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RESEARCH ARTICLE

Preparation of Chitosan Nanoparticles by Nano Spray Drying Technology

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SUMMARY

Spray drying is a fast, continuous process, leading to transform of a liquid feed into dry particles. This process is completed in three steps; the atomisation, the mixing of fine droplets with gas stream and the separation and collection of dried powder. Besides, nano-spray technology is a breakthrough innovation in order to obtain submicron particles from a solution. Particle size is a one of the indicator of product quality and performance. The main aim of this study was to determine critical process parameters of nano-spray dryer and to evaluate the effects of these parameters on particle characteristics and operation capacity. Nano-spray dryer B-90 was used. In terms of manufacturing process, orifice, spray capacity and polymer concentration were chosen as critical process parameters and their effects on product characteristics as particle size and polydispersity index and zeta potential were evaluated. Moreover, their effects on operation capacity as product yield and process yield were also evaluated. Process yield was calculated as sprayed sample volume per unit process time. Chitosan was chosen as a model polymer with the concentration of 10 and 20% (w/v). According to results of particle characteristics and operation capacity, smaller orifice leads smaller particle size for final product but lower operation capacity. On the other hand, the sprayed volume showed linear proportion with operational capacity but no effect on particle size. Increased polymer concentration improved the yield of dry powder and showed more positive zeta potentials as it exhibited decreased operation capacity.

Key Words: *Spray dryer, chitosan, nanoparticle, nano-spray dryer B-90*

Nano püskürterek kurutma tekniğinin kullanımı ile kitozan nanopartiküllerinin hazırlanması

ÖZET

Püskürterek kurutma; sıvı maddenin işlem sonunda katı toz haline geldiği hızlı, sürekli bir işlemdir. İşlem; atomizasyon, damlacıkların sıcak gaz ile muamelesi ve katılaştırılan tozun ayrılarak toplanması olmak üzere temelde üç adımda tamamlanmaktadır. Nano boyutlu püskürterek kurutma (Nanospray dryer) teknolojisi ise çığır açan bir yenilik olup çözeltilerden mikron altı boyutlarda partikül eldesini sağlamaktadır. Partikül büyüklüğü, ürünün kalitesini ve performansını etkileyen en önemli parametrelerden birisidir. Bu çalışmanın temel amacı; nano boyutlu partikül eldesine olanak tanıyan püskürterek kurutma cihazına ait kritik işlem parametrelerini belirlemek ve üretim işleminde kullanılan bu parametrelerin partikül karakteristikleri ve cihaz kapasitesi üzerine etkilerini değerlendirmektir. Cihaz olarak Nano-Spray Dryer B90 cihazı kullanılmıştır. Üretim prosesi açısından cihaza ait püskürtme başlığı por çapı, püskürtme kapasitesi ve polimer konsantrasyonu kritik işlem parametreleri olarak düşünülmüştür. Bunların; ürünün partikül karakteristikleri açısından partikül büyüklüğü ve dağılımı yanı sıra zeta potansiyelleri üzerine etkileri değerlendirilmiştir. Ayrıca işlem parametrelerinin uygulama kapasitesine etkileri de elde edilen katı ürün üzerinden işlem verimi olarak değerlendirilmiştir. İşlem verimi; birim işlem süresi boyunca püskürtülen örnek hacmi üzerinden incelenmiştir. Model madde olarak kitozan polimeri %10 ve %20 konsantrasyonlarında kullanılmıştır. Partikül karakteristikleri ve uygulama kapasitesi açısından sonuçlar değerlendirildiğinde; küçük püskürtme başlığı por çapı ile sonuç ürün partikül büyüklüğünün küçüldüğü, ancak bunun yanı sıra uygulama kapasitesinin azaldığı gözlenmiştir. Püskürtme hacminin; işlem verimi ile arasında orantısallık olduğu fakat partikül büyüklüğü üzerine bir etkisi olmadığı görülmüştür. Polimer konsantrasyonundaki artış ise zeta potansiyeli ve geri elde edilen katı ürün verimini artırırken, işlem verimini azaltmıştır.

Anahtar kelimeler: *Püskürterek kurutma, kitozan, nanopartikül, nano-spray dryer B-90*

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INTRODUCTION

Preparing nano-sized particles; the surface area, improve solubility, dissolution rate and thus oral bioavailability can be increased. Nanoparticles defined as ultrafine particles, ranging in size from 1nm to 1μm. (Lockman *et al*., 2002; Singh and Lillard, 2009; Kumar *et al*., 2014). Nanoparticles can be formulated by two different approaches; bottom up and top down. The bottom-up approach is relevant with more complex nanoscale assemblies created by molecular or atomic components, whereas the top–down approach reveals nanoscales structures starting from larger dimensions and reducing them into nano-scales. Spray drying is one of the top-down approaches (Biswas et. al., 2012; Kumar et. al., 2015; Wais et. al., 2016). Spray drying is a fast, continuous process, providing the transformation of a liquid feed into dry powders and also has possible control over particle size. It consists of three main stages; beginning with the atomization of the liquid feed into small droplets via atomizer, going forward with the mixing of fine droplets with a drying gas stream to evaporate the liquid phase, resulting with the separation and collection of dried particles (Liu *et al*. 2015; Kulkarni *et al*., 2016; Wan and Yang, 2016). In many conventional spray dryers, the fluid is reached to drying chamber via a peristaltic pump through an atomizer or nozzle. The atomization stage is performed by centrifugal, pressure or kinetic energy. Then, thanks to heated gas stream, fast solvent evaporation leading to the formation from fine droplets to dry particles occurs. At the end of these stages, dry particles are separated from the drying gas into a glass cyclone collector (Sosnik and Seremeta, 2015).

Particle size is a one of the indicator of product quality and performance. It influences several parameters from appearance to dissolution rate and effectiveness. Generally the smaller particles lead to faster dissolution. Nano spray drying technology is a breakthrough innovation in order to obtain submicron particles from a solution. In this study, it was aimed to find out some critical parameters of nano spray dryer and to evaluate their influences on manufacturing process, particle characteristics and capacities. Chitosan was chosen as a model polymer. It was dissolved in water with glacial acetic acid at 10 and 20% (w/v). As critical operation parameters, the hole size of spray caps (nozzles), spray capacities and various polymer concentrations were chosen and eight different formulations were determined then prepared with using spray drying technology using various parameters.

MATERIALS AND METHODS

Materials

Chitosan from shrimp shells (75% deacetylated, at around 200cP viscosity) was purchased from Sigma Aldrich, Germany. Glacial acetic acid was obtained from Fischer Scientific. Nano spray dryer B-90 obtained from Buchi, Germany. Microscopic image was taken by high contrast Transmission Electron Microscope (TEM), Tecnai G2 Spirit Biotwin from FEI, USA.

Methods

Sample preparation

Chitosan was chosen as a model polymer. Chitosan was directly dissolved in water with glacial acetic acid without the need to milling. Chitosan in 1% (v/v) aqueous glacial acetic acid was left under stirring at 1000 rpm at room temperature for 24 hours. Two different chitosan solution at the concentration of 10 and 20% (w/v) were prepared. Final solutions were filtered with membrane filter before spraying with Nano Spray Dryer B-90. For the TEM image, dryed powder was dispersed in water then a small drop of it was pipetted onto a TEM grid and dried at room temperature.

The Nano Spray Dryer B-90 technology

The nano spray dryer B-90 is the last generation of laboratory scale spray dryer. This novel technology differs from conventional spray dryer in respect to spray head, heating system and particle collector. A complete diagram of it is given in Figure 1.

Atomization is achieved by centrifugal pressure or kinetic energy for conventional spray dryer, after the fluid is fed to spray head through a peristaltic pump. On the other hand, for Nano Spray Dryer B-90, a vibrating mesh technology is utilized to generate fine droplets. Spray head based on piezoelectric crystal incorporate with a small perforate membrane in spray cap which can vibrate upwards and downwards (Figure 2.)

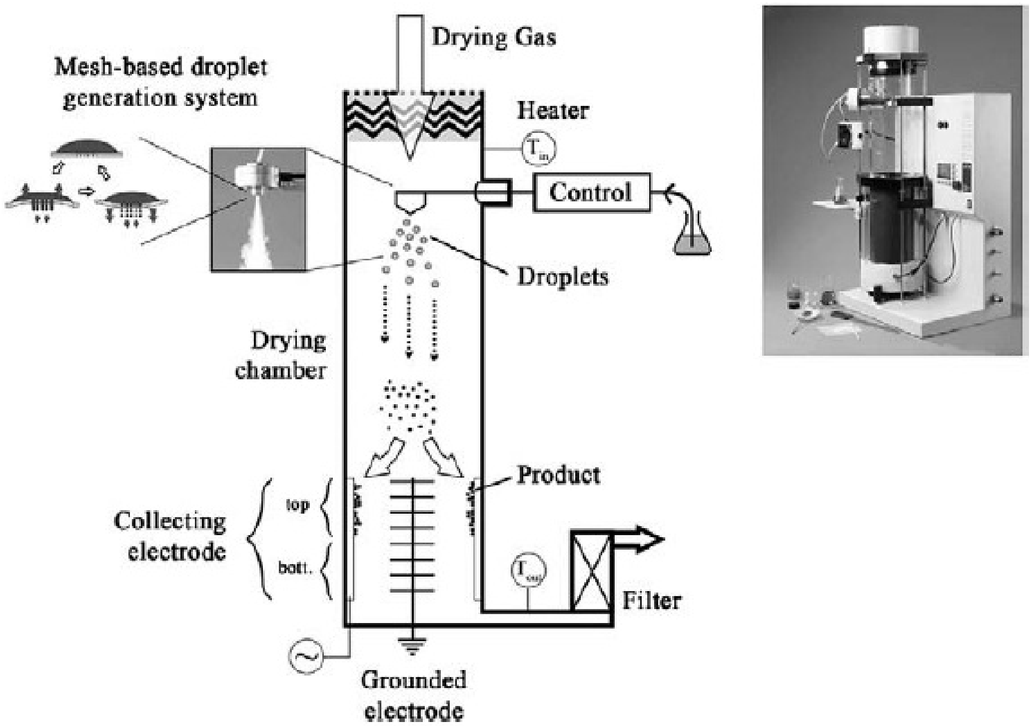

Figure 1. Schematic of working principle of Nano Spray Dryer B-90 (Li *et al.*, 2010)

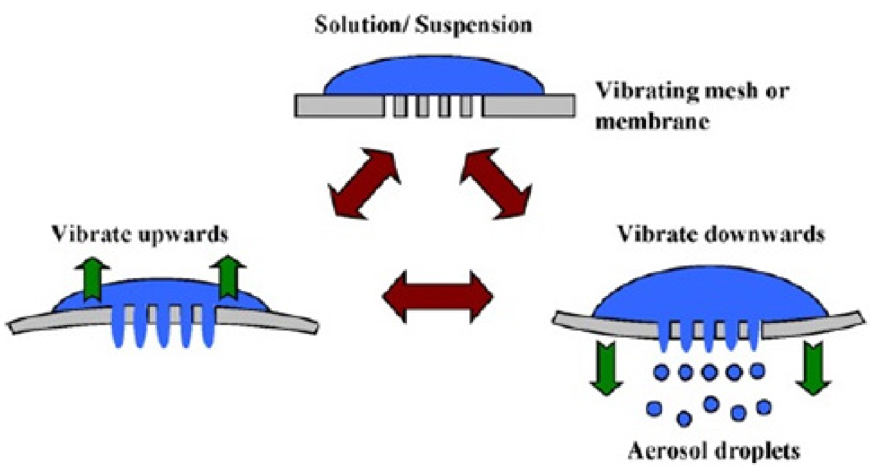

Figure 2. Spray cap membrane vibration at Nano Spray Dryer B-90

Unlike conventional spray dryers use turbulent flow for drying gas, it is taken advantage of the laminar flow using the Nano Spray Dryer B-90 so that the system can be applicable for heat-sensitive products.

The least but not last, electrostatic particle collector is used in Nano Spray Dryer B-90 instead of cyclone collector. Thus, the new system becomes more sufficient and favorable to gather dried powder and also ensure high product yield (Li *et al.* 2010; Lee *et al.*, 2011; Schmid *et al.*, 2011; Heng *et al.*, 2011; Bürki *et al.*, 2011; Baba and Nishida, 2012).

Spray drying conditions/parameters

Operation conditions for eight different formulations were listed in Table1.

While inlet temperature was set to 120°C, outlet and head temperature was around 40-50°C and 70-80°C, respectively. Pressure was approximately 37 mbar and air flow was 120 L/min.

Table 1. Operation conditions for different formulations

Formulation	Inlet Temp. (°C)	Outlet Temp. (°C)	Head Temp. (°C)	Pressure (mbar)	Air Flow (L/min.)
F1	120	41	55	38	120
F2	120	50	58	37	118
F3	120	52	80	34	110
F4	120	45	70	38	120
F5	120	52	75	35	112
F6	120	40	54	36	117
F7	120	45	81	36	117
F8	120	41	69	36	115

Under these conditions, three critical operation parameters, the hole size of spray caps (nozzles), spray capacities and polymer concentrations, were identified. Chitosan nanoparticles were obtained by spraying chitosan solution through Nano Spray Dryer B-90, Buchi, using different spray caps (smallest or biggest) with the hole size of 4μm or 7μm. Furthermore, two different spray capacities, full-capacity (%100) and half-capacity (%50) were performed. Details on the parameters for sprayed formulations were summarized in Table 2.

Table 2. Nano-spray dryer conditions

Formulation	Polymer Concentration (%)	Orifice (μm)	Sprayed Capacity (%)
F1	10	4	50
F2	10	4	100
F3	10	7	50
F4	10	7	100
F5	20	4	50
F6	20	4	100
F7	20	7	50
F8	20	7	100

The studies were performed in duplicate to understand variabilities.

Evaluated parameters

Feed liquids have a big influence on spraying performance. In order to determine the properties of feed liquids, chitosan solutions were considered for their density, drop volume and surface tension. Densities of both chitosan solutions at different concentration were measured using pycnometer. Drop volume and surface tension were determined by Theta Lite Optical Tensiometer, Attension-Biolinscientific, Finland.

Duration of spray process was also recorded to determine the process yield. It was calculated as sprayed sample volume per unit process time. After dried powder was collected and weighed, the product yield was calculated as simply dividing the final amount of chitosan powder to the initial amount of chitosan powder. Final products were also evaluated in terms of particle size, polydispersity index and zeta potential using Malvern Zeta Sizer Nano Series, England. After the spraying process was over, dried powder was collected from the collector. As soon as it was dispersed in pure water, all measurements were performed at least six parallels. TEM image was also captured for the sample which was produced in the best determined condition.

RESULTS

After the density was found by pycnometer, drop volume and surface tension were measured with six parallels with one hundred twenty times measurement for each parallel. Measurements were repeated for both chitosan solutions at 10 and 20% concentration. Results were shown in Table 3.

Table 3. Properties of chitosan solutions at different concentrations

Properties	10% Chitosan Solution	20% Chitosan Solution
Density	1,012	1,070
Drop volume (μL) ±SE	76.06±0.16	80.39±0.32
Surface Tension (mN/m) ±SE	14.02±0.02	14.10±0.03

Two different concentration of chitosan solutions were prepared and then sprayed in various conditions to evaluate the effect of process parameters on particle characteristics and operation capacity. While particle characteristics were considered for particle size, polydispersity index and zeta potential; operation capacity were evaluated with product and process yields. All these results were given in Table 4.

Table 4. Dry powder characteristics and operation capacity

Formulation	Particle Size (nm) ±SE	Polydispersity Index±SE	Zeta Potential (mV) ±SE	Product Yield (%)	Process Yield
F1	282±17	0.962±0.050	19.2±0.3	13	0.018
F2	206±20	0.403±0.103	20.6±0.4	80	0.021
F3	489±15	0.928±0.176	21.7±1.0	19	0.119
F4	462±10	0.484±0.006	19.7±0.4	41	0.125
F5	304±48	0.841±0.245	23.5±0.3	14	0.011
F6	322±63	0.496±0.115	24.0±0.8	70	0.046
F7	404±35	0.625±0.202	28.5±0.3	85	0.022
F8	381±36	0.538±0.109	30.5±0.5	72	0.079

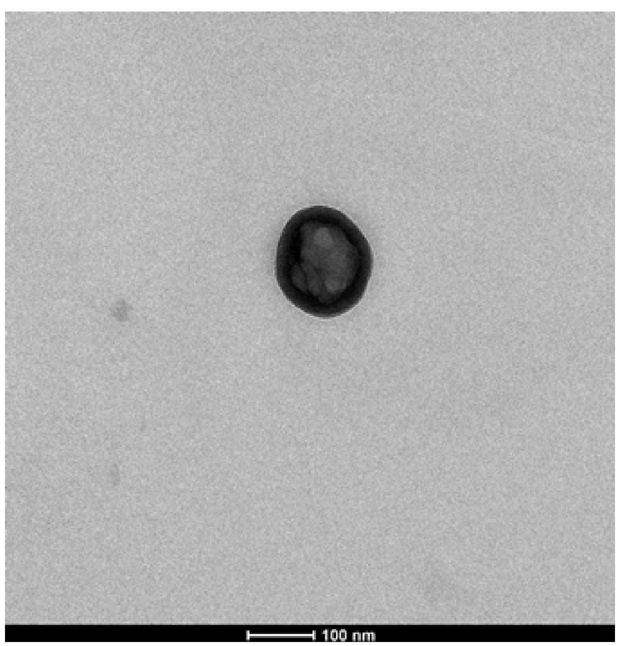

Figure 3. The TEM images for the formulation F2

The smallest chitosan particles were obtained by using Nano Spray Dryer B-90 under the conditions set at for the formulation number F2. The TEM image for this batch was given in Figure 3. Particles had spherical and smooth surfaces.

DISCUSSION

Spray drying technology was used for obtaining dry powder. Nano Spray Dryer B-90 was the latest generation of this technology. It not only presented the dry final product but also made the particle nano-scale, more soluble and spherical. It improved the particle size and morphology thanks to its unique spraying technique based on the piezo-driven spray nozzle. In order to obtain the highest output from this device, some critical parameters should be determined. The parameters may related with feed sample or operation. Here, we examined the feed solution and some operational steps that may affect on particle characteristics or product yield.

It was obviously seen in Table 3, density and surface tension of concentrate chitosan solution were higher than diluted one. Surface tension had an impact on spraying process. Feed liquid with lower surface tension put up less resistance to penetrate the hole of nozzle. As a consequence of that, liquid sprayed easily and product yield rised a lot.

On the basis of the results of the samples at low concentrations (F1 to F4); decreasing of the orifice (hole size of spray caps) led to much smaller particles. This results were found to be similar with other studies (Beck-Broichsitter *et al.*, 2012; Aquino *et al.*, 2014). On the other hand, using smallest orifice caused the dramatic reduction of process yield (6 fold or more). Besides the orifice of spray caps, the effects of spray capacity on these formulations were also investigated. Increasing spray capacity from 50% to 100%, slightly reduced the particle size, but at the same time markedly reduced polydispersity index and also significantly enhanced the product yield (at least 2 fold). Zeta potentials were found almost the same for all samples at low concentrations.

Polymer concentration was found to play a key role for drying efficiency and the yield (Gu *et al.*, 2015). Likewise to the samples at low concentration, the smaller orifices led to smaller particles for the samples at higher concentrations (F5 to F8) too. However, the effect lost its clarity in comparison with lower concentrations. Moreover, opposite to the samples at low concentration, process yield was not directly impressed by using smallest orifice but it was obviously impressed by spray capacity. As the usage of full spray capacity (100%), the process yield was enhanced for 3 or 4 times and also polydispersity index was improved. Although, the product yield could be enhanced by stepping the spray capacity up, producing the samples at high concentration became tricky because of some problems like sticking or powdering occurred so that particle collection became difficult and as a result, product yield might decrease contrary to be envisaged. Increasing the polymer concentration resulted in increasing of zeta potential based on the presence of more cationic chitosan powder.

CONCLUSION

The latest technology for spray drying based on piezoelectric spray head, laminar air flow and electrostatic particle collector was used to produce chitosan nanoparticles. Critical process parameters for spray drying were determined. The results indicated that the orifice, spray capacity and polymer concentration played a crucial role to obtain satisfactory performance. As the orifice directly had an impact on the particle characteristics; spray capacity affected total process time. Besides, both product yield and polymer concentration showed their effect on the process yield. Moreover, using small caps to get smaller particles required to use diluted samples in order to obtain optimum manufacturing process.

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REFERENCES

Aquino, R. P., Stigliani, M., Del Gaudio, P., Mencherini, T., Sansone, F. and Russo, P. (2014), Nanospray drying as a novel technique for the manufacturing of inhalable NSAID powders. *The Scientific World Journal, 1-7,* Article ID 838410.

Baba, K. and Nishida, K. (2012), Calpain inhibitor nanocrystals prepared using Nano Spray Dryer B-90. *Nanoscale Research Letters, 7*:436, 1-9.

Beck-Broichsitter, M., Schweiger, C., Schmehl, T., Gessler, T, Seeger, W. And Kissel, T. (2012), Characterization of novel spray-dryed polymeric nanoparticles for controlled pulmonary drug delivery. *Journal of Controlled Release, 158*, 329-335.

Biswas, A., Bayer, I. S., Biris, A. S., Wang, T., Dervishi, E. and Faupel, F. (2012), Advances in top–down and bottom–up surface nanofabrication: techniques, applications & future prospects. *Advances in Colloid and Interface Science, 170*, 2–27.

Bürki, K., Jeon, I., Arpagaus, C. and Betz, G. (2011), New insights into respirable protein powder preparation using nano spray dryer. *International Journal of Pharmaceutics, 408*, 248-256.

Gu, B., Linehan, B. and Tseng, Y. C. (2015), Optimization of the Büchi B-90 spray drying process using central composite design for preparation of solid dispersions. *International Journal of Pharmaceutics, 491*, 208-217.

Heng, D., Lee, S. H., Ng, W. K. and Tan R. B. H. (2011), The nano spray dryer B-90. *Expert Opinion Drug Delivery, 8(7),* 965-972.

Kulkarni, A. D., Bari, D. B., Surana, S. J. and Pardeshi C. V. (2016), In vitro, ex vivo and in vivo performance of chitosan-based spray-dried nasal mucoadhesive microspheres of diltiazem hydrochloride. *Journal of Drug Delivery Science and Technology, 31,* 108-117.

Kumar, S., Shen, J. and Burgess, D. J. (2014), Nano-amorphous spray dried powder to improve oral bioavailability of itraconazole. *Journal of Controlled Release, 86(3)*, 215-223.

Kumar, S., Shen, J., Zolnik, B., Sadrieh, N. and Burgess, D. J. (2015), Optimization and dissolution performance of spray-dried naproxen nano-crystals. *International Journal of Pharmaceutics, 486,* 159-166.

Lee, S. H., Heng, D., Ng, W. K., Chan, H. K. and Tan R. B. H. (2011), Nano spray drying: a novel method for preparing protein nanoparticles for protein therapy. *International Journal of Pharmaceutics*, *403,* 192-200.

Li, X., Anton, N., Arpagaus, C., Belleteix, F. and Vandamme, T. F. (2010), Nanoparticles by spray drying using innovative new technology: the Buchi nano spray dryer B-90. *Journal of Controlled Release, 147,* 304-310.

Liu, W., Chen, X. D. and Selomulya, C. (2015), On the spray drying of uniform functional microparticles. *Particuology, 22,* 1-12.

Lockman, P. R., Mumper, R. J., Khan, M. A. and Allen, D. D. (2002), Nanoparticle technology for drug delivery across the blood-brain barrier. *Drug Development and Industrial Pharmacy, 28(1),* 1–13.

Schmid, K., Arpagaus, C. and Friess, W. (2011), Evaluation of the nano spray dryer B-90 for pharmaceutical applications. *Pharmaceutical Development and Technology, 16(4),* 287-294.

Singh, R. and Lillard, Jr. J.W. (2009), Nanoparticle-based targeted drug delivery. *Experimental and Molecular Pathology, 86(3),* 215-223.

Wais, U., Jackson, A. W., He, T. and Zhang, H. (2016), Nanoformulation and encapsulation approaches for poorly water-soluble drug nanoparticles. *Nanoscale, 8,* 1746-1769.

Wan, F. and Yang, M. (2016), Design of PLGA-based depot delivery systems for biopharmaceuticals prepared by spray drying. *International Journal of Pharmaceutics, 498,* 82-95.