SUMMARY
Pulmonary aerosol delivery (inhalation) presents an elegant and effective approach to getting therapeutically active agents into the body, when the delivery method is properly designed to meet the requirements of the administration. Different administrations have different requirements, and therefore, there is not one optimal strategy in pulmonary aerosol delivery. Many variables are involved, which restrict the possibilities in a specific situation. In this thesis, a strategy is proposed for choosing the most suitable approach to optimisation of inhaled drugs and diagnostics. In addition, this thesis describes a number of studies conducted at the interface of the pharmaceutical technological laboratory and clinical practice, in which the importance is stressed of choosing the right delivery device for a specific situation.

Optimisation of pulmonary aerosol delivery requires recognition of three determinants: the purpose of the administration, the patient for whom the administration is intended, and the product – consisting of the device that is used for generation and administration of the aerosol and the formulation of the active agent. In Chapter 1, an introduction is given on the main purposes of pulmonary aerosol delivery, the different patient populations and their population specific characteristics and limitations that affect the freedom of choice, and the various products available for delivery of an active agent to or through the respiratory tract. Subsequently, the interplay between these three determinants is described as starting point for choosing the optimal approach.

In addition to the three determinants, this starting point also depends on whether a new inhaled application is developed or whether an existing one is prescribed. In the latter case, only a limited array of therapeutic options is usually available, and from these the most suitable one should be chosen for a specific individual patient and his specific needs, wishes, and abilities. The interaction between patient and device is the most important interaction to consider when prescribing an inhalation product, since the device has to be prepared and used correctly to achieve sufficient pulmonary deposition required for the desired therapeutic effect. In other words, an inhalation product is only as good as the patient’s ability to use it, or his motivation to use and maintain it correctly.

When developing a new inhaled application, the optimal product design should be based primarily on the question what device would best suit the capacities and needs of the anticipated patient population – in the context of the purpose – rather than which formulation is easiest to realise. However, deciding which device best suits the
needs of the patient is more easily said than done. After all, there is no such thing as *the* patient; there is rather a very diverse collection of individuals with different needs and abilities. Still, defining an average patient or patient population for the intended purpose may serve as a good base for choosing the most appropriate type of device. In addition to the patient, other factors can restrict the choice for a device. Most important are the active substance and its intended dose, thus the formulation, as well as the dose regimen. Not all substances can be formulated into all possible formulations that are considered suitable for inhalation. Biopharmaceuticals, such as proteins, are delicate compounds, which can easily be degraded by various stresses during manufacturing processes (*e.g.* drying for DPI formulations), storage (*e.g.* the hostile environment of pMDI propellants), or even administration (*e.g.* shear stresses during nebulisation). However, it is not just complex molecules that may present restrictions. The study described in Chapter 2 illustrates how even a straightforward small-molecule formulation can be incompatible with the chosen delivery system. The study presents a good example of a simple and fast, but inadequate solution to a problem that is more complex than anticipated. A different delivery device may require a more time-consuming development process, but can eventually improve the application significantly (Chapters 3–5).

The studies described in Chapters 2 to 5 focus on bronchial challenge testing, a diagnostic application of pulmonary aerosol delivery. Bronchial challenge tests are performed to measure bronchial hyperresponsiveness, a key characteristic of asthma. In clinical practice, it is generally not possible to develop a delivery device specifically for an administrative purpose. Rather solutions have to be found to facilitate the use of existing devices for specific, often off-label applications. Chapter 2 describes the *in vitro* evaluation of such a solution encountered in bronchial challenge testing: the use of the Sidestream nebuliser in bronchial challenge testing with AMP. AMP is usually administered according to dosing protocols developed for another challenging agent, methacholine. In the laboratory, the effects of AMP concentration, jet pressure, and suction flow rate on nebuliser output were determined. The dosing protocols require operation of the Sidestream nebuliser at a different jet pressure than specified, and it was shown that this strongly increases the droplet size in the aerosol. Furthermore, the high AMP concentrations used in bronchial challenge testing were found to strongly affect both droplet size and nebuliser output rate. These effects on nebuliser performance imply that the administered dose and site of deposition of AMP change at dose escalation. Since all investigated parameters influence nebuliser performance, it is recommended that they should not only be specified in challenge testing, but be actively controlled as well. Moreover, the study shows that nebulisers may not be the most appropriate devices for administration of AMP in a bronchial challenge test.
Chapters 3 to 5 were conducted in collaboration with the department of Pulmonary Medicine and Tuberculosis from the University Medical Centre Groningen. One of their research lines involves the contribution of small-airways dysfunction to the clinical expression of asthma. It has been shown that the presence of small-airways dysfunction is associated with worse asthma symptoms, including poorer control of asthma and a higher number of exacerbations. However, no simple and direct methods are available to diagnose small-airways dysfunction. Our collaboration investigated the use of a small-particle bronchial challenge test as a fast and reliable tool for assessment of small-airway dysfunction.

Dry powder inhalation of adenosine, the active moiety of AMP, by use of an effective inhaler may provide an interesting alternative to nebulisation. Chapters 3 and 4 describe the development, in vitro performance, and first clinical testing of a dry powder adenosine inhalation product. The study reported in Chapter 3 involved the testing of various powder formulations and classifier-based dispersion principles and investigation of the in vitro performance of the most promising formulation/classifier combination in a new test inhaler system. Spray-dried formulations of either pure adenosine (100%) or adenosine and lactose as diluent (1% and 10% adenosine) were prepared to cover the entire expected dose range for adenosine (0.01–20 mg). All three powders, in all twelve suggested doses, dispersed well with the newly developed test inhaler with a multiple air jet classifier disperser, into aerosols with an average volume median diameter of 3.1 µm (3.0–3.3 µm). For eleven out of twelve dose steps, the fine particle fractions < 5 µm as percent of the loaded dose varied within the range of 67–80% (mean: 74%). The new test concept allows for more consistent aerosol delivery over the entire dose range with aerosols having narrower size distributions than those obtained with nebulisation, which may improve adenosine administration in bronchial challenge testing. In Chapter 4, the applicability of the dry powder adenosine inhalation product is described in comparison to nebulised AMP. Two higher dose steps of 40 and 80 mg were added to the escalating dose range. Five asthmatic subjects performed two bronchial challenge tests, one with nebulised AMP following the 2-minute tidal breathing method; the second with dry powder adenosine administered with the investigational inhaler and single slow inhalations (inspiratory flow rate 30–40 L/min). All subjects reached a 20% fall in FEV$_1$ with the new adenosine test (PD$_{20}$) compared to four subjects with the AMP test (PC$_{20}$). Dry powder adenosine was well tolerated by all subjects and better appreciated than nebulised AMP. In conclusion, this new bronchial challenge test appears to be a safe

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and convenient alternative to the nebulised AMP test to assess bronchial hyperresponsiveness in asthmatic subjects.

Chapter 5 describes a clinical study that investigated whether dry powder adenosine can be used as diagnostic tool to identify asthma patients with small-airway dysfunction. It was hypothesised that these patients could be identified by selectively challenging the large and small airways, by using challenge tests consisting of large- and small-particle adenosine, inhaled with either a high or low flow rate. The study confirmed that dry powder adenosine challenge is able to induce bronchial hyperresponsiveness in asthmatic subjects. However, it was found that, regardless of inspiratory flow rate, both large- and small-particle adenosine induced responses in the small airways in 41 out of 42 tests. Three possible explanations are given for the unexpected results. Firstly, it may be that the expected selective deposition of large particles in the large airways and small particles in the small airways was not obtained. Secondly, it may be that the lung function tests that were used in the study as read-out of large and small airway constriction have limited specificity in discriminating between responses in the large and small airways. A final explanation may be that deposition of adenosine in the large airways not only leads to obstruction in the large airways but also in the small airways. The small airways may also respond to inflammatory mediators transported distally via superficial capillary vessels, or to stimulation of sensory nerves with excitation of cholinergic reflex pathways. A neural mechanism has not extensively been investigated in human bronchial hyperresponsiveness yet, but this may be worthwhile in perspective of these findings. This last explanation could have profound implications for the understanding of the underlying mechanisms of bronchial hyperresponsiveness and possibly the treatment of asthma and COPD.

Bronchial challenge testing is one of the few non-therapeutic purposes of pulmonary aerosol delivery. A bronchial challenge results in bronchoconstriction, which of course needs to be countered once the test is over, for which reliever medication is administered. In Chapter 6, the effects of a disposable extension mouthpiece on the aerosol from the breath-actuated salbutamol Redihaler are characterised. This mouthpiece has been developed to allow the use of this multi-dose product by multiple patients as reliever medication after a bronchial challenge test is performed. The extension mouthpiece was found to greatly reduce the delivered dose from the Redihaler and to selectively retain the larger particles from the aerosol. It was furthermore found that an inspiratory flow rate of 30 L/min was insufficient to open the valve in the extension mouthpiece completely. When using a standard pressurised metered dose inhaler with a valved holding chamber comparable losses can be found. Therefore, in spite of the high losses, the Redihaler with disposable mouthpiece can
be considered a suitable alternative to a standard pMDI with VHC, provided that the same dose is given (four times 100 μg) and that the patient is instructed to inhale moderately forceful to allow proper opening of the valve.

Given the importance of a successful interaction between patient and device, knowing the patient population and measuring their abilities and capacities is paramount to developing a new pulmonary application. In Chapter 7, this interaction was studied for the important and highly challenging patient population of primary school children and DPIs. Age appropriateness is a major concern of pulmonary delivery devices in general, but especially for DPIs, since their performance strongly depends on the inspiratory flow manoeuvre of the patient. The study investigated the requirements for a paediatric DPI by use of an instrumented test inhaler with variable airflow resistances and mouthpiece designs. The impact of airflow resistance on the inspiratory flow profiles of the children as well as the children's preferences for airflow resistance and mouthpiece design were investigated. 98 children aged 4.7 to 12.6 years participated in the study, of whom 91 were able to perform at least one correct inhalation through the test inhaler. Both airflow resistance and the children's characteristics (age, height, and gender) were found to affect the inspiratory flow parameters, and it was concluded that a medium-high resistance is both suitable for and well appreciated by children aged 5 to 12 years. Moreover, high incidences of obstructions in the oral cavity were observed, which may be reduced by optimisation of the mouthpiece design. Based on the results, design recommendations are given for future paediatric inhaler development.

Lastly, Chapter 8 reviews an application of pulmonary aerosol delivery that is still in a more experimental stage of development: pulmonary vaccination. It is described how pulmonary vaccination can be a promising alternative to vaccination by injection, since it takes away one of the main concerns of this administration route, i.e. the use of needles. An additional advantage of vaccination through the lungs can be the induction of a mucosal immune response on top of the systemic immune response, which could increase the effectiveness of vaccination against airborne pathogens. The review focuses on the inhalation devices and formulation strategies that may be suitable for the pulmonary administration of vaccines, by taking into account the most critical and limiting parameters: the labile compounds that vaccines generally are and the target population for whom the vaccination is intended. It is proposed that the development of the inhalation device and vaccine formulation should be done concurrently, to obtain a delivery system that can effectively be used by the target population and can be produced at low costs; two prerequisites for successful application of pulmonary vaccination in large-scale vaccination programs.