ELSEVIER

International Journal of Pharmaceutics 104 (1994) 73–85

international journal of pharmaceutics

Some functional aspects of air-jet nebulizers

Ralph W. Niven [a,*], Joseph D. Brain [b]

[a] *Amgen Inc., 8-1-A 215, Thousand Oaks, CA 91320, USA,* [b] *Respiratory Biology Program, Harvard School of Public Health, Department of Environmental Health, 665 Huntington Avenue, Boston, MA 02115, USA*

(Received 18 November 1992; Modified version received 3 September 1993; Accepted 21 September 1993)

Abstract

Mass median aerodynamic diameters (MMADs, μm), flow rates (l air/min) and mass outputs (ml/min and μl/l air) were determined over a range of air pressures (psig) for a variety of clinically used nebulizers. Nebulizers were also modified to determine the rate at which fluid was aspirated from the nebulizer cup into the nebulizer jets (ml/min and μl/l air). Only a small fraction of this fluid emerges as aerosol that can be inhaled. The ratio of mass output to fluid uptake from the reservoir is termed mechanical efficiency. The stability of the enzyme lactate dehydrogenase (LDH) to nebulization at two different starting volumes was examined in the 3-jet Collison nebulizer. The results are discussed with respect to the measured operating characteristics. In all the nebulizers tested, MMADs were inversely related (e.g., 6.4 to 3.8 μm at 10 and 40 psig, respectively, for the 'Cirrus' nebulizer), whereas mass outputs (e.g., 0.11 to 0.35 ml/min at 10 and 40 psig for the 'Misty') and flow rates (e.g., 4.5 to 10.4 l/min at 10 and 40 psig for the Misty) were directly related, to the driving air pressure. MMADs and mass output are measures of how well nebulizers atomize liquid into droplets of respirable sizes. Aspiration rates varied substantially between the nebulizers and ranged from 9 ml/min (5 psig) to 28 ml/min (40 psig) for the DeVilbiss No. 45 whereas the Collison 3-jet nebulizer aspirated liquid at 118 ml/min (5 psig) to 333 ml/min (40 psig). All nebulizers exhibited an exponential decline in the aspiration rate of liquid expressed in terms of the throughput of air (ml/l air). Rate constants ranged from 0.6 to 21.6 ($\times 10^{-2}$ min/l air) in the DeVilbiss No. 45 and Airlife Misty, respectively. These values give an indication of how the aspiration of fluid is affected by the air flow. Nebulizer mechanical efficiency was never > 2% (vents closed); this parameter is inversely related to the probable number of times that a drug will be aerosolized within the nebulizer. LDH was inactivated by nebulization. The degree of inactivation was greater at a 10 ml starting volume than at a 40 ml starting volume. The results indicate that monitoring the functional characteristics of nebulizers may be helpful in the assessment of drug stability.

Key words: Aerosol; Cascade impaction; Drug damage; Inhalation therapy; Jet nebulizer; Lactate dehydrogenase

1. Introduction

Nebulizers utilize aqueous solutions or suspensions. They are simple to use and widely employed in hospitals and in home care therapy. Consequently, numerous studies (Mercer, 1965; Ryan et al., 1981; Clay et al., 1983; Smaldone et al., 1988) have evaluated different facets of their clinical function. Generally these include a measurement of the droplet size, mass output, dead

* Corresponding author.

SSDI 0378-5173(93)E0277-W

volume and occasionally solute output. Rightly so, the results are often presented in terms of the importance to a patient inhaling a nebulized drug. For example, the droplet size is often measured at a point where it would enter the oral cavity. This is critical in determining if the aerosol size is suitable for inhalation purposes and if it falls within the prescribed mass median aerodynamic diameter (MMAD) of 0.5–3 μm. Assessment of the amount of drug leaving a device gives information on the amount of drug that a patient will inhale. The dead volume remaining after nebulization has ended gives an idea of waste and overall cost.

Such results do not focus on nebulizer function and how its operating characteristics can impact the drug or drug formulation. For instance, it has been shown that solute concentration occurs within air-jet nebulizers (Mercer et al., 1968; Niven and Schreier, 1990) and this, in conjunction with a lowering of temperature during nebulization, could result in precipitation of the drug due to it exceeding its solubility limit in the nebulizer solution. This has been observed with pentamidine (Debs, personal communication) and the result is a substantially lowered drug output. Other factors associated with nebulizer function may directly influence drug stability. Ultrasonic nebulizers are known to produce unwanted effects such as changes in the pH and odor of antibiotic solutions (Takanami and Goto, 1990). More importantly, the breakdown of ^{99m}Tc-DTPA (Waldman et al., 1987) and denaturation of α-interferon has been observed in ultrasonic nebulizers (Sato et al., 1992). Air-jet nebulizers also have been seen to damage liposomes containing encapsulated solute (Niven and Schreier, 1990; Taylor et al., 1990) and therefore may be capable of inactivating or affecting some proteins.

In spite of these points, it must be recognized that proteins are normally purified and stabilized in aqueous solution and likewise liposomes are normally prepared as aqueous dispersions. It is therefore logical to expect that drugs will be nebulized prior to the development of any other type of formulation, such as a dry powder. In fact the latter may present a formidable challenge in the case of proteins (Niven, 1993). Consequently, it seems reasonable to believe that a significant role for nebulizers will remain in future inhalation aerosol therapy.

This study evaluates a series of clinically used air-jet nebulizers and catalogues several parameters of nebulizer function. The results are discussed in terms of their possible impact on drug stability. The effect that air-jet nebulization has on the enzyme lactate dehydrogenase at two different starting volumes serves as an example. The air flow, aerosol mass output and the normal aspiration rate at which liquid is forced through the nebulizer jets are all measured.

2. Materials and methods

2.1. Nebulizers

The commercial jet nebulizers studied were 'Misty' (Airlife Inc., Montclair, CA), 'Cirrus' (DHD Medical Products, Canastota, NY), DeVilbiss No. 45 (The DeVilbiss Co., Somerset, PA), 'UpDraft' (Hudson Respiratory Care Co., Temecula, CA), 'Aerotech II' (Cadema Medical Products, Middletown, MY), 'Raindrop' (Puritan Bennett, Lenexa, KA) and the 'Whisperjet', 'Acorn I' and 'Acorn II' (Marquest Medical Products Inc., Englewood, CA). A 1-jet and 3-jet Collison nebulizer were also studied. They were machined from Delron acetal resin plastic (E.I. Dupont DeMours and Co., Wilmington, DE) according to the specifications detailed by May (1993).

2.2. Nebulizer droplet size

A 5% w/w aqueous solution of sodium chloride (NaCl; Fisher, Medford, MA) and 5,6-carboxyfluorescein (CF; Kodak Eastman, Rochester, NY) in a ratio of 98:2 was used as the nebulizer solution for the measurement of droplet sizes. A variable output compressor (PCS-4; Timeter Corp., Lancaster, PA) supplied pressurized ambient air to the nebulizers and an Andersen Mark I cascade impactor (Andersen Samplers Inc., Atlanta, GA) was used to size the dried aerosol particles. The flow rate of the cascade

impactor was checked using a calibrated rotameter (Aalborg Instrument and Control Inc., Monsey, NY) attached to the pump outlet. A mixture of different colored latex bead standards (Polysciences, Warrington, PA) of diameters 0.88, 2.92 and 5.78 μm was prepared in methanol at a concentration that would predict less than 1 particle every 10 droplets (Bangs, 1984). This suspension was aerosolized to check if the impactor was depositing particles on stages in the range of their predicted cutoff size intervals. Since the 28.3 l/min air flow entering the impactor exceeds the air flow produced by the nebulizers, diluent air was required to make up the volume flow entering the impactor. The diluent air was dried and heated to ≥ 40°C before mixing with the nebulizer aerosol flow to aid thorough drying of the droplets (Fig. 1). The resulting mixture was then passed through a 15 cm length of 2 cm internal diameter corrugated plastic tube and into a vertical diffusion dryer which was 67 cm in length and of 2 cm internal bore surrounded by Natrasorb T silica beads (4 × 12 mesh; Multiform Desiccants Inc., Buffalo, NY) before entering the impactor. The droplet sizes produced by each nebulizer were characterized at a variety of pressures ranging from 5 to 40 pounds per square inch gauge (psig) and at one constant flow rate of 6 l/min. The 6 l/min air flow rate is often used in hospital settings. In addition, the DeVilbiss No. 45 nebulizer was analyzed with an open vent.

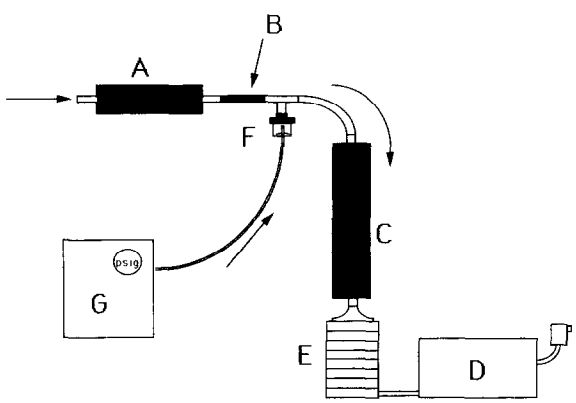

Fig. 1. Diagram of the aerosol drying and sizing system. A and C are diffusion dryers with A used to dry the ambient make up air and C to aid drying of the wet aerosol and gas mixture. Make up air was heated to ≥ 40°C by passing it through heated metal tubing, B, before mixing with aerosol produced by the nebulizer, F, driven by compressed air from a pump, G. The dry aerosol was sized by pulling it through a cascade impactor, E, by a vacuum pump, D, at 28.3 l/min.

Nebulizers were operated from 30 to 120 s, depending upon the flow rate used, to allow collection of assayable quantities of material on the impactor stages. After an experiment, 100 μl of water was placed on each stage of the impactor and wiped clean with a ≈ 6 cm^2 piece of lint-free tissue. Each tissue, as well as the filter (EPM 2000 glass microfibre; Whatman International Ltd, Maidstone, U.K.) on the last stage of the impactor, was placed in a separate borosilicate glass vial and filled with 5 ml of 0.1 M NaOH. After thorough mixing, 100 μl from each solution was placed in 96-well microtiter plates (Costar, Cambridge, MA) in duplicate. Solutions of vials containing filters from the final stage of the impactor were first centrifuged to remove fibrous material and then placed in the microtiter plates. Plates were analyzed using a fluorescent plate reader (Cytofluor 2300; Millipore, Bedford, MA) with excitation and emission bandpass filters of 485 and 530 nm, respectively. The size distributions obtained from these fluorometer readings are those of the dried particles. Initial wet droplet sizes were determined from dry particle values using the relationship:

$$d_{ae} = d_p \cdot \sqrt{\frac{\kappa_s}{\kappa}} \cdot \sqrt[6]{\left(\frac{\rho_l}{\rho_s \cdot (g_s)^2}\right)} \qquad (1)$$

where d_{ae} is the mass median aerodynamic diameter (MMAD) of the aerosol droplets, d_p denotes the MMAD of the measured dried particles, ρ_s and ρ_l are the densities, at ambient temperature and pressure, of the solute and liquid, respectively, g_s represents the weight fraction of solute in the droplets (Ferron et al., 1976) and κ_s and κ are the dynamic shape factors of a cuboidal salt particle (1.07) and a droplet (1.00), respectively (Ferron et al., 1976).

The weight fraction of solute in the droplets can be estimated from:

$$g_s = \frac{a_s}{a_s + a_{sv}} \qquad (2)$$

where a_s and a_{sv} are the masses of solute and solvent in the nebulizer solution, respectively. The equation is applicable for the primary, unevaporated droplets created inside the nebulizer. Finally, the density of the liquid can be approximated by

$$\rho_l = \frac{a_{sv} + a_s}{\left(\frac{a_{sv}}{\rho_{sv}}\right) + \left(\frac{a_s}{\rho_s}\right)}$$

where ρ_{sv} is the density of the solvent, or

$$\rho_l = \frac{a_{sv} + a_s}{a_{sv} + \left(\frac{a_s}{\rho_s}\right)} \quad (3)$$

where the solvent is water. It was assumed throughout that the density of the solute was that of sodium chloride (2.165 g/ml; Handbook of Physics and Chemistry, 1980) and was unaffected by the presence of the fluorescent marker.

2.3. Aerosol mass outputs

The air flow rates of the empty nebulizers in l air/min (A_t) were measured over 5–50 psig after attaching a calibrated rotameter to each of the nebulizer outlets. Each nebulizer was then filled with a known weight of distilled water. The nebulizers were operated for a 10 min period at a variety of air pressures. After each trial the nebulizer was reweighed to measure the total loss of fluid. Results are expressed in terms of mass output in ml water/min (M_t) and in μl water/l air flow ($M_t/A_t = M_f$) (Mercer et al., 1968). For convenience the mass loss is expressed as a volume since the density of water is 1. The mass loss represents the total loss of liquid from the nebulizers and includes both aerosol droplets as well as solvent loss due to evaporation (Mercer et al., 1968; Dennis et al., 1990).

2.4. Liquid aspiration rates and nebulizer mechanical efficiency

As liquid passes through the inner portion of the nebulizer where aerosolization takes place, the vast majority of the resultant liquid discharge is impacted against strategically placed baffles and/or the container walls. It then returns to the liquid reservoir. To measure the amount of liquid passing through the nebulizer jets, per min (Q_t) and per l air flow ($Q_t/A_t = Q_f$), the nebulizer cup was sealed above the fluid reservoir (J: Fig. 2) to prevent recirculation of fluid. The seal did not restrict aerosolization through the nebulizer jet. For all nebulizers, except the Collison 1- and 3-jet nebulizers, an external reservoir containing $\approx$ 100 ml of a known weight of distilled water was supplied by a microprocessor-controlled peristaltic pump (Model 7524–00; Cole-Parmer Instrument Co., Chicago, IL), to the nebulizer cup at a rate that maintained $\approx$ 4 ml of water in the reservoir during aerosolization. Aspiration rates of water (Q_t) were measured, for a range of air pressures supplying the nebulizers, by weighing the external fluid reservoir before and after a known time of aerosolization. Aspiration values per l air flow (Q_f) were also found by calculating the ratio of the volume of water discharged by the nebulizer jet to the volume of air passed through the nebulizer jet in the same time period. A fixed volume of fluid in the nebulizer cup was maintained by pumping liquid from the external reservoir at the same rate as the liquid was being aspirated to the nebulizer jets.

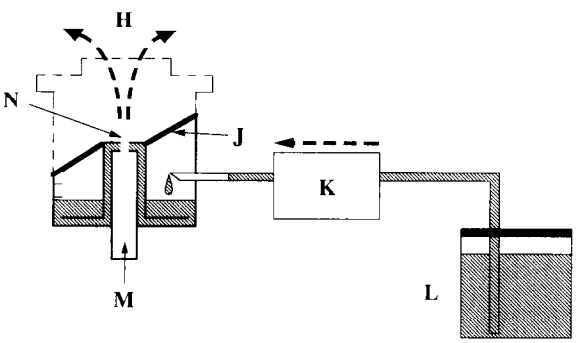

Fig. 2. Schematic of the system used to estimate the aspiration of water by the various nebulizers. A seal, J, was made above the nebulizer cup, N. It did not restrict fluid from being aerosolized at the nebulizer jet but prevented any of the liquid discharge, H, from returning to the reservoir. A constant fluid volume was maintained in the nebulizer cup by supplying water from an external fluid reservoir, L, using a microprocessor-controlled peristaltic pump, K. This liquid throughput is called (Q_t). The air flow (A_t) was generated from a compressor and entered via the gas inlet, M.

The shape of the Collison nebulizer 'heads' (P; Fig. 3) allowed their liquid inlets to be submerged directly into a known weight of water while the return of the aerosolized fluid to the liquid reservoir was prevented by sealing the nebulizer below the orifice outlets (Fig. 3). The ratio of the aerosol mass output to the aspiration rate (Q_t) is termed the mechanical efficiency (E_m) and is computed as:

$$E_m = \left(\frac{M_t}{Q_t}\right) \cdot 100 = \left(\frac{M_f}{Q_f}\right) \cdot 100 \quad (4)$$

The expression indicates the mechanical ability of the device to transfer the bulk mass of liquid to the external atmosphere either as liquid droplets or evaporated water. Thus, the values of E_m are conservative estimates of the true aerosol efficiency of the nebulizers since a substantial component of the calculated aerosol mass output may be due to solvent evaporation.

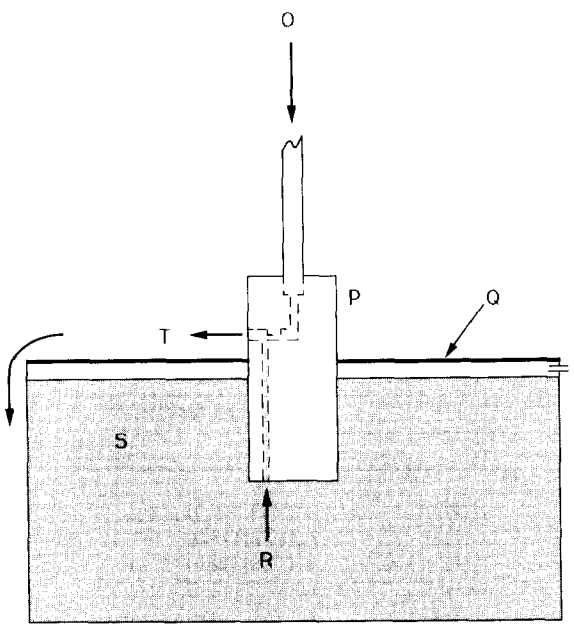

Fig. 3. Schematic of the system used to estimate the aspiration of water by the Collison 1- and 3-jet nebulizers. The Collison head, P, was placed in a large fluid reservoir (> 500 ml), S, such that the aerosol outlet, T, was above the water. The nebulizer was then fixed in position and the fluid reservoir was sealed, Q, below the aerosol outlet. Air was supplied via a gas inlet, O, causing fluid to be aspirated through a liquid inlet, R, and subsequently discharged as a gross aerosol at the aerosol outlet.

2.5. Intra-device variability

The variabilities of aerosol mass output (M_t and M_f) and aspiration rate (Q_t and Q_f) of six Marquest Whisperjet nebulizers were also evaluated as described above.

2.6. Effect of nebulization on lactate dehydrogenase

The protein (LDH; M4 isozyme from rabbit muscle, ICN, Irvine, CA) was used without further purification. Prior to nebulization, the crystalline suspension of LDH was dialyzed against 20 mM potassium phosphate buffer, pH 7.5 and was diluted to a concentration of 25 μg/ml. The LDH was aerosolized at two different starting volumes of 10 and 40 ml in a 3-jet Collison nebulizer at 40 psig. For a fixed aspiration rate it would be expected that the number of times a fixed volume of fluid cycles through the nebulizer jets would be greater for a smaller starting volume and consequently any observed damage might be excacerbated. Enzymatic activity in 25μl samples was measured at 25°C, as follows. A 1 ml reaction mixture of 25 mM Tris-Cl buffer, 0.1 M KCl, pH 7.5 containing 2 mM pyruvate and 0.215 mM reduced nicotinamide adenine dinucleotide was prepared. The reaction was initiated by the addition of 5 μl of the nebulizer sample. Activity was measured by the decrease in the absorbance at 340 nm using a Beckman DU 650 UV spectrophotometer (Beckman Instruments, Inc., Allendale, NJ). Activity is expressed as a percentage of initial activity.

3. Results and discussion

3.1. Nebulizer droplet size

The mass median droplet size decreases as the driving air pressure is increased (Table 1). These results are in agreement with studies that have used laser light diffraction instruments to size

particles produced by nebulizers (Newman et al., 1986; Miller et al., 1990). The estimated droplet sizes, over the range of driving pressures studied, are generally larger than the 0.5–3.0 μm size often described as acceptable for deep lung penetration (Gonda and Byron, 1978). They are also larger than droplet sizes estimated by laser techniques (Newman et al., 1986; Miller et al., 1990). The primary reason for these differences is that Eq. 1 estimates the size of droplets that escape the nebulizer, whether primary or secondary, the instant after they are formed within the nebulizer. In contrast, laser sizing techniques measure the size of droplets at some distance from the outlet of the nebulizer; thus some time elapses between the formation of a droplet and when it reaches the laser beam. Evaporation takes place during this time period and therefore the sizes measured by laser instrumentation will be highly dependent upon the conditions under which the measurements were taken and so it becomes difficult to compare data obtained from one laboratory to another. The use of the described 'back' calculation method avoids such problems. However, neither technique actually measures the size of the aerosol that emerges from the aerosol jets. This is a crude aerosol of larger median size given that 99 + % typically returns to the reservoir fluid. It is assumed that measurements of the droplet sizes that leave the nebulizer will reflect changes occurring in the gross aerosol produced within the nebulizer chamber.

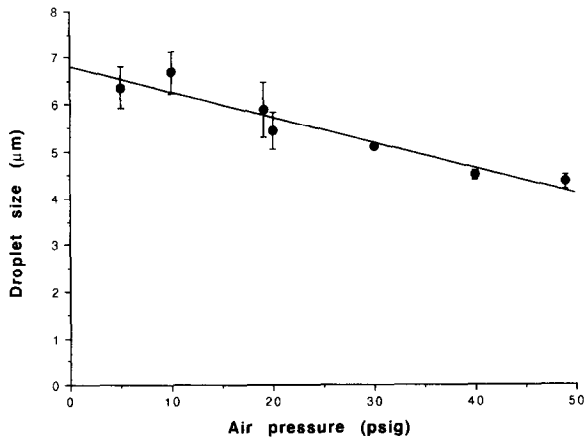

Fig. 4. The relationship between the estimated droplet size of the Marquest Whisperjet nebulizer and that of the driving air pressure. The data demonstrates a linear inverse relationship ($R^2 = 0.92$) between the droplet size and air pressure. Three experiments were conducted at each pressure. The means are shown with their ranges.

The effect of air pressure on droplet size was more extensively examined with a Whisperjet in Fig. 4. The resultant changes in MMADs are

Table 1
Effect of driving air pressure on the droplet size [a] leaving the nebulizers

Nebulizer	Droplet sizes (μm)			
	Air pressure (psig) [b]			6 l/min jet flow (psig) [c]
	10	20	40	
Puritan Bennett Raindrop	5.6 [2.3]	5.2 [2.1]	3.3 [1.3]	4.8 [1.9] (16.0)
Airlife Misty	6.3 [2.5]	5.3 [2.1]	4.8 [1.9]	6.3 [2.5] (13.0)
Cadema Aerotech II	4.7 [1.9]	3.6 [1.5]	3.2 [1.3]	4.4 [1.8] (14.0)
DHD Cirrus	6.4 [2.6]	6.0 [2.4]	3.8 [1.5]	6.3 [2.5] (11.6)
Devilbiss No. 45 closed vent	5.3 [2.1]	4.6 [1.8]	4.6 [1.8]	6.3 [2.5] (6.0)
Devilbiss No. 45 open vent	5.8 [2.3]	5.4 [2.2]	3.3 [1.3]	6.1 [2.5] (6.0)
Hudson Updraft	5.7 [2.3]	5.7 [2.3]	5.0 [2.0]	5.6 [2.3] (34.0)
Marquest Whisperjet [d]	6.4 [2.6]	5.5 [2.2]	4.5 [1.8]	5.8 [2.3] (19.1)
Marquest Acorn I	4.7 [1.9]	4.5 [1.8]	4.0 [1.6]	4.9 [2.0] (13.7)
Marquest Acorn II	4.8 [1.9]	4.9 [2.0]	4.5 [1.8]	4.7 [1.9] (25.0)

[a] Droplet sizes (μm) as calculated from the dry particle sizes (shown in square brackets) using Eq. 1.
[b] Pressure reading in pounds per square inch gauge (psig).
[c] Values in parentheses are the air pressures producing 6 l/min air flow at the outlet of the nebulizer.
[d] These results together with droplet sizes obtained using the additional air pressures of 5, 30 and 48 psig are shown in Fig. 4.

shown in Fig. 4, where a linear inverse relationship is observed over the range of pressures studied. The extent to which droplet size changes as a function of driving pressure varies according to the nebulizer studied. In some cases, such as the Acorn II nebulizer, the reduction in droplet size was only 0.3 μm (< 15%) over 40 psig. Whereas for the Cirrus nebulizer, the size reduction is > 2.5 μm (> 40%) over the same pressure range. It is not known to what extent this behavior is a general characteristic of the type of nebulizer or is a feature of the individual nebulizer.

The distribution of droplet sizes changes in all nebulizers with increasing driving pressure. An example is shown for droplet size distributions of the Cirrus nebulizer, normalized for the size interval between impactor stages, at air pressures of 10 and 40 psig (Fig. 5). These results are in agreement with the work of Rizk and Lefebvre (1982). Although the mean data obtained from the cascade impactor could not be adequately described by a log normal distribution, previous laser diffraction studies by Clay et al. (1983) have indicated that the spread of droplet sizes increases with increasing flow rate.

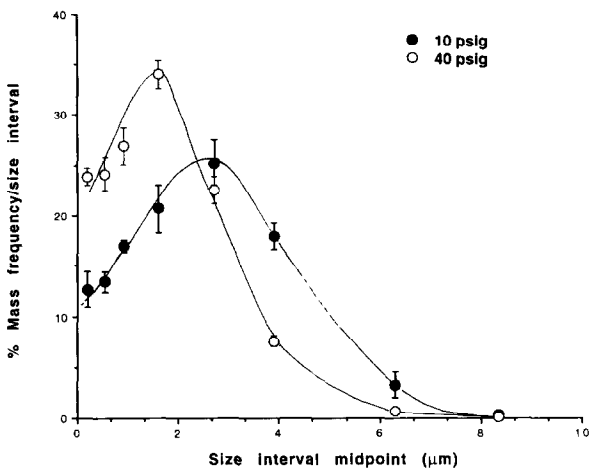

Fig. 5. The effect of driving air pressure on the size distribution of aerosolized droplets. The frequency distributions were obtained from a DHD Cirrus nebulizer containing a 5% w/v NaCl:CF solution (98:2) which was operated at 10 psig (●) and 40 psig (○). Three experiments were conducted at each air pressure. The means are shown with their ranges.

Pressure and therefore energy is required to force air through the gas orifice of nebulizers. The droplets are created primarily by the shearing action of the air flow as it passes the region where liquid is being supplied (Lefebvre, 1989). The energy required to create the droplets will be related to the amount of de novo liquid surface area that is formed during atomization. The surface area is in turn, related to the square of the droplet diameter. Therefore, for a given volume of aerosolized liquid, a small reduction in droplet size, can cause a large increase in the total surface area and this implies a greater transfer of energy to create aerosol. The large air interface produced over time also may induce surface denaturation of surface-active proteins. This can be brought about through conformational changes at the hydrophobic air-water interface. The median droplet size may prove to be one useful indicator of the damage that different nebulizers could inflict on nebulized drug formulations.

3.2. Aerosol mass outputs

The mass output of the nebulizers increases with air pressure but the differences among the nebulizers are quite small in terms of M_t (ml water/min) given the variety of nebulizer designs (Table 2). Larger differences in output are seen in terms of M_f (μl water/l air) (Table 2). Of greater impact than the nebulizer type is the presence of a vent as in the case of the Devilbiss No. 45. M_t is increased approx. 3-fold when the vent is open (Table 2). The effect is due to negative pressure in the proximity of the nebulizer jet. This draws auxiliary air flow into the nebulizer through the vent. This mixes with jet air and carries additional aerosol in the outflow (Mercer et al., 1969).

3.3. Aspiration rates and nebulizer mechanical efficiency

The aspiration rates (Q_t) vary by as much as 12-fold among the nebulizers and this indicates significant differences in their comparative efficiencies despite their relatively similar aerosol outputs (Table 2). No consistent relationship is

noted between Q_t and the driving air pressure for the different devices. Some demonstrate an increase followed by a decrease in Q_t as the air pressure or air flow is increased (Fig. 6A). Others follow an increasing trend throughout the pressure range studied (Fig. 6B). A two-phase choked flow condition may be being exhibited by some of the nebulizers (Morley, 1989). All the nebulizers demonstrate a nonlinear decrease in the aspiration rate as air pressure is increased when data is expressed in terms of the air flow, Q_f (Fig. 7A). This decline in the throughput of liquid relative to the throughput of air as a function of the air pressure is apparently exponential in nature (Table 4, Fig. 7B). The reason for this phenomenon is unknown, but given the diversity of the nebulizers it is quite distinct and markedly different from the Q_t profiles. It is possible, in one sense, that the air is competing with the liquid to escape through the nozzle. As the air flow approaches the velocity of sound, the air orifice will become critical and prevent any increase in the velocity of air at the outlet. However, volume air flow can still increase due to the fact that the air is compressible. Perhaps it is a transitory increase in the density of air that limits the interaction of liquid beyond the gas orifice. This may be sufficient to limit and even decrease the liquid flow as air pressure is increased, as observed (Fig. 6A). Presumably, the same effect might be seen in the other nebulizers if the applied air pressure were increased beyond the range studied. It is interesting to note that the DeVilbiss No. 45, the only nebulizer tested with external, as opposed to internal mixing, liquid feed jets, has Q_f values that are almost independent of air pressure. In this instance there is a single liquid feed outlet positioned at some distance from the gas orifice. As the air has time to expand and accelerates beyond the orifice the interaction with the liquid is relatively unaffected.

Very little information is available on aspiration rates for nebulizers used for inhalation therapy. Hickey and Byron (1987) modified the Bird and Acorn nebulizers to control the liquid feed rate, but they made measurements at very low feed rates of up to 0.6 ml/min. Moreover, the input was controlled by the syringe pump supplying the liquid and not by the gas pressure. Mercer et al. (1968) measured Q_f of the Lovelace, DeVil-

Table 2
Output air flow rates and aerosol mass outputs of the nebulizers

Nebulizer	Air pressure (psig)								
	10			20			40		
	Air flow (l/min)	Mass output (ml/min)	(μl/l)	Air flow (l/min)	Mass output (ml/min)	(μl/l)	Air flow (l/min)	Mass output (ml/min)	(μl/l)
Raindrop	4.9	0.11	21.5	7.2	0.19	25.9	11.7	0.35	29.6
Misty	4.5	0.11	24.1	6.6	0.18	27.5	10.8	0.35	32.8
Aerotech II	4.9	0.09	19.0	7.0	0.14	19.7	11.4	0.20	17.4
Cirrus	4.8	0.10	21.0	6.9	0.20	43.0	11.2	0.37	33.1
Devilbiss No. 45 closed vent	6.9	0.15	21.5	10.2	0.31	30.0	16.8	0.46	27.5
Devilbiss No. 45 [a] open vent	–	0.39	–	–	0.87	–	–	1.20	–
Updraft	2.7	0.05	19.1	4.1	0.17	41.7	6.8	0.32	46.3
Whisperjet [b]	4.2	0.10	22.5	5.9	0.20	33.1	9.4	0.39	40.8
Acorn I	5.2	0.13	25.7	7.4	0.27	36.4	11.9	0.47	39.9
Acorn II	3.6	0.06	15.9	5.2	0.15	23.7	8.4	0.42	50.4
Collison 1-jet	1.7	0.03	15.5	2.5	0.06	22.3	4.2	0.09	21.9
Collison 3-jet	4.9	0.08	16.5	7.3	0.15	20.7	12.0	0.38	31.7

[a] Output air flow rates of the DeVilbiss No. 45 nebulizer were not measured when the vent was open.
[b] Results are the mean values for six Whisperjet nebulizers.

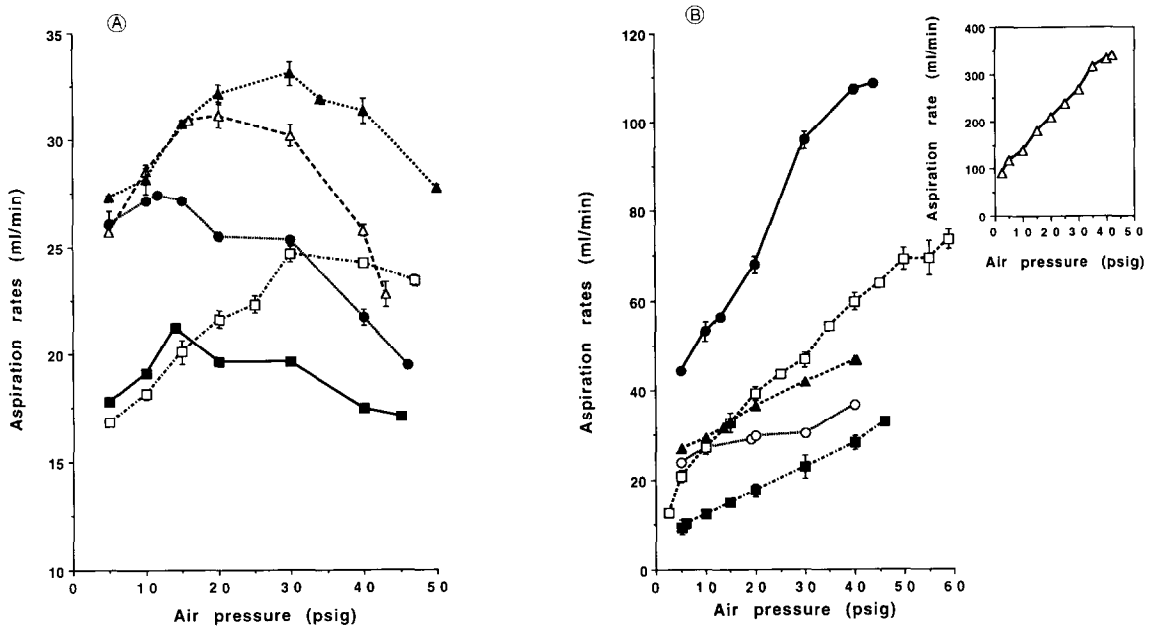

Fig. 6. The effect of air pressure on the aspiration rate (Q_1 ml/min) of nebulizers. (A) The aspiration rates for those nebulizers that exhibit a rise and fall in liquid throughput as air pressure is increased [DHD-Cirrus (●), Puritan-Bennet Raindrop (△) Cadema Aerotech (■), Hudson Updraft (▲), Marquest Acorn II (□)]. (B) The aspiration rates of nebulizers that demonstrate a continual increase in aspiration rate [Collison 1-jet (□), Airlife Misty (●) DeVilbiss No. 45 (■), Marquest Acorn 1 (▲) and Whisperjet (○)]. The inset shows the plot for the Collison 3-jet nebulizer which exhibited higher flows than the other nebulizers. The plots illustrate the tremendous variability in the quantity of fluid uptake as well as the nature of uptake process. Points are the mean of three experiments with each nebulizer. Vertical bars represent the range.

biss and Lauterbach experimental nebulizers, but thought that the values had no relationship to the output of 'useful' aerosol. Liu and Lee (1975) in their discussion of a constant output atomizer note that in modifying a Collison nebulizer they could lower the liquid feed rate from 2.2 ml/min

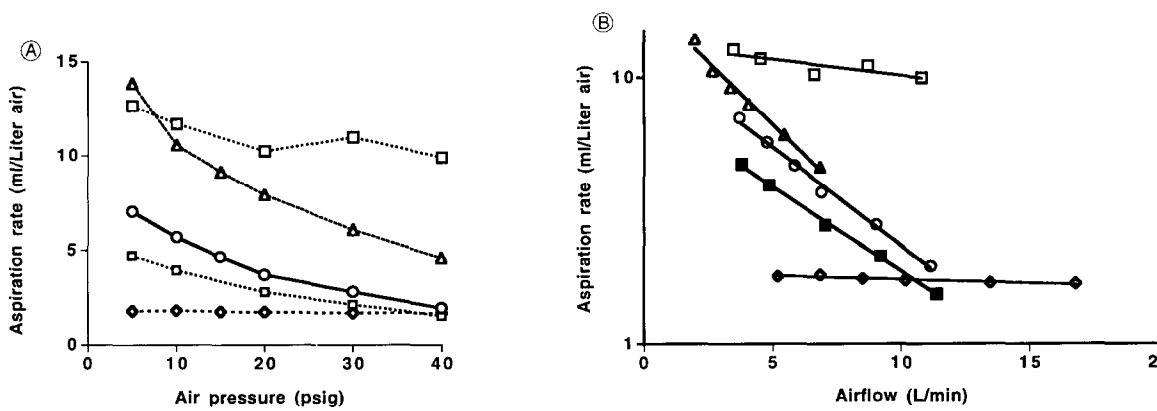

Fig. 7. The effect of air pressure (or air flow) on the aspiration rate (Q_f ml/l air) of nebulizers [DHD-Cirrus (○), Airlife Misty (□), Hudson Updraft (△), DeVilbiss No. 45 (◇), Cadema Aerotech (■)]. (A) The decline in aspiration of fluid as air pressure is increased. (B) The same data replotted on a log-linear plot with the x-axis having units of air flow (l/min). The best fits (broken line) by least-squares regression are plotted though the data. Only five of the nebulizers are illustrated for the purpose of clarity but all the nebulizers tested were linearized in this fashion.

down to nearly 0.59 ml/min without affecting either the mass output or the MMAD of the aerosol at a constant air flow.

All the nebulizers are highly inefficient devices as constructed since only a small fraction of the liquid aspirated from the reservoir emerges as potentially inhalable aerosol. The 'best' nebulizer in this sense was the DeVilbiss No. 45 nebulizer. It was optimal at 20–30 psig and had an E_m of about 1.7% (Table 3). This was improved to over 5% at 30 psig when the vent was open. Presumably this increase in efficiency would be expected if vents were incorporated in the other nebulizers and this would be one simple method of reducing drug damage if it occurred during aerosolization.

Nebulizer efficiencies are important since they are related to the number of times a solute or drug must pass through the nebulizer jet before it can expect to be both aerosolized and exit the nebulizer in a droplet small enough to escape. If 100% efficient then the drug molecules in solution would only be exposed to the potentially damaging effects of aerosolization a single time. As efficiency drops, then the drug in solution can expect to pass through the nebulizer jet an increasing number of times before escaping in the aerosol output.

Table 3
Mean efficiencies of nebulizers

Nebulizer	% efficiency [a] at air pressure (psig):				
	5	10	20	30	40
Puritan Bennett Raindrop	0.20	0.37	0.60	0.92	1.34
Airlife Misty	0.13	0.21	0.27	0.26	0.33
Cadema Aerotech II	0.37	0.48	0.71	0.80	1.14
DHD Cirrus	0.20	0.37	1.16	1.19	1.70
Devilbiss No. 45 closed vent	1.09	1.19	1.74	1.69	1.64
Devilbiss No. 45 open vent	2.61	3.18	4.94	5.18	4.24
Hudson Updraft	0.10	0.18	0.53	0.80	1.01
Marquest Whisperjet [b]	0.19	0.33	0.62	0.93	1.03
Marquest Acorn I	0.29	0.46	0.68	0.83	1.02
Marquest Acorn II	0.16	0.32	0.70	1.00	1.74
Collison 1-jet	0.05	0.06	0.07	0.12	0.11
Collison 3-jet	0.08	0.10	0.14	0.15	0.15

[a] The % mechanical efficiency as defined by the % ratio of the mean aerosol output to the mean aspiration rate.
[b] The mean % mechanical efficiency obtained for six Whisperjet nebulizers.

Table 4
Rate constants for liquid aspiration rates of nebulizers as a function of air flow [a]

Nebulizer	Rate constant ($\times 10^{-2}$min/l air) [b]
Puritan Bennett Raindrop	14.1
Airlife Misty	21.6
Cadema Aerotech II	14.5
DHD Cirrus	17.0
Devilbiss No. 45 closed vent	0.6
Hudson Updraft	2.8
Marquest Whisperjet [b]	12.2
Marquest Acorn I	6.5
Marquest Acorn II	12.0
Collison 1-jet	8.3
Collison 3-jet	3.5

[a] The rate constants were obtained by nonlinear regression of plots of the aspiration rate (ml/l air) vs air flow (l air/min).
[b] The greater the rate constant the faster the decay in aspiration rate relative to the air flow through the nebulizer.

It is proposed that two of the dominant factors influencing how much a drug product is altered by the rigors of aerosolization are (a) the number of times it must pass through the nebulizer jet before leaving the nebulizer and (b) the amount of energy it is exposed to during each passage through the nebulizer jet. Energy can be defined in this case as any energetic effect that could result in destabilization of a drug (e.g., chemical reaction, bond disruption, surface denaturation and so on). In effect, it appears logical to use a highly efficient nebulizer at low air pressures rather than vice versa. Although it is straightforward to produce more efficient nebulizers this is largely at the expense of the small droplet sizes. It should therefore be emphasized that any 'improvement' to nebulizer design must, as a prerequisite, maintain a droplet size distribution acceptable for inhalation therapy.

3.4. Nebulizer intra-variability

Mass outputs for six different Whisperjets ranged from 0.35 to 0.44 ml/min at an air pressure of 40 psig. The mass output increased with

increased air pressure for all devices. The aspiration rates, however, varied from 21.1 to 47.4 ml/min at 40 psig, an approx. 2.25-fold difference. Moreover, there was not a clear linear relationship with driving pressure and this results in sizeable differences in the mechanical efficiencies of the Whisperjet nebulizers. Fig. 8 illustrates the variation of E_m with air pressure for the Whisperjet with the highest and lowest efficiency. If a nebulizer is to be employed in inhalation therapy where there is the likelyhood of drug instability during aerosolization then it will be necessary to manufacture devices with minimal variation in efficiency.

3.5. Effect of nebulization on lactate dehydrogenase

Aerosolization of the LDH at the two volumes studied resulted in an irreversible time-dependent loss of enzymatic activity in the reservoir solution (Fig. 9). The loss of activity was substantially faster in the 10 ml initial volume compared with the 40 ml initial volume. Without prior knowledge of nebulizer operation and Q_t, this effect would be surprising. However, since Q_t at 40 psig is ≈ 300 ml/min it can be recognized that the number of times the reservoir fluid is cycled through the nebulizer jets/min for 10 ml is approx. 4-times more than for the 40 ml volume. This relationship, although not strictly correct because of the effects of aerosol output on volume, and the mixing that occurs in the resorvoir, does serve to illustrate that the LDH in the 10 ml volume is exposed to the nebulizer jets more times and hence more accumulated energy than the LDH in the 40 ml volume. This result also shows that fluid volume and aspiration rate are important determinants of the stability of LDH to nebulization.

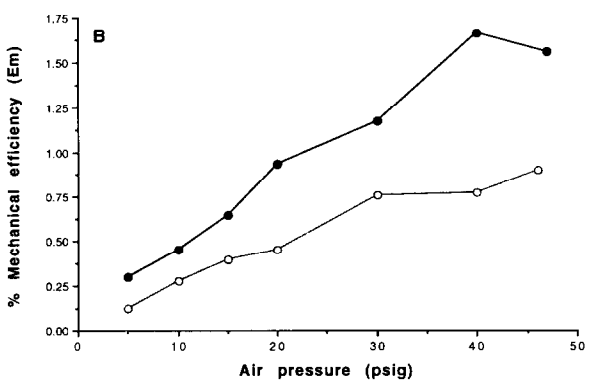

Fig. 8. The maximum (□) and minimum (○) mechanical efficiencies (E_m) observed from testing six Whisperjet nebulizers as a function of the applied air pressure. The results show the $\approx$ 2-fold range in the efficiency of the nebulizers. Points are the mean of three experiments. Vertical bars represent the range.

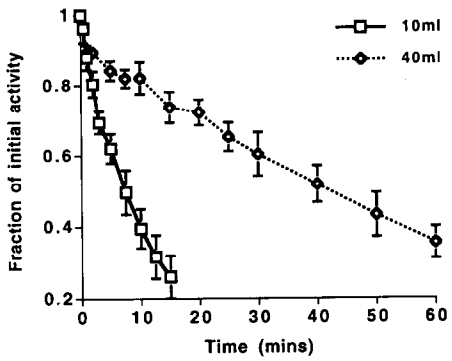

Fig. 9. The effect of nebulization on the stability of lactate dehydrogenase to air-jet nebulization with a 3-jet Collison nebulizer. The enzyme is inactivated during nebulization in a time-dependent manner. The loss of activity is substantially faster using a 10 ml starting volume (□) compared with a 40 ml starting volume (◇). The points represent the mean of ≥ 5 experiments and the error bars are the standard deviation.

4. Conclusions

The results have quantified several parameters of nebulizer function including the aspiration of fluid through the nebulizer jets. Aspiration rates are substantially larger than their respective aerosol outputs and can be dramatically affected by the air flow passing through the gas orifice of the nebulizers. These rates dictate the cycling of fluid within the nebulizer reservoir and are indirectly shown to influence the stability of LDH. In turn, the aspiration rate in conjunction with the aerosol mass outputs, defines the mechanical efficiency of the nebulizers. This is observed to be very low and rarely more than 1%. The median

diameter of droplet sizes decreases as a function of increasing air pressure in all the nebulizers. This information, coupled with the mass output gives some indication of the transfer of energy to the liquid to create droplets. The smaller the droplet size the greater the transfer of energy and production of surface area. Shear effects may damage liposomes, and shear together with surface denaturation may be responsible for the inactivation of the LDH. It is not possible to conclude which effect is important in this study. It is hoped that this work will spur some interest in the study of nebulizer function with respect to drug stability.

5. Acknowledgements

The help of John Thomson and Jim Blanchard while this work was being conducted and written is greatly appreciated. We thank the efforts of Ernie Mitchell for manufacturing the Collison nebulizers. The work was supported by a grant from Merck-Frosst, Canada and NIH grant No. HL019170.

Glossary

psig	pounds per square inch gauge (1 psig = 0.0703 kg/cm^2)
d_{ae}	mass median aerodynamic diameter (μm) of unevaporated droplets
d_p	mass median aerodynamic diameter (μm) of dried particles
κ_s	dynamic shape factor of a cuboidal sodium chloride particle
κ	dynamic shape factor of a droplet
ρ_l	density of the solution forming the droplets (g/ml)
ρ_s	density of the solute (g/ml)
ρ_{sv}	density of the solvent (g/ml)
g_s	weight fraction of solute in the droplets
a_s	mass of solute in a fixed mass of nebulizer solution (g)
a_{sv}	mass of solvent in a fixed mass of nebulizer solution (g)
A_t	output air flow rate of a nebulizer (l air/min)
M_t	aerosol mass output as a function of time (ml/min)
M_f	aerosol mass output as a function of output air flow (μl/l air)
Q_t	aspiration rate as a function of time (ml/min). The throughput of liquid is the quantity of liquid which passes through the nebulizer air jet
Q_f	aspiration as a function of output air flow (ml/l air)
E_m	% ratio of the aerosol mass output to the liquid throughput

6. References

Bangs, L.H., *Uniform Latex Particles*, Seragen Diagnostics, Indianapolis, 1984.

Clay M.M., Pavia D., Newman, S.P. and Clarke, S.W., Factors influencing the size distribution of aerosols from jet nebulizers. *Thorax*, 38 (1983) 755–759.

Dennis, J.H., Stenton, S.C., Beach, J.R., Avery, A.J., Walters, E.H. and Hendrick, D.J., Jet and ultrasonic nebulizer output – Use of a new method for direct measurement of aerosol output. *Thorax*, 45 (1990) 10–16.

Ferron, G.A., Kerrebun, K.F. and Weber, J., Properties of aerosols produced with three nebulizers. *Am. Rev. Respir. Dis.* 114 (1976) 899–908.

Gonda, I. and Byron, P.R., Perspectives on the biopharmacy of inhalation aerosols. *Drug Dev. Ind. Pharm.*, 4 (1978) 243–259.

Handbook of Physics and Chemistry, 61st Edn, CRC Press, Boca Raton, 1980, p. B147.

Hickey, A.J. and Byron, P.R., Effect of solution flow rate on the output of two modified commercially available jet nebulizers. *J. Pharm. Sci.*, 76 (1987) 338–340.

Lefebvre, A.H., *Atomization and Sprays*, Hemisphere, New York, 1989, pp. 65–68.

Liu, B.Y.H. and Lee, K.W., An aerosol generator of high stability. *Am. Ind. Hyg. Assoc. J.*, 36 (1975) 861–865.

May, K.R., The Collison nebulizer: description, performance and application. *J. Aerosol. Sci.*, 4 (1973) 235–243.

Mercer, T.T., Output characteristics of several commercial nebulizers. *Ann. Allergy*, 23 (1965) 314–326.

Mercer, T.T., Goddard, R.F. and Flores, R.L., Effect of auxiliary air flow on the output characteristics of compressed-air nebulizers. *Ann. Allergy*, 27 (1969) 211–217.

Mercer, T.T., Tillery, M.I. and Chow, H.Y., Operating characteristics of some compressed air nebulizers. *Am. Ind. Hyg. Assoc. J.*, 29 (1968) 66–78.

Miller, R.F., Godfrey-Faussett, P. and Semple, S.J.G., Nebulized pentamidine as treatment for *Pneumocystis carinii* pneumonia in the acquired immunodeficiency syndrome. *Thorax*, 44 (1990) 565–569.

Morley, P.G., Sizing pressure safety valves for flashing liquid duty. *Chem. Eng.*, October (1989) 47–49.

Newman, S.P., Pellow, P.G.D. and Clarke, S.W., Droplet size distributions of nebulized aerosols for inhalation therapy. *Clin. Phys. Physiol. Meas.*, 7 (1986) 139–146.

Niven, R.W., Delivery of biotherapeutics by inhalation aerosols. *Pharm. Tech.*, 17 (1993) 72–82.

Niven, R.W. and Schreier, H., Nebulization of liposomes: I.

Effects of lipid composition. *Pharm. Res.*, 7 (1990) 1127–1133.

Rizk, N.K. and Lefebvre, A.H., Airblast atomization: studies on drop-size distribution. *J. Energ.*, 6 (1982) 323–327.

Ryan, G., Dolovich, M.B., Obminski, G., Cockcroft, D.W., Juniper, E., Hargreave, F.E. and Newhouse, M.T., Standardization of inhalation provocation tests: influence of nebulizer output, particle size, and method of inhalation. *J. Allergy Clin. Immunol.*, 67 (1981) 156–161.

Sato, Y., Sato, M., Kita, M. and Kishida, T., A comparison of 28 kHz and 160 kHz ultrasonic aerosolization of interferon-α. *J. Aerosol Med.*, 5 (1992) 59–64.

Smaldone, G.C., Perry, R.J. and Deutsch, D.G., Characteristics of nebulizers used in the treatment of AIDS-related *Pneumocystis carinii* pneumonia. *J. Aerosol Med.*, 1 (1988) 113126.

Takanami, C. and Goto, Y., Physical properties of antibiotic aerosols produced by jet and ultrasonic nebulizers. *J. Aerosol Med.*, 3 (1990) 45–52.

Taylor, K.M.G., Taylor, G., Kellaway, I.W. and Stevens, J., The stability of liposomes to nebulization. *Int. J. Pharm.*, 58 (1990) 57–61.

Waldman, D.L., Weber, D.A., Oberdörster, G., Drago, S.R., Utell, M.J.,Hyde, R.W. and Morrow, P.E., Chemical breakdown of technetium-99m DTPA during nebulization. *J. Nucl. Med.*, 28 (1987) 378–382.