 to 15 kV. The mean diameter of the particles was calculated using Image J software (National Institutes of Health; MD); 100 random measurements from several SEM images were used to obtain the mean values.

Experimental design and statistical analysis

RSM is a statistical and mathematical method for optimization of processes. In this study, the CCD method was used for the experimental design. Two solution parameters were used as variables in the experiment design. Particle diameter was the response. The experimental design is shown in Table 1. All the other parameters were sets derived from pretesting.

Table 1- Central Composite Design in 13 experiment and corresponding response

Run No	Independent Variables		Dependent Variable
	C _{Menthol} (Wt %)	C _{PEG} (Wt%)	Y (nanometer)
1	15	15	3180
2	10	10	1328
3	10	5	1269
4	20	5	1378
5	15	10	3080
6	15	10	3480
7	15	10	3120
8	20	15	4599
9	10	15	3180
10	15	5	1522
11	20	10	4247
12	15	10	2763
13	15	10	2739

A third-order polynomial function was derived to determine the mathematical relationship between response function and the independent variables (X_i : menthol concentration; PEG concentration) as:

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_1 \beta_2^2 X_1 X_2^2 \quad (1)$$

Where Y is the predicted response, β_0 is the function intercept, β_1 and β_2 are the linear coefficients, and $\beta_1 \beta_2^2$ is the cubic effect. A 3D surface plot was used to represent the response-factor relationship (Figure 2).

Results

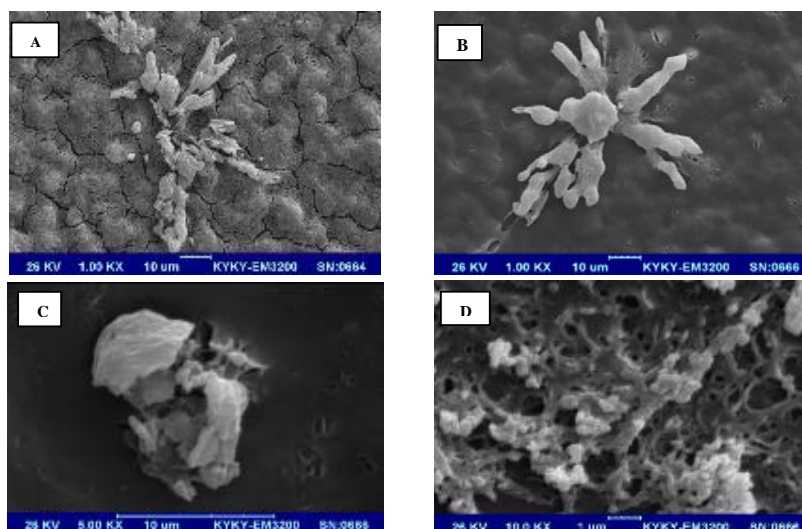
Preliminary studies

A set of preliminary studies were done to optimize the relevant solution and spraying conditions. Studies show that a binary solvent system of ethanol/DCM (1:1) is suitable for

electrospraying. Pretesting showed that the best Taylor cone formation operated at 20 kV and the optimal flow rate was 1 ml/h for the mixed menthol/PEG solution. Increasing the flow rate destabilized the jet and drop formation in the needle tip. It also was observed that increasing the operational voltage between the cannula tip and collector, significantly increased powder formation on the collector surface. Higher voltages also increased the particle diameter.

Particle morphology

The SEM images (Figure 1) revealed that an increase in PEG concentration in solution increased the particle diameter. The images showed the minimum particle size of 1200 ± 30 nm for the 5% (w/v) menthol/DCM solution.



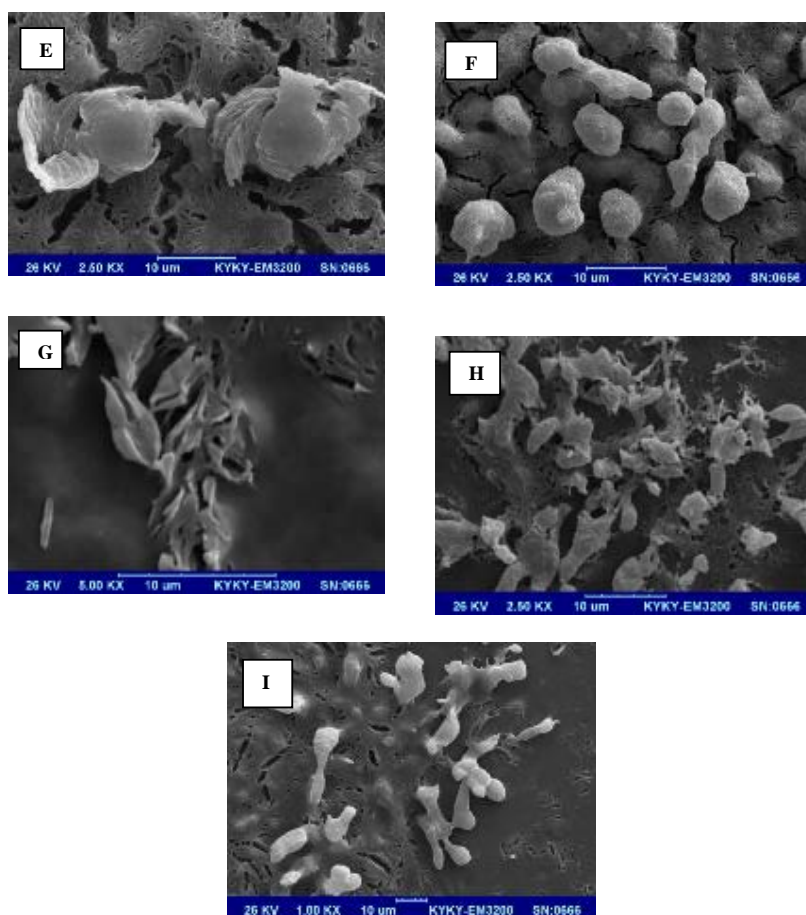


Figure 1- SEM micrograph of electrospayed solution containing different concentration of Menthol and PEG

It can be inferred that the polymeric nature of the PEG resulted in larger particles. The molecular structure of the menthol increased formation of compact particles, indicating that the concentration did not affect particle diameter as did the PEG concentration. The amorphous polymer structure limited menthol crystallization during electrospaying.

Kumar *et al.* [5] used PEG as a dispersant for menthol in a liquid formulation for housefly repellent. Sahoo *et al.* [7] show that the addition of PEG to spray-dried solutions increase fine powder formation and amorphous powder fabrication.

Discussion

This results show that PEG increases the noncrystallinity of the powder by dispersing the polymer molecular chain between the drug molecules during drying. These findings are in agreement with the results of the present study.

Particle diameter optimization

Analysis of the response surface plot and Equation 1 for the solution led to predictions of optimal concentrations of 10.7% for menthol and 7.4% (wt) for PEG. These values are based on minimum particle diameter. Testing using the optimal values showed that

the fabricated particle diameter (1136 nm) was in good agreement with predicted value (1219 nm).

Zarchi et al. [8] reported similar results in their research for polymer concentration. They showed that increasing the polymer concentration increased particle diameter.

They also reported that an increase in flow rate had only a minor effect on particle diameter in electrospraying. The 3D graph of the results (Figures 2, 3) of the present study illustrate that the dependency of particle diameter on PEG concentration is much greater than on menthol concentration.

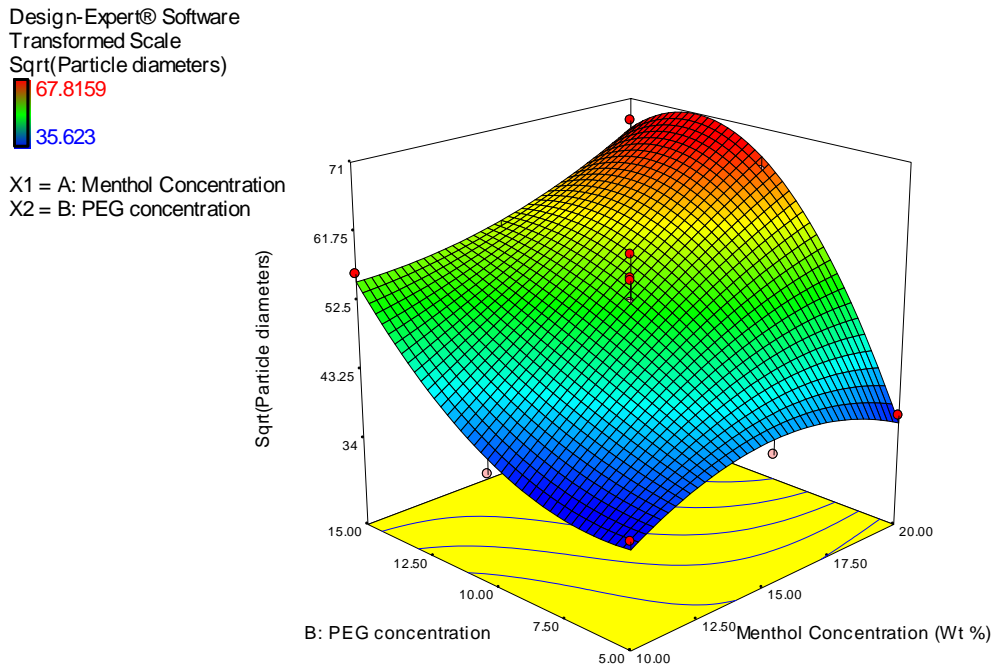


Figure 2- 3D plot of response analysis and its dependency to variables

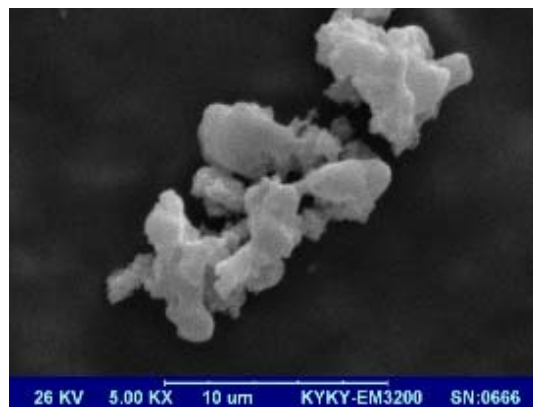


Figure 3- Optimum conditions particle

Conclusion

It can be inferred that menthol/PEG fabrication by electro spraying depends strongly on PEG concentration. Optimized values of each variable were predicted with the

proper response function.

Acknowledgement

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