NANOPARTICLE AND MICROPARTICLE GENERATION WITH SUPER- OR NEAR-CRITICAL CARBON DIOXIDE

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ABSTRACT

Fine dry particles can be generated by a patented process, CO_2-Assisted Nebulization with a Bubble Dryer(R) (CAN-BD) [1,2]. These particles, with aerodynamic diameter range of 400 nm to 5 μm, are suitable for pulmonary drug delivery or other applications in which nanoparticles or microparticles are desired. To generate these particles, two fluid streams (super- or near-critical CO_2 and an aqueous or organic solution), driven by two separate pumps, are mixed intimately in a small volume tee at or near room temperature and a predetermined pressure (e.g., 1200 psig). The resultant mixture is expanded through a flow restrictor to atmospheric pressure to generate an aerosol, which is then dried at a temperature at or below 60°C to produce fine dry powders.

Lately, a newly configured CAN-BD system utilizing one pump to drive both fluids has been developed. The fluid flow rate of each stream can be controlled to a certain degree with check valves and needle valves to produce a stable dense aerosol plume. The dry powders generated by the one-pump CAN-BD have similar characteristics as those generated by the two-pump CAN-BD, and yet the latter is more versatile. The result of comparison between these two CAN-BD systems will be discussed.

Fine dry particles of different solutes (e.g., proteins, sugars, water-soluble drugs, or organic solvent-soluble drugs, etc) suitable for pulmonary delivery have been micronized using the two-pump CAN-BD process [3 to 8]. A parametric study, utilizing the two-pump CAN-BD system and ethanolic solutions containing betamethasone 17,21-dipropionate, has been conducted. The results of the study will be presented.

MATERIALS AND METHODS

Absolute ethyl alcohol with a purity of 99.9% was obtained from AAPER Alcohol, Shelbyville, Kentucky. Betamethasone 17,21-dipropionate (hereafter simply "betamethasone") with a purity of 99.2% was from SICOR-Societa Italiana Corticosteroidi S.p.A. Images of dry powders were obtained using a scanning electron microscope (ISI, model SX-30), and the mean aerodynamic particle size distribution was measured using a TSI Aerosizer (Model 3225 DSP).

EXPERIMENTAL APPARATUS

Fig. 1 is a schematic diagram of a one-pump two-fluid CAN-BD system. Liquid CO_2 is pressurized at room temperature to a predetermined pressure (typically 1200 psig) in a syringe pump (S), and is delivered at this constant pressure into the system. A portion of CO_2 is delivered toward point A to enter a high-pressure cylinder (C) that contains a floating piston to separate liquid CO_2 from an ethanolic solution. This CO_2 stream drives the solution to a mixing tee (T), while the other port of the tee receives liquid CO_2 from (S) via point B. A needle valve (N1 or N2) is used to control the flow rate of each fluid stream, and two check valves (CV1 and CV2) prevent reverse flows of the fluids. The two fluid streams (the ethanolic solution and liquid CO_2) are mixed in (T), and expanded through a flow restrictor (9.5 cm long, fused silica capillary tubes with an inner diameter of 74 μm) to generate aerosols. The pressure of the CAN-BD system during nebulization is maintained constant at the predetermined pressure by (S). The aerosols formed are dried using pre-heated nitrogen gas in a drying chamber (D), which is operated at near atmospheric pressure and a temperature below 60°C. The dry powder is collected on a filter membrane (F) located at the bottom of the drying chamber.

The schematic of a two-pump CAN-BD system has been described elsewhere [3,4,5,7,8]. It is similar to Fig. 1, except that points A and B are not connected, and two pumps (S) are required. One pump drives the liquid solution at point A and the other drives liquid CO_2 at point B.

DISCUSSION OF RESULTS

One reason to develop the one-pump system is to reduce the cost of the system equipment, since high-pressure pumps are expensive and are the major cost for the entire system. For this newly configured one-pump two-fluid system, three solutes (sodium chloride, palmitic acid and mannitol) have been micronized at 1200 psig. The particle size and morphology of the dry powders generated are similar to those micronized by the two-pump system. The SEM images of the fine particles generated from the two-pump system for sodium chloride [6,7], palmitic acid [7] and mannitol [8] have been published. One shortcoming of the one-pump system is that it is difficult to control the flow rates of the two fluid streams independently, since the range of the fluid flow rates that can be controlled with the needle valve is rather narrow. The average flow rates of a given experiment can be calculated knowing the total volume of liquid CO_2 delivered into the system at the predetermined pressure (1200 psig), and the volume of liquid solution pumped out of the high-pressure cylinder. For the three experiments conducted, the liquid solution flow rate was about 0.5 cc/min, and the CO_2 flow rate was about 2 cc/min.

In Table 1 are shown the results of a process parameter study for the two-pump system. The solution used as a model for nebulization was betamethasone dissolved in ethanol. Experiments were performed at a drying temperature of 60°C to investigate the effects of varying solute concentration, CO_2 delivering pressure and inner diameter (ID) of flow restrictors. To investigate the influence of each process parameter, the experimental conditions were chosen such that comparison of results could be made in which only one process parameter was manipulated. The base case conditions (Run 1 in Table 1) were as

follow: concentration of betamethasone in ethanol = 3 % (w/v), ethanol solution flow rate = 0.3 cc/min, CO_2 pressure = 1200 psig, nitrogen flow rate = 20 L/min, fused silica capillary tube flow restrictor ID = 74 µm with a length of 9.5 cm. At the end of each experiment, the liquid solution lines from the pump to the flow restrictor were flushed with pure solvents.

The aerodynamic particle diameter of Run 1 as measured by a TSI Aerosizer had a mean value of 0.81 µm with 95% of the particles less than 1.34 µm and 5% less than 0.51 µm (Figs. 2 and 3). [All the particle size data in this paper are reported as number weighted]. The effects of lowering solute concentrations (to 1% and 0.2%) indicate virtually no change in the mean particle size compared to Run 1. In our previous work on aqueous solutions containing mannitol or *myo*-inositol [8], the Aerosizer data did show that the mean particle size decreased from 1.6 µm to 1 µm as the solute concentration decreased from 10 % to less than 1 %.

From the visual observation of the SEM images (Figs. 3 and 4), the particle size of the powder from 0.2% solution is smaller than that from 3% solution. [Column "SEM (µm)" in Table 1 shows our best estimates of particle sizes that can be assessed visually from the SEM's]. Fig. 4 shows that the geometric size of the particles appears to be between 0.05 µm to 0.50 µm, and yet the Aerosizer data indicate a mean aerodynamic particle diameter of 0.79 µm.

The effect of CO_2 aerosolizing pressure is next discussed. When the pressure was increased form 1200 to 1800 psig, no noticeable change in particle size was seen. However, when the pressure was further reduced below the CO_2 vapor pressure (~900 psig), a significant increase in particle size was observed (Figs. 5 and 6). The mean aerodynamic particle size increased to 1.69 µm when the pressure was lowered to 300 psig. This could be attributed to less dispersion of microdroplets and less formation of microbubbles from gaseous CO_2 at low pressures than from near-critical or supercritical CO_2 at 1200 psig. Note that CO_2 to solution mass flow rate ratio for the 300 psig run (Run 6) was only 0.007 g/g, while that for the base case 1200 psig run was 14.5 g/g. This same phenomenon was also observed during our previous work with *myo*-inositol dissolved in water, though the result was not included in that paper [8]. As the pressure was decreased from the base case 1200 psig to 300 psig, the mean *myo*-inositol particle diameter increased from 1.5 to 4.5 µm.

When the ID of the flow restrictor was increased from 50 to 74 to 100 µm with all the other parameters unchanged (e.g., solution flow rate remained at 0.3 cc/min), the CO_2 flow rate increased, which led to the increase in the CO_2 to solution flow rate ratio. Table 1 shows no particle size change when the ID decreased from 100 to 74 µm (i.e., the flow ratio reduced from 52.7 to 14.5 g/g). However, the particle size increased when the ID was further reduced to 50 µm (with a flow ratio of 4.9 g/g). The SEM images (Figs. 7 and 8) independently confirm the Aerosizer particle size measurements. The particle size changes due to the changes in CO_2 pressure as well as the restrictor ID are correlated with the CO_2 to solution mass flow rate ratio in Fig. 9. This plot implies that, when this mass ratio is greater than about 12, the particle size stabilizes at about 0.8 µm (according to the TSI Aerosizer data). When the ratio decreases , the aerodynamic and geometric particle sizes increase. Such a correlation has also been demonstrated for the aqueous solutions containing mannitol or *myo*-inositol as a solute [8]. The mean aerodynamic particle size for the aqueous solutions at high mass flow rate ratio was about 1.5 µm.

CONCLUSIONS

(A) A newly developed one-pump two-fluid CAN-BD system has been presented. The powders generated from this system were similar to those generated by the two-pump system. This system is less expensive, but needs more elaborate needle valves to control the flow rates of the fluid streams, and the system cannot provide the range of process conditions achievable with the two-pump system.

(B) For the two–pump system, a process parameter study, utilizing ethanolic solutions containing betamethasone, was conducted to investigate the effect of process parameters on particle characteristics. Particles with diameters as low as 50 nm were attained.

(C) The results of process parametric studies for ethanolic solutions containing betamethasone as well as aqueous solutions containing mannitol or *myo*-inositol [8] indicated that CO_2 to solution mass flow rate ratio has a significant influence on the particle size. By manipulating this ratio, the particle size of fine powders generated by the CAN-BD can be controlled from nanometer size to micrometer size.

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REFERENCES

[1] SIEVERS, R.E., KARST, U., U.S.Patent 5,639,441, **1997**.
[2] SIEVERS, R.E., KARST, U., European Patent 0677332 B1, **2002**.
[3] SIEVERS, R.E., MILEWSKI, P.D., SELLERS, S.P., MILES, B.A., KORTE, B.J., KUSEK, K.D., CLARK, G.S., MIOSKOWSKI, B., VILLA, J.A., Ind. Eng. Chem. Res., Vol. 39, **2000**, p. 4831.
[4] SELLERS, S.P., CLARK, G.S., SIEVERS, R.E., CARPENTER, J.F., J. Pharm. Sci., Vol. 90, **2001**, p. 785.
[5] SIEVERS, R.E., HUANG, E.T.S., VILLA, J.A., WALSH, T.R., Proceedings of 8th Meeting on Supercritical Fluids, Bordeaux, France. **2002,** p. 73.
[6] SIEVERS, R.E., HUANG, E.T.S., VILLA, J.A., KAWAMOTO, J.K., EVANS, M.M., BRAUER, P.R., Pure Appl.Chem. 73(8), **2001**, p. 1299.
[7] SIEVERS, R.E., HUANG, E.T.S., VILLA, J.A., ENGLING, G., BRAUER, P.R., J. of Supercritical Fluids, **2003** (in press).
[8] HUANG, E.T.S., CHANG, H.Y., LIANG, C.D., SIEVERS, R.E., **2003** ACS Symposium Series Books (in press)

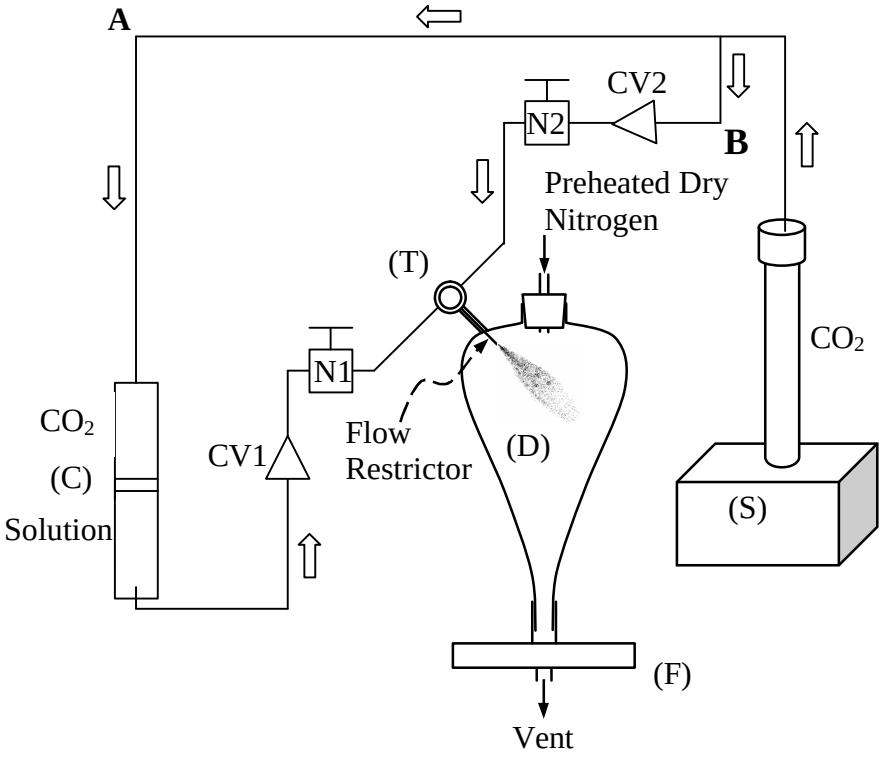

Figure 1 Schematic diagram of a one-pump CAN-BD system.
[Notes: syringe pump (S), high-pressure cylinder (C), low volume mixing tee (T), drying chamber (D), filter holder (F), needle valves (N1 & N2), check valves (CV1 & CV2)].

Table 1 Parametric Study for 2-pump CAN-BD at 60°C (Betamethasone in EtOH)

Run No.	Conc. % (w/v)	ID (µm)	Press. (psig)	Ave. CO_2 Rate (cc/min)	CO_2 to solution flow ratio (g/g)	SEM (µm)	Particle size (µm) >5%	Ave. Diam.	<95%
Effect of concentration of solute									
Run 1	3	74	1200	4.4	14.5	0.1 - 1.0	0.51	0.81	1.34
Run 2	1	74	1200	3.4	11.2	0.1 - 1.5	0.48	0.78	1.24
Run 3	0.2	74	1200	3.4	11.2	0.05 - 0.5	0.48	0.79	1.41
Effect of CO_2 pressure									
Run 4	3	74	1800	4.2	14.9	0.1 - 0.7	0.48	0.79	1.38
Run 1	3	74	1200	4.4	14.5	---	---	0.81	---
Run 5	3	74	700	0.94	0.50	0.1 - 2.5	0.64	1.15	2.26
Run 6	3	74	300	0.04	0.007	0.2 - 6.0	0.73	1.69	6.65
Varying restrictor ID to change fluid flow ratios									
Run 7	3	100	1200	16.0	52.7	0.1 - 0.5	0.47	0.82	1.85
Run 1	3	74	1200	4.4	14.5	---	---	0.81	---
Run 8	3	50	1200	1.5	4.9	0.8 - 1.2	0.56	0.93	1.60

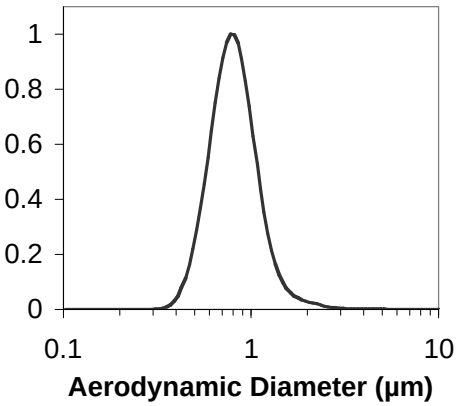

Fig. 2 Particle distribution, Run 1

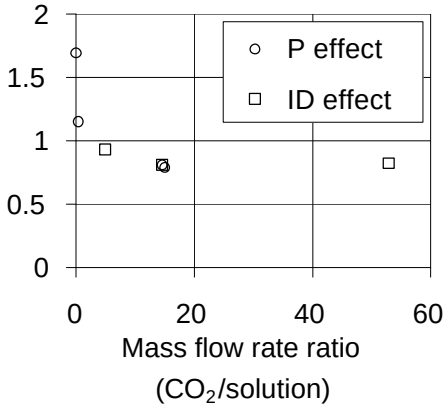

Fig. 9 Effect of flow ratio on size

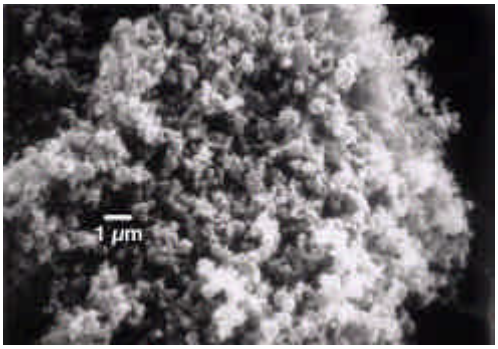

Fig. 3 Run 1 (Base case) – 3% Conc., 74 µm ID, 1200 psig

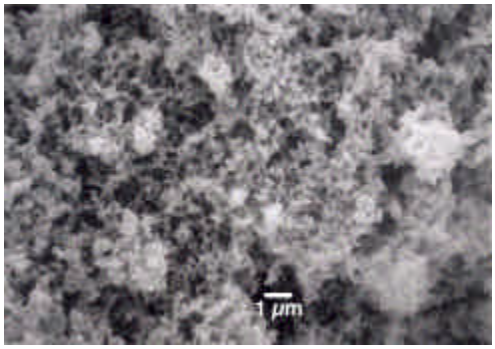

Fig. 4 Run 3 – 0.2% Conc.

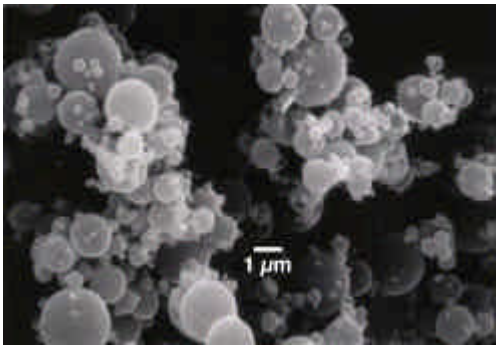

Fig. 5 Run 5 – 700 psig

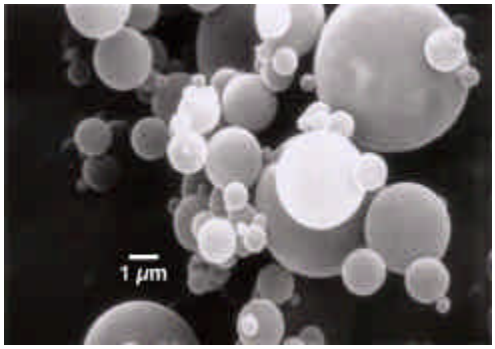

Fig. 6 Run 6 – 300 psig

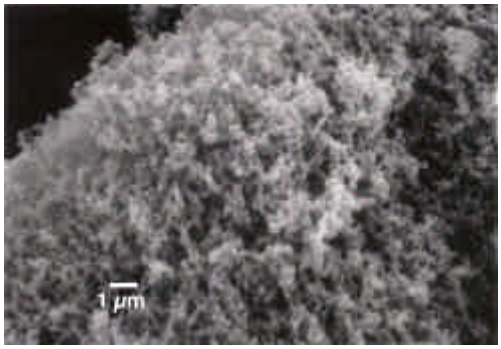

Fig. 7 Run 7 – 100 µm ID

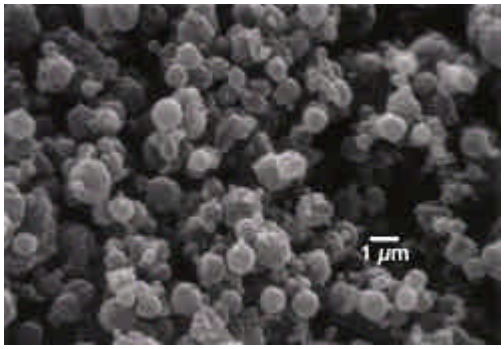

Fig. 8 Run 8 – 50 µm ID