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Correlations between cascade impactor analysis and laser diffraction techniques for the determination of the particle size of aerosolised powder formulations

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ABSTRACT

The purpose of the study was to examine the suitability of the Spraytec® laser diffraction technique for measuring the size distribution of aerosol particles generated from dry powder inhalators. A range of formulations with different dispersion properties were produced by spray-drying. The percentage of particles below 5.0 μm of these formulations was measured by laser diffraction (Mastersizer® 2000 and Spraytec®) and inertial impaction (MsLI and NGI) using various inhaler devices and at different flow rates between 30 and 100 l/min. Linear relationships and correlations ($R^2 > 0.9$) existed between the results obtained from, on one hand, the Mastersizer® 2000 and the Spraytec®, and, on the other hand, the MsLI and the Spraytec® regardless of flow rates and inhaler devices. The Spraytec® could be a reliable technique for the development, evaluation and quality control of dry powder aerosol formulations.

1. Introduction

Particle size is a key parameter in defining the deposition pattern and bioavalability of drug material delivered to the respiratory system using inhalers. Therefore, in order to deliver a drug to the lungs in an efficacious manner, it is believed that the optimum particle size for deposition into the pulmonary system should be between 1 and 5 μm (Heyder et al., 1986; Elversson et al., 2003; Bosquillon et al., 2004).

The aerodynamic diameter, which is defined as the diameter of a sphere with a unit density, that has the same terminal settling velocity in still air as the particle in consideration (De Boer et al., 2002) is routinely measured by sizing techniques that are based on inertial impaction (Heyder et al., 1986). Inertial impaction of particles in movement in an air stream is well understood and can be controlled. Many different types of impactors and impingers have been proposed for inhalation aerosols in the last decades. They vary from a simple device, such as the glass impinger or the twin impinger, to more complex apparatus that have more collection plates, such as the Andersen cascade impactor (ACI), the multi-stage liquid impinger (MsLI) or the recently developed new generation impactor (NGI). Inertial impactors have to be designed and constructed according to certain aerodynamic rules, such that they must be operated under strictly defined conditions in order to obtain the desired cut-off efficiencies and to avoid excessive flattening of the cut-off curve (De Boer et al., 2002). Impingers and impactors have been widely employed for particle size characterization.

Nevertheless, laser diffraction is the most widely used technique for particle size analysis. Instruments employing this technique are considered easy to use and particularly attractive for their capability in analysing particles over a broad size range in a variety of dispersion media (Clark, 1995). However, laser diffraction provides measurement of geometrical instead of aerodynamic particle size and the apparent particle density and dynamic shape factors of drug agglomerates are not considered. Nevertheless, laser diffraction has been successfully employed to examine aerosols for inhalation from nebulisers (Clark, 1995; Mc Callion et al., 1996; Bridges and Taylor, 1998) and pressurised metered dose inhalers (pMDI) (Moren, 1981), although its application to dry powder inhalers (DPI) has been little studied. Even so, the increasing popularity of DPI in the development of drug pulmonary administration needs new advances in the evaluation of the particle size distribution of the aerolized formulation (Olsson et al., 1988; Martin et al., 2006; Zeng et al., 2006).

It is therefore the purpose of this study to establish whether a Spraytec® laser diffraction method has the potential to characterize

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the properties of various dry powder aerosol formulations and to compare the results with those obtained from inertial impaction methods. Dispersion properties of various dry powder formulations were investigated using different laser diffraction and impaction apparatus at different flow rates and using different inhalator devices.

2. Materials and methods

2.1. Materials

Tobramycin was supplied as micronized powder from Plantex Chemicals B.V. (The Netherlands).

Cholesterol was purchased from Merck (Belgium). Phospholipon 90H (Ph90H), hydrogenated soy lecithin, with more than 90% hydrogenated phosphatidylcholine, consisting of approximately 85% distearoyl phosphatidylcholine and 15% dipalmitoyl phosphatidylcholine, was donated by Nattermann Phospholipids GmbH (Koln, Germany).

All chemicals used were of analytical grade.

2.2. Methods

2.2.1. Production of the dry powders

Dry powders considered as "carrier-free" presenting different degrees of particle aggregation were prepared by spray-drying (Büchi Mini Spray Dryer B-191a, Büchi laboratory-Techniques, Switzerland) various suspensions of tobramycin in isopropanol.

Briefly, various concentrations of lipids were dissolved in micronized tobramycin suspensions which were spray-dried at 70 °C, resulting in a coating of the drug particles and in a modification of their surface and agglomeration tendency (Formulations F1) (Pilcer et al., 2006). On the other hand, micronized tobramycin was added to a solvent system composed of isopropanol:water 80:20 and spray-dried at various temperatures (from 120 °C to 200 °C) leading to differences in residual humidity content and various aggregation and dispersion properties (Formulations F2). Powders were stored in a dessicator at ambient temperature.

Table 1 gives an overview comparison of all powders studied. All the powders presented a low bulk powder tap density (<0.3 g/cm^3), and sizes theoretically suited for administration to the deep lungs. In both cases, all powder formulations were considered as homogeneous and presented a Gaussian size distribution curve (log normal).

2.2.2. Particle size characterization by laser diffraction

The volume particle size distribution was measured using a Malvern Mastersizer 2000® laser diffractometer using a dry sampling system (Scirocco, 2000, Malvern, UK) with a suitable Standard Operating Procedure (SOP) (refractive index: 1.52, vibration feed rate: 25%, measurement time: 7 s, dispersive air pressure: 4 bar).

The particle size distribution was characterized by the mass median diameter (D (0.5)), i.e. the size in microns at which 50% of the sample is smaller and 50% is larger. Values presented are the average of at least 3 determinations.

2.2.3. Simultaneous characterization by inertial impaction and laser diffraction

The second laser diffraction-based technique consisted of a Malvern Spraytec® (Malvern, UK) equipped with an inhalation cell, specifically modified for measuring the particle size diameter (PSD) generated from medicinal aerosols, including MDI, DPI and nebulizers. It consists of a Spraytec® unit with a throat held in place by the inhalation cell and a connection for an MsLI. The entire assembly is a closed system and allows for a controlled airflow rate in the measurement zone. This allows the size properties of DPIs to be measured under simulated breathing conditions (Haynes et al., 2004). The difference between the Mastersizer 2000® and the Spraytec® is the particle dispersion capacity of the two methods. The compressed air values applied in the dispersion unit of the Mastersizer 2000® (up to 4 bar), allows all the agglomerates to break down whereas the airflow generated in the Spraytec® is much lower and does not permit the de-agglomeration of all particles.

The focal length of the Spraytec® lens used was 100 mm, which has a particle size range of 0.5–200 μm. The fine particle fraction was obtained from the Spraytec® software based on the percentage of particles having a diameter of <5.0 μm. Values presented are the average of at least 3 determinations.

The assembly was connected to an impactor in order to allow the same aerosol, generated under inspiratory airflow, to pass through both the laser beam crossing the end of the metal throat and the stages of the inertial impactor. Sizing could therefore be carried out using the two techniques in-line. Importantly, this modification does not change the determination of the size results from the MsLI. Thus, this assembly allows the simultaneous determination of the geometric (laser diffraction) and aerodynamic (MsLI) diameters.

2.2.4. Aerodynamic particle size analysis

The aerodynamic particle size distribution was measured by either an MsLI or an NGI. The MsLI and NGI were operated under pharmacopoeial conditions. The flow rate was adjusted to a pressure drop of 4 kPa, as is typical for inspiration by a patient. Dry powder inhalation devices (Aerolizer®, Novartis; Spinhaler®, Sanofi-Aventis and Handihaler®, Boeringher Ingelheim) were filled with HPMC no. 3 capsules (Capsugel, France). Three capsules loaded with 15 mg powder were taken for each test. Drug deposition

Table 1
Composition of the spray-dried suspensions F1 and F2 formulations

	Suspensions		Ratio cholesterol/phospholipon (%)	Inlet temperature (°C)
	Tobramycin (%, w/v)	Lipids (%, w/v)		
F1.1	5	0.1	75/25	70
F1.2	5	0.25	66/34	70
F1.3	5	0.25	90/10	70
F1.4	5	0.5	75/25	70
F1.5	2	0.1	75/25	70
F1.6	10	0.5	75/25	70
F1.7	5	0.25	75/25	70
F2.1	5	/	/	120
F2.2	5	/	/	140
F2.3	5	/	/	160
F2.4	5	/	/	180
F2.5	5	/	/	200

in the device, the throat, all the stages and the filter was determined by high-pressure liquid chromatography (HPLC) analysis. For accuracy, each test was repeated three times. The suitable and validated quantification method was previously described (Pilcer et al., 2006).

The total dose of particles with aerodynamic diameters smaller than 5.0 μm was calculated by interpolation from the cumulative mass against cut-off diameter of the respective stages and considered as the fine particle dose (FPD) (mg) or fine particle fraction (FPF), expressed as a percentage of the emitted dose (ED). The ED was determined as the percentage of total powder mass exiting the capsule and device. The mass median aerodynamic diameter (MMAD) of the particles was defined from the same plot as the particle size at which the line crosses the 50% mark. This parameter takes account of particle aggregates and gives therefore the ultimate aerodynamic behaviour of the aerosol. It brings information on dry powder cohesiveness and aerosolization properties.

2.2.5. Validation of the acquisition, computation and comparison of the different sets of data

In practice, the quality of the analytical input data is crucial for interpretation of method comparison studies. A first criterion generally used is the correlation coefficient R^2 which measures the strength of a linear relation between two variables, not the agreement between them (Bland and Altman, 1986). Nevertheless, as R^2 is still commonly used, it will be computed in this study. A second method is to draw a comparison plot (one method per axis) coupled to a linear regression study (Westgard, 1998). Ordinary least-square regression (OLR) has the disadvantage of assuming an error-free x variable and a constant analytical imprecision of the y variable (homoscedatic variance), assumptions that are seldom met in practice (Stockl et al., 1998). If both measurements sets are subject to random errors, an alternative to OLR is the Deming regression computed in this study, also named "principal component analysis", which requires specifications of the ratio between variances of both analytical methods (Linnet, 1990, 1998; Stockl et al., 1998).

3. Results

3.1. Particle size distribution by laser diffraction

The particle size distribution results of the F1 and F2 formulations are presented in Table 2. All the particle size distributions of the F1 and F2 formulations obtained from the Mastersizer 2000® were unimodal and narrow. The measured particle size distributions of these formulations were found to be accurate, with errors in geometric mean diameter measurements in the order of 10%, as reported previously in the sizing of narrow unimodal systems of similar particle size (Annapragada and Adjei, 1996).

Table 2
Particle size characteristics of the F1 and F2 formulations D (0.5) and % <5.0 μm (mean ± S.D., n = 3), measured with the Mastersizer® 2000 laser diffractometer in dry powder form and with the Spraytec®

	D (0.5)		%<5.0 μm	
	Mastersizer®	Spraytec®	Mastersizer®	Spraytec®
F1.1	1.27 ± 0.01	3.0 ± 0.5	99.6 ± 0.1	88 ± 1
F1.2	1.38 ± 0.02	2.6 ± 0.9	99.8 ± 0.1	90.8 ± 0.3
F1.3	1.29 ± 0.01	3.3 ± 0.6	99.6 ± 0.1	90.3 ± 0.5
F1.4	1.38 ± 0.03	2.9 ± 0.4	99.9 ± 0.1	92 ± 1
F1.5	1.24 ± 0.02	3.3 ± 0.8	99.8 ± 0.1	93.7 ± 0.1
F1.6	1.28 ± 0.03	3.4 ± 0.7	99.7 ± 0.1	96.1 ± 0.9
F1.7	1.23 ± 0.01	3.3 ± 0.9	99.6 ± 0.1	98 ± 1
F2.1	2.54 ± 0.02	35 ± 6	88 ± 1	36 ± 6
F2.2	2.47 ± 0.05	18 ± 2	93 ± 1	59 ± 9
F2.3	1.6 ± 0.1	7 ± 1	96.4 ± 0.7	79 ± 6
F2.4	1.41 ± 0.08	2.0 ± 0.8	97.3 ± 0.3	84 ± 9
F2.5	1.6 ± 0.1	1.8 ± 0.1	98.33 ± 0.06	90.6 ± 0.3

The median particle sizes for F1 appeared to be similar for all powder formulations exhibiting a D (0.5) value of about 1.2–1.4 μm with Mastersizer 2000® and about 2.6–3.4 μm with Spraytec®. For the F2 formulations, the median particle sizes appeared to be different for all powder formulations exhibiting a D (0.5) value of about 1.4–2.6 μm with the Mastersizer 2000® and about 1.4–35.0 μm with the Spraytec®. Concerning F2.1, the D (0.5) measured with the Spraytec® appeared to be twelve times greater than that measured with the Mastersizer 2000®. As we can see in Table 2, the problems of agglomeration of particles are more noticeable with the Spraytec® than with Mastersizer 2000®. The differences in the size determination results obtained by the two laser diffraction methods can be explained by the differences in the particle dispersion capacity of the two methods used. The higher compressed air values applied in the dispersion unit of the Mastersizer 2000® (up to 4 bar), permitted all the agglomerates to break down, especially for micron-sized powders, as is the case for DPI formulations. For all the powders investigated, the mass median diameter measured by the Mastersizer 2000® in the dry state rapidly decreased with increased pressure of compressed air used to de-aggregate the powder. Moderately to strongly aggregate powders could be dispersed by a pressure between 3 and 5 bars (Bosquillon et al., 2001). 4 bars presented a good compromise between separation of the agglomerates and breaking of the particles. In contrast, the airflow generated in the Spraytec® was much lower and did not allow the de-agglomeration of all particles. In fact, the entire assembly is a closed system and allowed for a controlled airflow rate (100 l/min during 2.4 s) in the measurement zone. This permitted the size properties of DPIs to be measured under simulated breathing conditions. In other words, the Mastersizer 2000® permitted the determination of the size characteristics of totally individualized particles, whereas size results obtained from the Spraytec® included the presence of some agglomerates such as it appears under the normal conditions of use of an inhaler by a patient.

As can be shown from these results, the behaviour of the two types of Formulations F1 and F2 are very different. For the F2 formulations, increasing the spray-dried temperature allowed a decrease of the moisture content of the formulations, which had implications in aggregation and dispersion properties. Moreover, the application of a lipid coating around the active particles (F1 formulations) allowed an improvement in the particle dispersion from the inhalator, thus enhancing the drug deposition deep in the lungs (loose agglomerates were easily scattered into small particles).

For the F1 formulations, there was no correlation between the results obtained from the Mastersizer 2000® and the Spraytec®, probably due to the fact that the size characteristics of the different powder formulations were very close: for example, size results obtained with the Mastersizer 2000®, at 4 bar, showed that the percentage of particles below 5 μm range from 99.3% to 99.9%. Nevertheless, after computation of the Deming regression, linear relationships between the most important size characterization parameters, i.e. D (0.5) (data not shown) and percentage of particles below 5.0 μm (Fig. 1), could be demonstrated between the results obtained with the Mastersizer 2000® and the Spraytec® for F2 formulations. The best index of correlation (R^2: 0.99) was obtained for the percentage of particles below 5.0 μm, which is the size range that is considered to be "respirable".

3.2. Particle size distribution by inertial impaction

The most effective Formulation (F1.7 and F2.5) of each of the two types of formulations produced were tested with an MsLI and an NGI apparatus at 100 l/min. The latter impactor has been designed

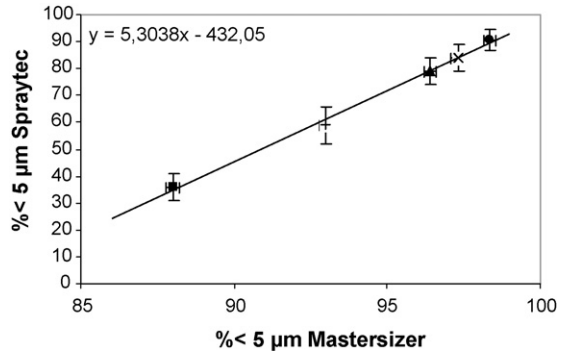

Fig. 1. Deming regression of the percentage of particles under 5 μm measured by the Spraytec® (mean ± S.D., $n = 3$) against the percentage of particles under 5 μm measured by the Mastersizer® 2000 (mean ± S.D., $n = 3$) for the F2 formulations: ■ F2.1, + F2.2, ▲ F2.3, × F2.4, ● F2.5.

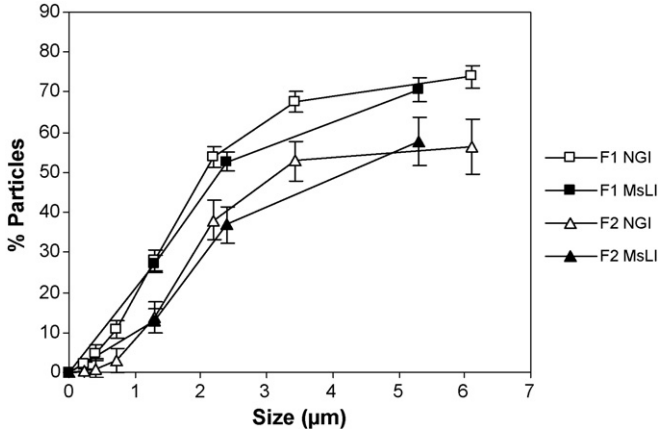

Fig. 2. Aerodynamic particle size distributions for F1.7 and F2.5 determined by MsLI (mean ± S.D., $n = 3$) and NGI (mean ± S.D., $n = 3$) for particles below 6 μm.

to provide pharmaceutically appropriate stage cut points and stage collection efficiency curves that, unlike the ACI, do not overlap significantly at volumetric airflow rates between 30 and 100 l/min (Kamiya et al., 2004). This should enable more accurate PSD determination of the delivered doses from dry powder inhalers.

The fine particle fraction for F1.7 was about 77.3 ± 0.9% with the MsLI and about 76.6 ± 0.6% for the NGI. The fine particle fraction for F2.5 was about 60.3 ± 2.0% with the MsLI and about 62.5 ± 2.3% for the NGI. These results are significantly similar ($p > 0.05$). As can be seen in Fig. 2, the cumulative percent undersize curves of the two impactors were similar.

However, the reduced number of stages of the MsLI may cause an approximation in the evaluation of the particle size distribution of powders, especially in the smaller sizes ranging from 1 to 3.5 μm. The principal difference between the two apparatus is the number of stages: the MsLI is divided in 5 stages with cut-off diameters at 100 l/min about 5.27, 2.40 and 1.32 μm at stages 3, 4 and 5, respectively. In contrast, the NGI has 7 stages, of which 6 have a cut-off diameter below 6.0 μm at 100 l/min: 6.12, 3.42, 2.18, 1.31, 0.72, 0.40 and 0.24 μm. This allows a more detailed particle size distribution, in particular for small particles. Nevertheless, the manipulation of the NGI is more time-consuming than the manipulation of the MsLI.

3.3. Correlation between inertial impaction and Spraytec® laser diffraction

The aerodynamic diameter of a particle, d_{aer}, is related to its geometric diameter, d_g, as well as density, ρ_p, according to the formula: $d_g = d_{aer}\sqrt{\rho_p/\rho_1}$ where $\rho_1 = 1\ g/cm^3$. This equation assumes that the particles are spherical and slip correction factors are insignificant (Crowder et al., 2002). Nevertheless, it has been demonstrated that the physical implications of slip, shape and density on aerosol behaviour are of extreme importance. Without correction for these factors, calculation of aerodynamic particle properties and deposition probabilities would be inaccurate (Crowder et al., 2002). Of primary interest is the use of the particle density in the calculation of a particle's aerodynamic diameter. The particle density is often evaluated by tap density measurements. Nevertheless, assuming efficient packing, the tap density of a monodisperse assay of spheres underestimates by 21% the true particle density because of the void spaces between particles (Vanbever et al., 1999). Considering the difficulty of a true measurement of the particle's density and the approximation of the shape and slip factors, no conversion of the geometric diameter in aerodynamic diameter has been performed before two sets of data.

In order to evaluate the Spraytec® as a reliable measuring instrument for characterizing powders for inhalation, the MMAD for tobramycin particles measured by inertial impaction were compared with and plotted against the D (0.5) measured by laser diffraction. As can be seen from Fig. 3, a linear relationship calculated with the Deming regression existed between the MMAD and the D (0.5) between, on one hand, the F1 formulations (R^2: 0.94) and, on the other hand, between the F2 formulations (R^2: 0.93). Similar correlations for the F1 and F2 formulations for the percentage of particles below 1.3 μm, between 1.3 and 2.4 μm and between 2.4 and 5.3 μm corresponding to the three last stages of the MsLI (stages 3, 4 and 5) have also been demonstrated (data not shown). The R^2 were ranged between 0.90 and 0.98. The existence of these linear relationships demonstrated that the results of the Spraytec®

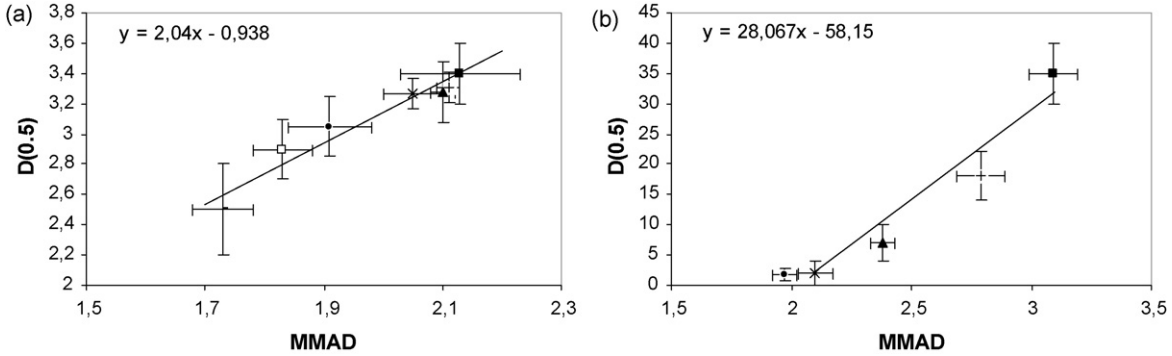

Fig. 3. Deming regressions between the MMAD measured by the MsLI (mean ± S.D., $n = 3$) and the D (0.5) measured by the Spraytec (mean ± S.D., $n = 3$) for (a) F1 formulations: ■ F1.1, + F1.2, ▲ F1.3, × F1.4, ● F1.5, □ F1.6, ■ F1.7 and (b) F2 formulations:: ■ F2.1, + F2.2, ▲ F2.3, × F2.4, ● F2.5.

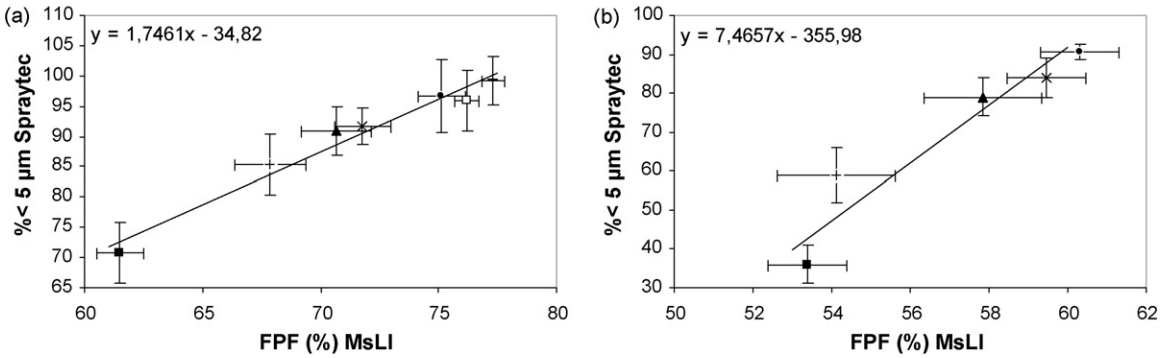

Fig. 4. Deming regressions between the percentages of particles below 5 μm measured by the MsLI (mean ± S.D., $n = 3$) and the Spraytec (mean ± S.D., $n = 3$) for (a) F1 formulations: ■ F1.1, + F1.2, ▲ F1.3, × F1.4, ● F1.5, □ F1.6, ■ F1.7 and (b) F2 formulations:: ■ F2.1, + F2.2, ▲ F2.3, × F2.4, ● F2.5.

are useful for size evaluation of particles below 5 μm, which were considered to be the respirable fraction. Of particular interest for pulmonary delivery, regression analysis of the data from the FPF of F1 and F2 formulations demonstrated a good correlation (R^2 of 0.96 and 0.90, respectively) between the two techniques (Fig. 4).

In order to evaluate if it is possible to determine the FPF of dry powders from this correlation, 5 different batches of the F1.7 formulation were tested with the Spraytec® and the MsLI. Table 3 gives a comparison of the experimental FPF determined by the MsLI and the theoretical FPF calculating from the equation of the Deming regression $y = 1.7461x - 34.82$ (Fig. 4), where y is the percentage of particles below 5.0 μm measured by the Spraytec® and x, the FPF measured by the MsLI. These results show that the experimental and the theoretical FPF appeared to be statistically similar ($p > 0.05$) and that the Spraytec® could quickly provide a very good evaluation of the aerodynamic behaviour of the powders produced.

A similar correlation was also demonstrated between the results of the Spraytec® and the NGI (data not shown), showing that the simultaneous measurements of laser diffraction and impaction could be provided with different impaction apparatus.

The shift of the values obtained by the two methods (laser diffraction vs. impaction) can be explained, as mentioned above, by the influence of slip, shape and, especially, density of the particles. As expected, the d_g presented higher values than the d_{aer}, since the density of the particles is lower than 1 g/cm^3. This implication of the combination of physicochemical factors was also found in the fact that it proved to be impossible to have a correlation among both of the Formulations F1 and F2 because the surface morphology and the density of the two types of formulations appeared to be too different.

The quality of a DPI product is determined by the formulation, device design and interaction between these two factors. Since different devices can be used to deliver the same active ingredient, it is still common practice to screen a large number of candidate formulations and devices before identifying the most suitable formulation for a specific and/or selected device (Kamiya et al., 2004).

Table 3
Comparison of the experimental and theoretical FPF of 5 different F1.7 batches (mean ± S.D., $n = 5$)

Batches	%<5.0 μm (Spraytec)	Experimental FPF (MsLI)	Theoretical FPF	
1	97.6 ± 1.0	76.8 ± 1.0	75.8 ± 0.6	$p > 0.05$
2	96.7 ± 0.7	75.7 ± 0.9	75.3 ± 0.4	$p > 0.05$
3	98.7 ± 0.8	76.9 ± 0.3	76.6 ± 0.4	$p > 0.05$
4	98.0 ± 1.1	75.7 ± 0.5	76.5 ± 0.6	$p > 0.05$
5	95.5 ± 1.0	74.3 ± 0.8	74.6 ± 0.6	$p > 0.05$

Tests with the Aerolizer®, the Spinhaler® and the Handihaler® were carried out. These devices operate by dispensing drug contained in a capsule, from either a spinning (Aerolizer® and Spinhaler®) or rumbling (Handihaler®) motion, once the capsule has been opened by piercing pins. The particles are dispersed through the turbulence generated by spinning blades (Aerolizer®, Spinhaler®) and/or a plastic grid (Aerolizer®, Handihaler®) at the time of inhalation.

The results of the particle deposition of the F1.7 formulation measured by the MsLI, revealed that the Aerolizer® seemed to be the most effective device, with a FPD about 9.7 ± 0.1 mg, in comparison to the Spinhaler® (8.14 ± 0.3 mg) and the Handihaler® (6.18 ± 0.6 mg). The Spinhaler® presented a deposition profile slightly different from the two others, with a deposition level more elevated in the throat and in the stage 4. The Handihaler® had the highest level of retention of the powder in the capsule and device (3.50 mg). Indeed, in contrast to the Aerolizer® and the Spinhaler®, the capsule did not whirl during inhalation and particles were less dispersed in the airstream.

Regression analysis of the data from the percentage of particles below 5.0 μm and the FPF of F1.7 with the different inhalers demonstrated a linear relationship with good correlation (R^2: 0.99) (Fig. 5). The Spraytec® can thus be a useful tool for rapid screening for the most suitable device for a specific formulation.

Experiments at different flow rates (30, 40, 60 and 100 l/min) were carried out on the F1.7 formulation with the Aerolizer® in order to evaluate the influence of the airflow on the dispersion of particles and on the measurements from the Spraytec®.

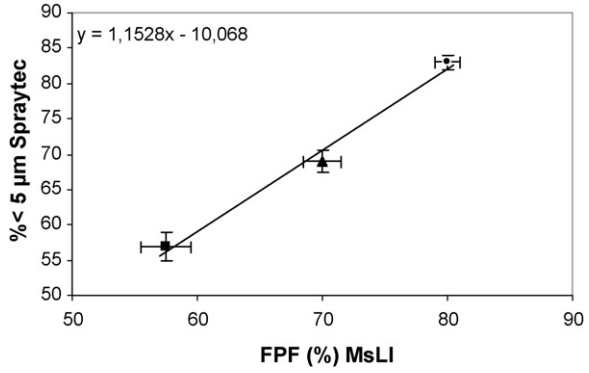

Fig. 5. Deming regression of the tobramycin FPF obtained by the MsLI (mean ± S.D., $n = 3$) plotted as a function of corresponding FPF obtained by the Spraytec® (mean ± S.D., $n = 3$) for F1.7 formulation: aerosol generated by ● the Aerolizer®, ▲ the Spinhaler® and the ■ Handihaler® devices.

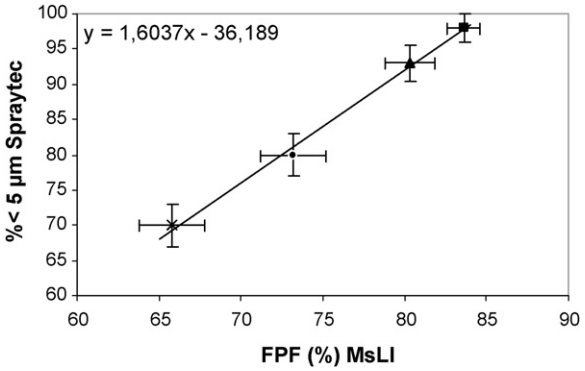

Fig. 6. Deming regression of the tobramycin FPF obtained by the MsLI (mean ± S.D., $n = 3$) plotted as a function of the corresponding fine fraction obtained by the Spraytec® (mean ± S.D., $n = 3$) for F1.7 formulation with the Aerolizer generated at × 30, ● 40, ▲ 60 and ■ 100 l/min.

The FPD appeared to decrease with reduction of the flow rate. The FPD decreased from 9.6 mg at 100 l/min to 6.4 mg at 30 l/min. The more the flow decreased, the more the powder was retained inside the capsule and device (2.2 mg vs. 4.5 mg) and a lower deposition of tobramycin was observed in the stages 3, 4 and 5. These results may suggest that a flow rate superior to 60 l/min is necessary for the breakdown of most of the drug particle agglomerates. Deming regression analysis of the data from the percentage of particles below 5.0 μm and the FPF of the F1.7 formulations with the Aerolizer® at different flow rates demonstrated a linear relationship with a good correlation (R^2: 0.99) (Fig. 6). So, as opposed to other laser diffraction techniques, the Spraytec® presents the advantage that the analysis of particle size distribution depends on controlled flow rates which are near those produced by a patient. This provides more realistic results of the tendency of the powder to de-agglomerate during inhalation.

4. Discussion

The aerodynamic particle size diameter is routinely measured using sizing techniques that are based on inertial impaction. The principle of classification by inertial separation is well-established and different types of apparatus such as the MsLI, the ACI and the NGI, and test procedures have been adopted by European and US Pharmacopoeias. Impingers and impactors have been widely employed for product development. However, such impaction techniques are invariably laborious and time-consuming to operate and thus are not the best choice for screening many candidate formulations during the early stages of product and process development. Moreover, in the last decade, cascade impactor analysis has been subjected to critical evaluations and some developments in their use (Hickey, 1990): High inter- and intra-laboratory variations have been described with impactors of the same design. Variation for the fine particle dose may be quite large, ranging from 5.5 to 20% for the DPIs (Olsson et al., 1996). Comparison of results from different types of impactors with different upper class limits for fine particle fractions at the same flow rate are even more problematic. This is especially so when the number of size classes is low and the composition of a cumulative mass distribution curve as a function of particle diameter is impossible (Marriott et al., 2006).

Therefore, alternative techniques need to be identified in order to cope with the limitations of inertial impaction techniques. Laser diffraction may prove to be such a technique since it is fast, reproducible and, above all, offers a much higher number of size classes for the relevant fine particle fractions that can be obtained from inertial impaction. However, the use of laser diffraction to characterize dry powder aerosol is limited for many reasons. First, most dry powder aerosol formulations are composed of micronized drug blended with a coarse carrier. Since the carrier almost always contains small particles that are similar in size to the drug, it is impossible to differentiate between drug and fine carrier particles. Second, particle size measurement by laser diffraction is based on the assumption that the particles are spherical. Nevertheless, for micronized particles, deviation from sphericity could be considered as negligible (Marriott et al., 2006). Finally, the method provides data on geometric instead of aerodynamic diameter and the transformation of the results requires a good knowledge of the density and the shape factor of the particles. So, it requires good understanding of the working principle of a DPI and the properties of powder formulations for inhalation to draw correct conclusions. This could limit the application of laser diffraction for DPI development. Nevertheless, different correlations between geometric and aerodynamic size data have been demonstrated in this study. Within the flow rate, the different inhalation devices and the drug formulations examined in this work, the tobramycin fine fraction could be predicted from measurements obtained from the laser diffraction technique using one linear relationship for one type of formulation. Indeed, a combination of physicochemical properties, particle size, density, shape, surface area, and morphology affects the forces of interaction between the drug particles, and these can subsequently change the aerodynamic behaviour of the powder (Telko and Hickey, 2005). Therefore, it appears that it is not possible to predict with exactitude the fine particle fraction of all inhalation powders with only one linear relationship since the inhaled drug properties vary not only in size distribution but also in particle density, shape and velocity (De Boer et al., 2002). Deposition of the drug depends upon a complex interaction between the device, the formulation, and the patient, who controls the flow rate of inhaled air through the system. Therefore, the Spraytec® may be a very useful technique in pharmaceutical development for screening many formulations, devices and flow rates because the particle size distributions of powders from the Spraytec®, as opposed to the conventional laser diffraction method, are dependent on those factors that also influence the fine particle fraction. Consequently, the laser diffraction technique has been proved to be an important tool for initial formulation and process screening for one specific type of formulation. Moreover, the use of the Spraytec® could be interesting especially in process control and quality control of finished products as it allows a rapid screening of many products. It is a robust technique that is capable of conducting in-line measurement of particle size distribution to ensure that a predefined quality can be achieved at the end of the manufacturing process.

5. Conclusion

The aerodynamic particle size distribution of aerolized drugs is an essential parameter to evaluate in formulation screening and the subsequent quality control of the final product. In this paper, the applicability of the laser diffraction technique was evaluated as an alternative, not a substitute, for cascade impactor analysis for in vitro characterization of inhalation particles. The method has the potential to solve some of the major problems related to cascade impactor analysis as it quickly permitted the generation of a sizing parameter, corresponding to the aerosol fine fraction. The most useful features of laser diffraction are time savings, reproducibility, high size resolution and automatic data recording and processing, which could be very interesting and useful in product development, the production and quality control of inhalation products.

In this study, only “carrier-free” dry powders were tested. As most dry powder inhaler formulations are binary interactive mixtures composed of micronized drug blended with a coarse carrier,

it would be interesting in the near future to examine the potential of the technique on such complex blends.

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