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**FORMULATION OF PARTICLES FOR PULMONARY DRUG
DELIVERY**

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SCHOOL OF CHEMICAL AND BIOMEDICAL ENGINEERING
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FORMULATION OF PARTICLES FOR PULMONARY DRUG DELIVERY

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Summary

Pulmonary drug delivery has become an attractive route of drug administration to the human body for the treatment of lung diseases and a number of systemic applications. Dry powder inhalation (DPI) is considered as the most appropriate aerosol therapeutics in terms of delivery efficiency, patient compliance and environmental issue. Successful delivery of the dry powder formulation to the deep lung requires proper understanding of the factor influencing particle deposition. Improvement of the drug formulation characteristics is the most convenient way to control delivery performance. In this study, the effect of shape and morphology of dry particles for inhalation is investigated. The optimum design of micro-particle for better delivery performance is determined. Preliminarily, different shape macro-particles with low sphericity are experimented in a confined medium to determine the impact of shape irregularities on wall effect during gravitational settling. Then different shape micro-particles including spherical, pollen, plate, cube, and needle shape particles are synthesized with a similar aerodynamic size range of $<5\mu\text{m}$. Flowability and *in vitro* dispersion and deposition experimentation using cascade impactor are performed with the different shape particles. Particles with shape that have preferable behavior are formulated as carrier particles for dry formulation. The flow behavior and turbulence occurrence of the particle laden gas flow are measured in an idealized inhalation path model. The feasibility of these carrier particles as a binary mixture with a model drug is also assessed. The effect of drug loading and

inhalation flow rate of these carriers are assayed and compared with conventional carrier particles.

Particle shape is found to influence the particle settling behavior and velocity by changing their settling orientation. Elongated particles can avoid adverse wall effect by shifting their settling orientation in the settling medium. Micro-particle flow, dispersion and deposition characteristics are not direct function of particle aerodynamic diameter (d_a) and characteristic size. Particle shape factor is also not able to account the inconsistencies of the irregular shape particles. Pollen-shape particles with fibrous surface morphology exhibit better flow properties, *in vitro* emission and deposition values than other shape particles. Among the particles with a d_a range of 1.4-5.9 μm , pollen-shape particles result in ED values over 80% while other shape particles exhibit 50-75% at 30 L/min. The FPF value of the pollen-shape is over 15% while for other shape particles it varies in the range 2-10%. It is anticipated that surface morphology impose low surface density to the particles. They experience lower interparticle interaction and aggregation tendency which improve their flow, dispersion and deposition performance. Large pollen-shape particles show preferable flow behavior in the idealized inhalation path model. They can follow the geometry conveniently and reduce early deposition unlike the traditional lactose carriers. The binary mixture of these pollen-shape carrier particles with a model drug also proved their potential in comparison with the lactose carriers. Pollen-shape particles can exhibit high drug loading due to their surface morphology. The drug concentration in the blends with pollen-shape hydroxyapatite (HA) model carriers after sieving is more than double compare to the drug concentration in the

conventional lactose (LA) blends (LA=3.73%, HA=6.51-7.90%). Blends with HA carrier particles are also found to have better drug emission and deposition in the lower stages of the impactor. The HA blends show high ED of 82–90% at 30 L/min while the LA blends are observed to have ED of 69–82% at the same conditions. The high emission of the HA blends also allows high fine particle fraction (FPF) of 10–18% while the FPF of the LA blends are 3–15%. At a gas flow rate of 60 L/min, all the HA blends show better ED than LA blends (83–95% for HA and 82–84% for LA) but compatible FPF results are found (19–41% for HA blends and 21–34% for LA blends). It is also found that the blends with pollen-shape particles with different size and morphology can exhibit good ED, though FPF results are observed with no regular trends.

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Nomenclature

$\dot{A}$	empirical coefficient used in Equation (4.12)
A	Hamaker constant
a	length of cube and flat plate particles
b	width of cube and flat plate particles
c	height of cube and flat plate particles
D	column diameter or diameter of flow channel
d	cylindrical particle diameter
d_e	particle equivalent diameter
d_a	aerodynamic diameter
d_g	geometric diameter
d_p	mean particle size
F	van der Waals force between two spherical particles
g	gravity
H	separation distance
h'	height of bicone shape particle
l	cylindrical particle length
p	exponent used by Kehlenbeck and Di Felice ¹
R	radius of the particles in interaction
r	radius of the contact area of the two interacting particles
r'	radius of bicone shape particle
$Re_{t\infty}$	Reynolds number in infinite dimension vessel at steady-state condition
S_{tk}	Stokes number
U	mean velocity of the flow, m/s
u_t	particle velocity in finite diameter column at steady-state condition
$u_{t\infty}$	particle velocity in infinite diameter column at steady-state condition
α	exponent by Di Felice ²
θ	angle of slide
S	dynamic shape factor

λ	particle equivalent diameter to column diameter ratio
λ_c	critical particle equivalent diameter to column diameter ratio
λ_o	empirical parameter used by Kehlenbeck and Di Felice ¹
μ	fluid viscosity
ρ	fluid density
ρ_{bulk}	bulk density
ρ_e	effective particle density
ρ_s	unit density
ρ_{tap}	tap density
ψ	particle sphericity
η	deposition efficiency

Publications arising from the thesis

Journals

1. Wang, Y., **Hassan, M. S.**, Lau, R., Gunawan, P., and Xu, R. Polyelectrolyte mediated formation of hydroxyapatite microspheres of controlled size and hierarchical structure. Journal of Colloid and Interface Science, 2009, 339(1): 69-77.
2. **Hassan, M. S.** and Lau, R. Flow behavior and deposition study of pollen shape carrier particles in an idealized inhalation path model. Particuology, 2010, 8:51-59.
3. **Hassan, M. S.** and Lau, R. Feasibility study of pollen-shape drug carriers in dry powder inhalation. Journal of Pharmaceutical Sciences, 2009, 99(3): 1309-1321.
4. **Hassan, M. S.** and Lau, R. Effect of particle shape on dry particle inhalation: study of flowability, aerosolization, and deposition properties. AAPS Pharmscitech, 2009, 10(4): 1252-1262.
5. **Hassan, M. S.** and Lau, R. Inhalation performance of pollen-shape carrier in dry powder formulation with different drug mixing ratio: Comparison with lactose carrier. International Journal of Pharmaceutics, 2010, 386(1-2):6-14.
6. **Hassan, M. S.** and Lau, R. Drug Formulations for Improved Dry Powder Inhalation Efficiency. Current Pharmaceutical Design, 2010, in press.
7. Lau, R., **Hassan, M. S.**, Wong, W. Y. and Chan, T. Revisit the wall effect on the settling of cylindrical particles in the inertial regime. Industrial & Engineering Chemistry Research, 2010, accepted.
8. **Hassan, M. S.** and Lau, R. Inhalation performance of pollen-shape carrier in dry powder formulation with different drug mixing ratio: Comparison with different pollen shape carriers. In preparation.

Conferences

1. **Hassan, M. S.** and Lau, R. Wall effect study of different shape settling particles in cylindrical columns, Regional Symposium on Chemical Engineering (RSCE), Dec. 2007, Yogyakarta, Indonesia.
2. **Hassan, M. S.**, Wang, Y., Xu, R., and Lau, R. Evaluation of the flow properties of spiked-sphere shape hydroxyapatite particles for dry powder inhalation, American Institute of Chemical Engineers (AIChE) Annual Meeting, Nov 2008, Pennsylvania, USA.
3. **Hassan, M. S.** and Lau, R. Pollen shape particles for pulmonary drug delivery: *in vitro* study of dispersion and deposition properties, International Conference on Biomedical Engineering (ICBME), Dec 2008, Singapore.
4. **Hassan, M. S.** and Lau, R. Flow behavior study of pollen shape model carrier particles in an idealized inhalation path model by particle image velocimetry. International Conference on Chemical Engineering (ICChe), Dec 2008, Dhaka, Bangladesh.
5. **Hassan, M. S.** and Lau, R. The influence of carrier morphology on drug loading and *in vitro* deposition in dry powder inhalation. The International Symposium on Advanced Bio, Nano, and Pharmaceutical Science and Technology (BNPST), May 2009, Beijing, China.
6. **Hassan, M. S.**, and Lau, R. Effect of carrier morphology on drug loading and *in vitro* aerosolization and deposition properties in dry powder inhalation, American Institute of Chemical Engineers (AIChE) Annual Meeting, Nov 2009, Nashville, TN, USA.

Chapter 1 INTRODUCTION

1.1 Background

1.1.1 Significance of pulmonary drug delivery

In the treatment of diseases, aerosol administration represents a valuable mean by which a therapeutic agent can be delivered through the pulmonary route. In this technique inhalable aerosol medication is aerosolized into the human airways to be delivered in the alveolar region. The delivered medication is absorbed by the epithelial cell into the systemic circulation. Recent technological advances in different aspects of innovative therapeutic dose and delivery device techniques make this route of drug administration become attractive.

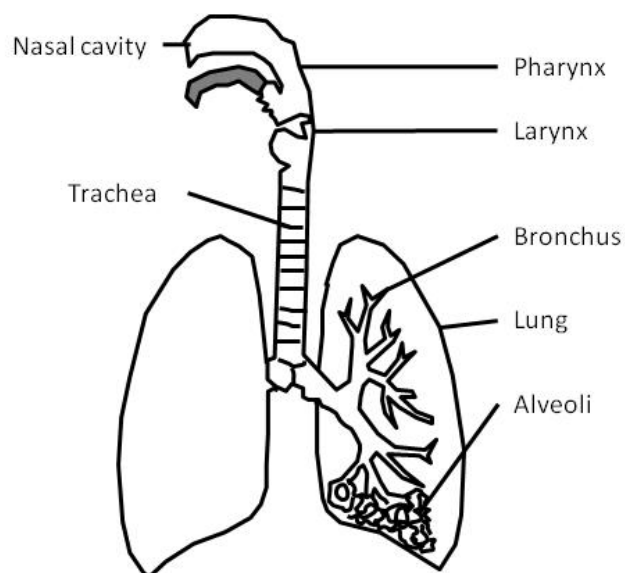


Figure 1.1 Schematic diagram of a human lung.

Human lung airways consist of several parts like pharynx, larynx, trachea, bronchioles and alveolar region. In Figure 1.1 a schematic of the human lung can be seen. The main issue in pulmonary drug delivery is to deliver the medication to the lower

airways of human lung. Precise drug deposition can be performed by specific targeting of disease or receptor locations, thereby increasing the efficiency and effectiveness of aerosolized drug delivery³.

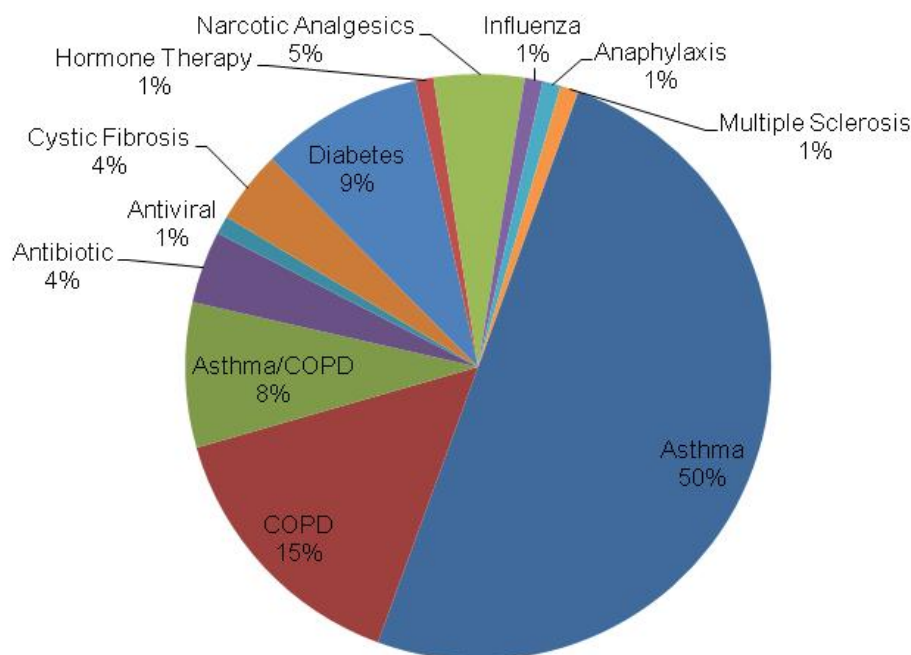


Figure 1.2 Different respiratory and systemic diseases treated by pulmonary drug delivery.⁴

Pulmonary route of drug administration have been used mainly to treat lung diseases. Different types of lung and systemic diseases that can be treated by this delivery technique are shown in Figure 1.2. Most lung diseases such as chronic persistent asthma, bronchiectasis, extrinsic allergic alveolitis, emphysema, sarcoidosis etc affect the lower airway regions. Formulation also needs to be in the lower region to deliver the drugs to the systemic circulation. Therefore, the lower airway of the lung is the target area for

pulmonary drug delivery. Different aerosol medications have become popular for the treatment of lung diseases. For instant, bronchodilator and anti inflammatory corticosteroids aerosol medications are used for asthma treatment and inhalable antibiotics are used for the treatment of cystic fibrosis.⁵ On the other hand aerosol medication for the treatment of systemic diseases is a recent development in pulmonary drug delivery e.g. insulin for diabetics.

1.1.2 Different inhalation delivery techniques

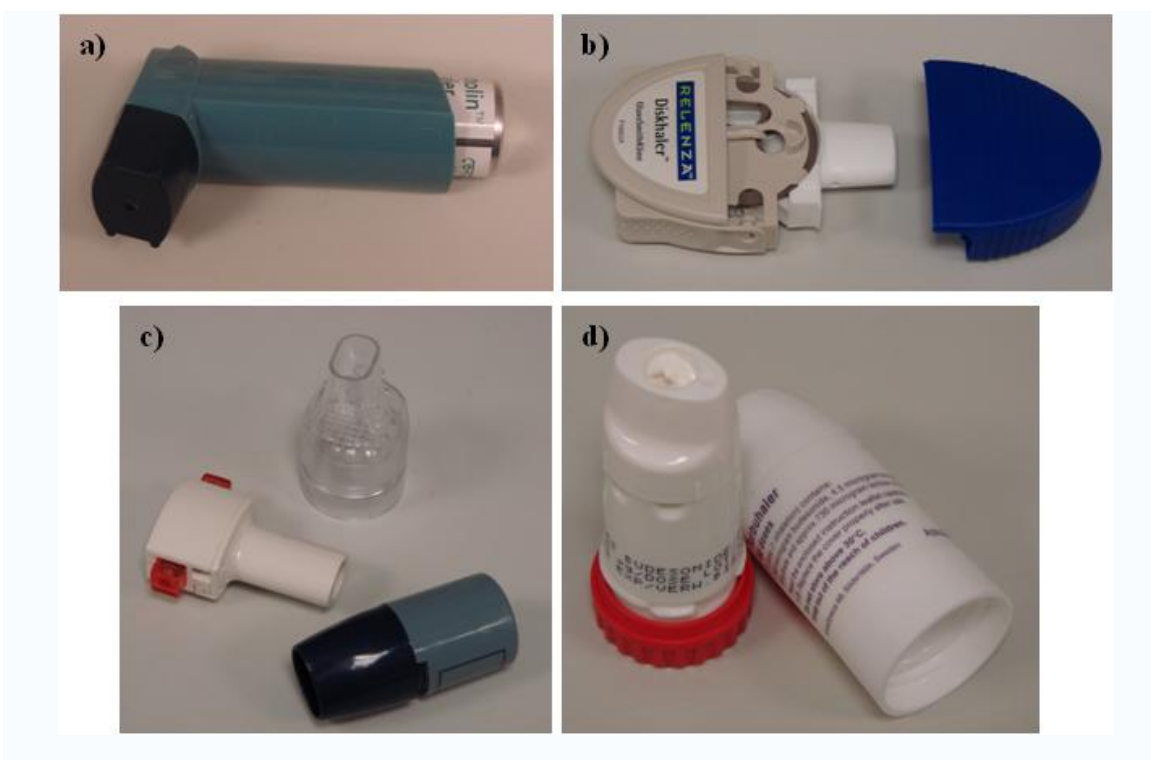


Figure 1.3 Different types of commercially available DPI and MDI. a) Ventolin® - pMDI, b) Diskhaler® - multi unit dose DPI, c) different types of rotahaler-single dose DPI device, and d) Turbuhaler® – multidose DPI device

Pulmonary route for drug administration was first used long before to treat asthma, though it took long time to get proper attention as a potentially effective way of drug administration. To date, three different kinds of inhalers are commonly used, i.e.

nebulizer, metered dose inhalers (MDI) and dry powder inhaler (DPI). Some common samples of the different types of inhaler are shown in Figure 1.3. Nebulizer was first used in 1925. Asthma disease was treated with nebulizer before the invention of the MDI. However, that device was delicate and the outcome was not reliable. In 1955, Riker Laboratories developed the first MDI. At that time the medications were driven by a CFC propellant. The suspended solution of the drug is kept in a pressurized canister. At the time of actuation, the pressure is released slightly and the drug particles are atomized into the human lung system. Then the drug particles travel to the lower lung airways with the air inhalation. The first MDI product was marketed in 1956.

DPI was invented as a good alternative of MDI in 1960. In DPI, pharmaceutical active ingredients are delivered as dry powder. The drug formulation is kept in a capsule for manual loading or directly stored inside the inhaler. After actuation, the patients will take the dose by deep inhalation and hold their breath for 5-10 seconds. Three different categories of DPI devices have been developed, e.g. single dose devices, multiple unit dose devices or multi dose devices. Single dose devices are loaded with capsules or blisters. In actuation, the doses are released from the capsule or blister and taken inside the lung through inhalation, e.g. Spinhaler[®] and Aerolizer[®] (Novartis). Multiple unit dose devices carry several capsules or blisters, e.g. Diskhaler[®] and Diskus[®] (GlaxoSmithKline). In multi dose inhalers the medications are kept in a reservoir in the inhaler and metered from there as free flowing powder, e.g. Turbuhaler[®] (Astra Zeneca).

Improved devices and formulations have changed, inhalation pharmaceutical technology over the last two decades.⁵ In 1986, researchers found that large molecules can be absorbed conveniently into the systemic circulation of rats through pulmonary

route.⁶ Other than pulmonary delivery or injection, no other routes are capable of delivering macromolecules unless (including oral, buccal, transdermal and nasal) penetration enhancers are used. Therefore, many pharmaceutical companies and academics worldwide concentrate on the research to develop novel formulation and ideal devices to deliver protein and peptide drugs.

1.1.3 Advantages of pulmonary drug delivery

Pulmonary route of drug administration offers several advantages over the other routes of drug administration. The advantages are described below.

1. Respiratory tree in the adult human has an estimated surface area of 140 m². 95% of the absorptive area, the alveolar surface is covered with a thin vesculated and perfused monolayer of epithelial cells.⁷⁻⁹ In Figure 1.4, epithelial cells of different regions of human lung are shown with relative sizes.⁶⁻⁸ Pulmonary route of drug delivery is effective because the epithelium for human lung is highly permeable and easily accessible by an inhaled aerosol dose.

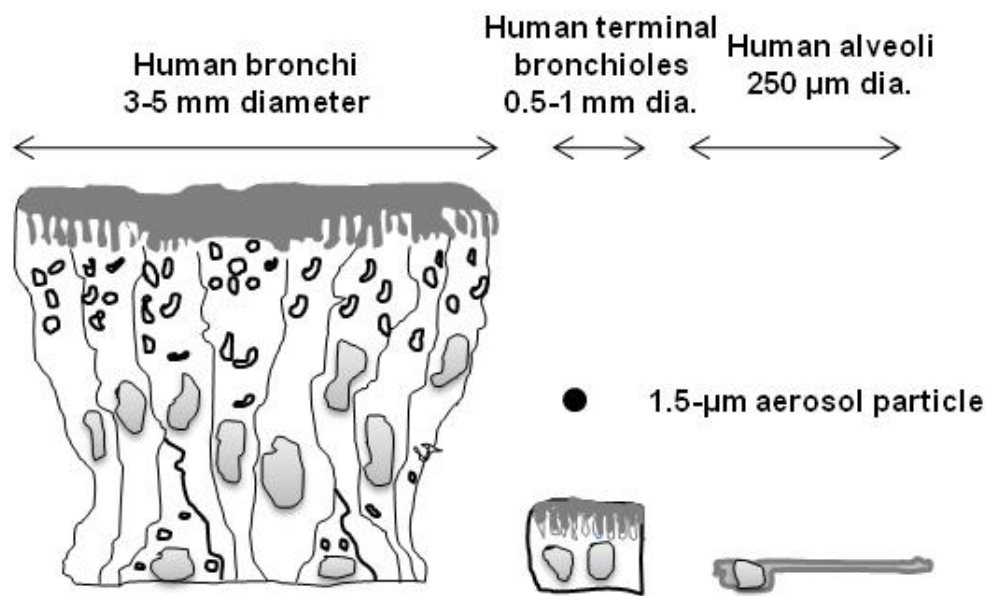


Figure 1.4 Epithelium at different sites in the lung. This Figure is drawn as per the reference.⁶

2. Pulmonary route is a noninvasive ‘needle-free’ delivery method.

3. Through pulmonary drug delivery, therapeutic efficacy can be improved by delivering the drugs directly to the site of action of the respiratory tract. By this site directed drug delivery, the local concentration of the therapeutics can be raised to the required level while the systemic concentration is kept low.¹⁰ Inhalation aerosols for the treatment of respiratory disease are an excellent example of site-directed delivery by physical means.

Though only a small portion of the inhaled drug particles can reach to the deep lung (typically less than 20%),¹⁰ the dose amount would still be smaller than that to be given systemically for the same therapeutic effect. The site-directed route also has the advantage of activation of a prodrug at the site of action.

4. Some respiratory drugs show serious systemic side effects when introduced by other means of drug administration. For these drugs, site directed delivery is essential.

Pulmonary drug delivery minimizes risk of systemic side-effects.^{10,11} Moreover, because of the slow systemic clearance of the pulmonary route, the duration of the drug action is longer than that through the systemic route.

5. This site directed drug delivery technique for lung diseases also offers rapid response in the therapeutic outcome.¹¹ The drugs do not need to pass through the barriers like other routes of administration, e.g. gastrointestinal absorption and first-pass metabolism in the liver. Therefore, lower amount of drug is needed for inhaled dose than the systemic dose to generate similar level of therapeutic effect. For example, 100–200 µg of inhalable salbutamol is therapeutically equivalent to 2–4 mg of its oral medication.¹¹

For the treatment of some diseases, a rapid absorption of the drug into the systemic circulation may be preferable.¹⁰ Pulmonary drug delivery offers great potential for a fast delivery of small-molecules to the systemic circulation. Absorption of the small molecules deposited in the lungs into the systemic circulation is rapid. The absorption occurs with the help of the large surface area of the lungs, high permeability of the epithelial and small aqueous volume at the absorptive surface.⁷ This is the fastest uptake of drug of than other routes of delivery except intravenous. For some human diseases the onset of symptoms happens very rapidly, generally in seconds, e.g. the myriad forms of pain, anxiety, spasms hypertensive crisis, arrhythmias, nausea etc. The diseases could be offset by fast-acting inhaled medicines.⁶

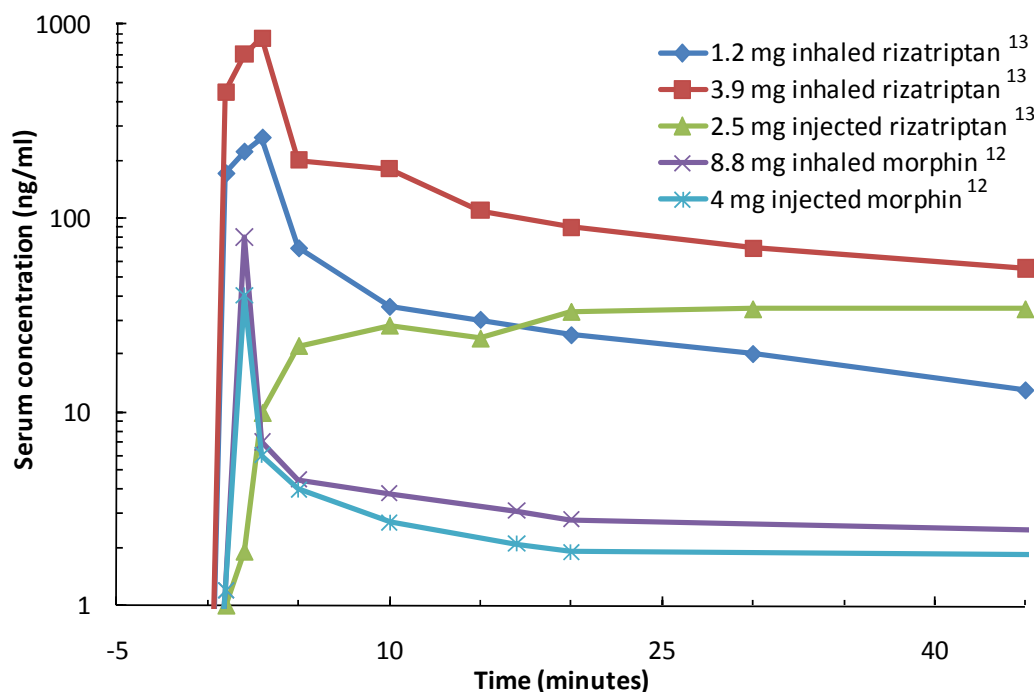


Figure 1.5 The illustration of fast pulmonary absorption.

Figure 1.5 shows the trend in plasma concentration with time following inhalation and injection. The behavior of inhaled morphine (dose=8.8 mg) compared with injected morphine (dose=4 mg) in human volunteers¹² and plasma profile of rizatriptan in dogs with inhaled and injected doses¹³ are shown. Rapid absorption of the drugs is found by the inhalation route.

6. Pulmonary drug delivery is suitable not only for the small molecules but also to very large proteins.^{11,14,15} The slow mucociliary clearance in the lung periphery increase the residence time of the large molecules in the lung.¹¹ Therefore, large molecules can be absorbed in significant quantities in spite of the low absorption qualities.

Though the pulmonary route of drug delivery was used solely for the treatment of lung disease initially, it has great potentials for delivering drugs for other diseases. Protein and peptide drugs are easily decomposed by oral ingestion and toxic drugs could

have severe side effects if not been delivered to proper sites. Studies have shown that the pulmonary administration of drugs has the potential to improve protein, toxic and easily decomposing drugs delivery. This delivery technique is safer for the patients in clinical respect and more economic for the drug industry in the financial respect.

7. The concentration of the drug-metabolizing enzymes is lower in the lungs than in the gastrointestinal tract and liver.¹⁶ This low enzymatic environment of the respiratory route can avoid the hepatic first-pass metabolism. Thus, the possibility of degradation of the inhaled molecules that enter the circulation through pulmonary route is lower than their delivery through oral administration. Moreover, pulmonary delivery is free from the of dietary complications, extracellular enzymes (to break large molecules) and the difference in inter-patient metabolism that affect gastrointestinal absorption^{11,15} There are some drugs that have adverse post-prandial effects on bioavailability or low gastrointestinal bioavailability. Pulmonary drug delivery would be a reliable and efficient route for them.⁶

1.1.4 Benefits of dry powder inhalation therapeutics

Patients and healthcare professionals mentioned their opinions about different aspects of delivery techniques. On the basis of that, some characteristics are set up for an efficient inhaler. The characteristics include ease of usage of the inhaler, versatility of delivering different type of drugs effectively to the lung, presence of dose counter, ease of transport and storage, free of propellant, construction by non-toxic materials etc.¹⁷

It is reported that patients' dissatisfaction with MDI is pushing forward to a new alternative.¹⁸ DPIs have several advantages over MDIs. DPIs are breath-activated, convenient in use, can exhibit better effectiveness and environment friendly.¹⁷ Previously, the suspended medication solution was driven by pressurized chlorofluorocarbon (CFC) propellants in MDI.¹⁹ Stratospheric ozone depletion is a vital environmental issue in the world and part of ozone degradation is caused by the CFC products. To protect the ozone depletion the use of CFC need to be minimized similar like other responsible chemical compounds for ozone loss. Under this consequence, new inhaler technology is developed by introducing hydrofluoroalkanes (HFAs) as propellant. However, this alternative propellant substance also has potential risk of environmental problems. Therefore, in the environmental aspect, it would be better to replace the MDI with DPI.

MDIs also have aerosol formulation and generation related problems. The pressurized solution in the MDIs may contain different kinds of surfactants which cause complexities in the airways. For example, oleic acid could be a possible risk for bronchospasm in patients suffering from advanced airway hyperreactivity.²⁰ Moreover, the medication is delivered from the inhaler as droplets and a huge portion of them

deposit in the initial airways because of their large size and high velocity.²¹ Therefore, lung deposition of the drug formulation from DPI (Turbuhaler[®]) was found to be twice of that from a MDI (32% versus 15% respectively).²² It can be demonstrated that large amount of medication would be lost in the initial airways from MDI. This would increase the amount and cost of the MDI medication to produce same level of disease control. It would also increase the systemic and local side effects.

It is also reported that asthma treatment by DPI is more effective than MDI.²³ For example, in the treatment of perennial asthma in children, DPI Turbuhaler[®] can generate higher clinical improvement by delivering similar dose of drug compared to the improvement produced by MDI Nebuhaler[®]. In most of the MDI, a precise coordination of the actuation and inhalation is needed for effective delivery. Sometimes this could go against patient compliance.²¹ On the other hand, DPI devices are breath-actuated which help the patient to avoid coordination problem in using the inhalers. Thus, elderly patients can use the DPIs correctly and prefer to use them most.²⁴

1.1.5 Motivation of this research

In 2004, 292 million MDI and 113 million DPI were sold worldwide.²⁵ Huge amount of inhalation doses are taken in every year. The quality control of the inhalers is essential. It is reported that, only 0.1% of the imperfect inhaler devices could make more than 400,000 cases of patient noncompliance per year.²⁵ Inefficient delivery, therefore, could induce a huge loss of therapeutics and cause side effects. Till now, only a small fraction of the dose of present formulations can be delivered to the lower airways. So, the main challenge in pulmonary drug delivery involves the improvement of the deep lung deposition. Dry powder inhalation is influenced by different factors. However,

controlling the characteristics of the drug formulations would be the most suitable way to improve their delivery efficiency. It is well known that aerodynamic behavior of dry powder formulation depends mostly on the physical properties of drug and carrier particles. Therefore, the insights of the effect of the physical properties of the formulation components e.g. drug and carrier on the delivery performance is experimentally investigated. The knowledge would be helpful in developing new formulation for efficient drug delivery, not only for the treatment of lung diseases but also for large range of local and systemic applications.

1.3 Thesis outline

The experimental investigations carried out in this study are demonstrated in eleven chapters.

Chapter 1 is an introduction of this study which includes the background, development, motivation, specific objective and aim of the study.

Chapter 2 enlightens the literature review on different aspects of pulmonary drug delivery. Prominent researches regarding particle deposition mechanism and different influencing factors of deposition are reviewed which includes lung physiological and drug formulation aspects.

Chapter 3 includes detail description of different experimentation methods used in this study. The sample preparation and their characterization methods are also discussed in details.

Chapter 4 presents the preliminary study comprising particle shape effect on gravitational settling in a macro-particle system. Millimetre sized particles are employed

in settling experiments in a confined medium and the effect of irregular shape particle on settling corresponding to different medium diameter is investigated.

Flowability, aerosolization and deposition behavior are the most essential for effective delivery of particles deep into the lung. In chapter 5, different shape particles with similar size range are utilized and their characterizing behaviors are measured to determine the shape effect on them. The optimum particle shape with favourable flowability, aerosolization and deposition properties would be identified.

Pollen-shape particles are found to have good flowability and aerosolization behavior. Hence, the feasibility of its application as drug carriers in dry powder formulation is studied. Flow behavior, velocity field and turbulence occurrence of large pollen-shape particle laden gas flow at distinct positions in an idealized inhalation path model using particle image velocimetry (PIV) is presented in Chapter 6. .

Pollen-shape particles with large size are implemented as model carrier particles in dry powder formulation along with a model drug. The study of the performance of pollen-shape carrier particles and their comparison with traditionally used rock- shape lactose carrier particles is presented in chapter 7.

Chapter 8 describes the effect of drug attachment on the inhalation performance of these pollen-shape carrier particles at different inhalation flow rate. Their performance is also compared with lactose carrier's performance under the same condition. The second part of this study is in chapter 9 where the impact of the size and morphology change of the pollen-shape carrier particles on delivery performance is assessed with different drug loading and flow rate.

Finally, overall concluding remarks of the study and the recommendation for future works are presented in chapter 10.

Chapter 2 LITERATURE REVIEW

2.1 Introduction

Drug formulations must overcome various physical obstacles to be delivered effectively for either local or systemic action.²⁶⁻²⁹ This inhalation medication therapy is successful for lung diseases, even though typically only less than 20% of the inhaled dose reaches the bronchial airways and alveoli of the respiratory tract. Inhalation drug delivery study requires knowledge of this administration route including the mechanism and factors of aerosol particle deposition in the lung airways.

2.2 Deposition mechanism

Deposition mechanisms in the airways can be categorized into five types, namely inertial impaction, sedimentation, diffusion, electrostatic precipitation and interception. Inertial impaction is caused by the tendency of particles and droplets to move in the initial direction instead of following the gas streamlines. Hence, impaction may occur on the obstacle downstream in the trajectory of the particle in the lung. Particles with high mobility can avoid inertial impaction in upper airway region, thus their probability of deep lung deposition is high. Sedimentation takes place due to gravity. Particle deposition from inertial impaction and sedimentation are directly related to the residence time of the aerosol particle into the lung. Diffusion is the dominant mechanism for small particle deposition. It depends on the diffusion coefficient that increases with the decrease of the particle size and is independent of the particle density. Hence, diffusion coefficient is more suitable to describe the deposition of ultra fine particles than other parameters. These three are the main deposition mechanism for particles. Electrostatic precipitation

plays a vital role for deposition of charged particles. Finally, interception mechanism is important for deposition in the respiratory tract when the dimensions of the anatomical spaces are comparable to the dimension of the particle.¹⁰

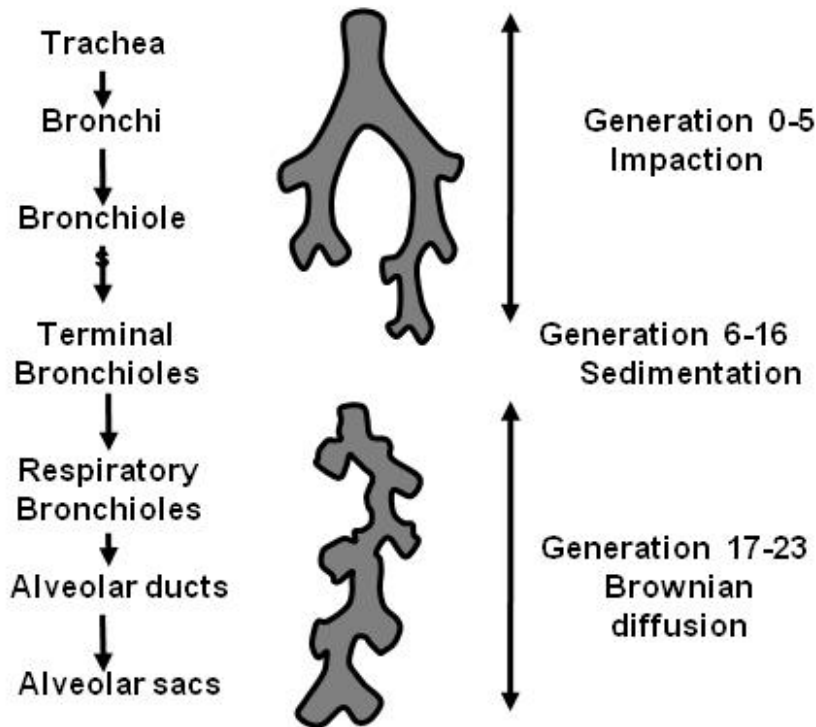


Figure 2.1 Deposition mechanism at different regions of human lung.

Figure 2.1 shows the dominating deposition mechanisms at different regions of the lung. Particles with higher velocity and turbulence can easily free from the flow streamline in the initial airways and deposit by inertial impaction. Impaction is the main deposition mechanism at the first five generations of the lung. Generally, large particle deposit in these region and swallowed eventually. Their contribution to the therapeutic response is negligible. Particles with slightly lower inertia can avoid the filtering and reach to the middle airways. The deposition of the particles is dominated by gravitational sedimentation. Therefore, sedimentation is the main deposition mechanism in the middle range of the generations (from 6-16). Only the very small particles ($<1 \mu\text{m}$) have very

lower inertia. They can reach to the alveolar region and with the influence of Brownian motion.^{6,8,30,31} Hence, Brownian motion is the dominating mechanism for deposition in the 17-23 generations.

2.3 Influencing factors

The above mentioned deposition mechanisms are influenced by three key factors. Firstly, the airway morphology which includes such parameters as airway diameters, lengths, and branching angles. Secondly, the ventilatory pattern which includes air flow rate in the lung airways and pattern of breathing. And finally, the inhaled drug formulation characteristics.^{10,32}

2.3.1 Airway morphology

Airway morphology is the most influential factor for the deposition of aerosolized drug particles in the lung. Diffusion and sedimentation are important in small airways because of their calibre and tortuosity, whereas inertial impaction is dominant in the upper airways. Interception mechanism for deposition of fiber particles mostly depends on airway morphology. Filtering capacity of the upper airway is also important in which about 70-90% of the aerosolized particles are removed³.

The human lung is separated into 23 generations as shown in Figure 2.1. A new generation possesses a narrower airway than the previous generation. Hence, it is getting difficult for the particles to flow in the lower generation of lung airways. Eventually, the particles get deposited. This deposition is high for large particles. Branching angle at the bifurcations is another anatomical factor for this deposition. If the branching angle is too large, particle with high inertia cannot follow the airway geometry. The particles would

break out from the air stream and collide with the lung wall. It is important to note that these factors vary from person to person and people of different ages.

If the drug particles are hygroscopic, the high relative humidity of human lung can enhance their deposition. Hygroscopic drug particles can grow or shrink due to addition or removal of water.³³ The hygroscopic growth rate depends on the initial diameter of the particles. Generally, the growth is higher for smaller particles compare to larger particles.^{11,34,35} This phenomenon also influences the drug deposition in different stages of lung airway.

2.3.2 Ventilatory pattern

2.3.2.1 Inhalation flow rate and pattern

Particle deposition in the lung airways is highly influenced by inhalation flow rate. High inhalation flow rate causes early deposition to the lung. For ultrafine particles, the dependence of particle deposition on the flow rate is relatively weak. At lower flow rate their deposition efficiency increases slightly.³⁶ Besides, breath-holding is also of particular importance because it allows more residence time. By altering breathing profiles, deposition values can be affected regarding quantity delivered. Longer breath-holding times and higher tidal volumes increase deposition in the pulmonary region, whereas increased inhalation flow rates increase deposition in the tracheobronchial region.^{3,37}

2.3.2.2 Inhaler design

It is essential for the patients to produce an air flow that is required for efficient aerosolization and de-agglomeration of the drug particles and their deposition.³⁸ Particle velocity and their possibility to go to the deep lung depends on the air flow generation.

Specific design of the inhaler is very critical to generate sufficient air flow to disperse the drug formulation and turbulence to break up the aggregates and deliver them into therapeutically effective region of the lung.²¹

In pulmonary drug delivery practice, flow rate through the inhaler depends also on intrinsic resistance of the dry powder inhaler device. Difference in design offers different resistance to the inhalation airflow rate.³⁹ The flow rate controls the particle lifting from the chamber. After the dispersion, de-agglomeration of the agglomerates of drug and carrier is occurred by shear forces and turbulences generated by the airflow in the inhaler and the initial airways.^{21,40,41} Hence, aerosolization of the formulation from the inhaler and deposition in the deep lung depends also on the inhaler resistance.^{42,43}

Low resistance inhaler would allow higher inhalation flow rate than the inhalers with high resistance. However, inhalers with low resistance cannot generate reproducible drug delivery performance and shows interpatient variability.⁴⁴ High resistance inhaler need high inhalation effort from the patients to deliver a good result. Nonetheless patients with persistent asthma disease (up to certain severity) can afford to use that up to certain severity.⁴⁵ It is also known that, inhaler resistance and flow rate are not the only factors for delivery efficiency. Their performance is also a function of the rate of increase in the flow or rise time to flow rate.⁴⁶ Therefore, patients need proper practice to optimize their inhalation technique for a optimum inhalation flow rate and rise time.⁴⁷

Some other aspects of the specific design of inhaler and their characteristics are also reported. Inhaler mouthpiece geometry has no substantial effect on the overall inhaler performance.⁴⁸ With the increase of the grid voidage the amount of powder retention into the inhaler increases.⁴⁹ Length of the mouthpiece also exhibit negligible

effect on the flow field generated into the inhaler⁴⁹ whereas air inlet size has a varying effect on powder dispersion. Reduced air inlet size increases the inhaler dispersion performance by enhancing flow turbulence and particle impaction.⁵⁰ From the handful of reported studies regarding inhaler design, it can be anticipated that only the design aspects related to the turbulence into the inhaler have the important impacts on the delivery performance.

2.3.3 Formulation characteristics

Different techniques are followed to prepare drug formulation in pulmonary drug delivery. Though in some cases pure drugs are used, but the common practice is to mix drug particles with inactive excipient. The optimum size of drug particles is very small. Particles of small size range are not suitable for handling and dispersion because of their cohesion and static charge. Small particles also can induce drug retention in the inhaler and cause non-uniformity in delivering doses. Therefore, large carrier particles are used in the formulation which can improve their overall delivery performance. Thus the physical characteristics of drug and carrier particles influence formulation behavior.

2.3.3.1 Drug particle characteristics

Researchers have paid most attention to drug particle characteristic because both airway morphology and ventilatory pattern cannot be controlled easily and varies with patients. The most viable way of achieving better deposition thus relies on the optimum physical properties of the drug formulation. The penetration distance and rate of deposition of dry powder formulation in the airways depend on different factors such as particle size, density, shape, electrostatic properties and hygroscopicity^{29,51}.

Size

Aerosol particles always comprise of wide range of sizes. Different type of size distributions can be found from a powder sample, e.g. based on the particle area, volume, mass or number. For the actual representation, size distribution based on the particle number may be misleading, as smaller particles contain fewer drugs than larger ones. Thus, in this distribution the log of the particle diameters need to be plotted against particle surface area or volume.

Several parameters are being used to define particle size from the distribution. Mass median diameter is one of the common parameter which defines that 50% of the aerosol resides above and 50% remains below that diameter. This parameter also sometime is related to the aerodynamic diameter of the particles, which named as mass median aerodynamic diameter (MMAD). MMAD is determined from the cumulative mass distribution as a function of the logarithm of aerodynamic diameter. It is estimated as the 50th percentile of the aerodynamic particle size distribution based on sample mass.

Particle size is the most important factor affecting particle deposition in the lung. As aerosol particles comprised of irregular size, shape and density, aerodynamic diameter is used as the common basis to characterise the deposition. Aerodynamic diameter is an equivalent diameter that standardizes aerosol particles to describe their aerodynamic behavior. The standardization is based on the terminal setting of the concerned particles. Aerodynamic diameter is defined as the diameter of a sphere of unit density that has the same terminal settling velocity as the particle under consideration. For instance, the terminal settling velocity of a particle can be expressed with the Stokes's law as:

$$V_{TS} = \frac{\rho_p g d_p^2}{18\mu} = \frac{\rho_o g d_a^2}{18\mu} \quad (1)$$

where d_g is the actual geometric diameter, d_a is the aerodynamic diameter, ρ_p is the actual density, ρ_o is the unit density, g is the gravitational acceleration and η is the viscosity of the medium. In general, the aerodynamic diameter of a particle is simply related to the actual diameter and density of the particle as:

$$d_a = d_g \left(\frac{\rho_p}{\rho_o} \right)^{\frac{1}{2}} \quad (2)$$

An important assumption in Stokes's law is that the relative velocity between the gas and the surface of the sphere is zero. However, this is not the case for the small particles whose size is close to the mean free path of the gas. The settling is faster than predicted by Stokes's law because of the surface slip of the particle. This error becomes significant for particles less than $1\mu\text{m}$ in diameter. For these particles a slip correction factor (C_c) should be applied in the Stokes's law^{10,52}. Hence, aerodynamic behavior of therapeutic aerosols for pulmonary drug delivery can be assumed in terms of the physical properties like particle size, shape and density.

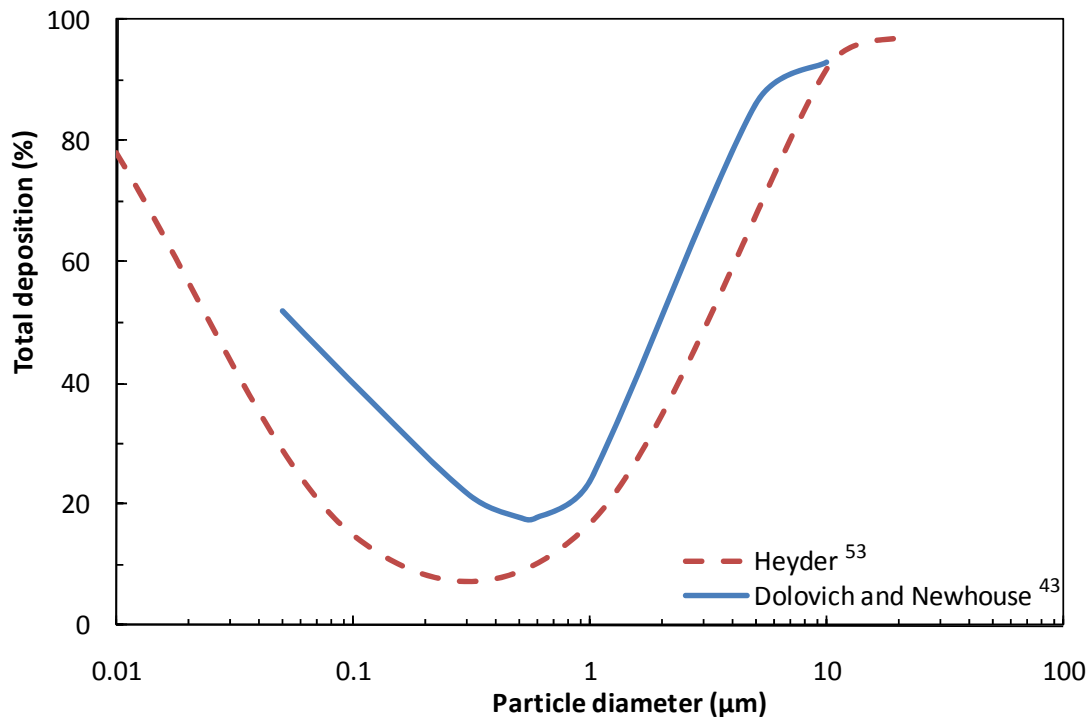


Figure 2.2 Total deposition of unit density spheres in respiratory tract.

Particle size also influences their deposition mechanism. With the decrease of the particle size their diffusional transport increases. Particles with $d_a < 0.1 \mu\text{m}$ in diameter mostly deposit by diffusion. This deposition decreases with an increase of particle size up to about $1 \mu\text{m}$. For larger particles, deposition due to diffusion is almost negligible. In Figure 2.2, it can be seen from Heyder's⁵³ model that particles larger than $0.1 \mu\text{m}$ deposit with the additional affect of gravitational sedimentation and it increases with particle size. However, sedimentation is also function of particle density and respiratory cycle period. According to Heyder⁵³, particles with a size range 0.1 to $1 \mu\text{m}$ range deposit with the simultaneous effect of diffusion and sedimentation. However, Dolovich and Newhouse⁴³ reported the range around 0.3 to $1 \mu\text{m}$. These studies demonstrated that particles with a size range 1 - $10 \mu\text{m}$ deposit by sedimentation and inertial impaction

simultaneously. For larger particles, impaction is the main deposition mechanism and it increases with particle size, density and flow rate of air.

Studies reveal that the upper size (MMAD) limit of aerosol particles for airway penetration under normal breathing conditions is 23 μm .⁵⁴ Effective deep lung deposition is achieved only by the particles which are significantly smaller than 10 μm .⁵⁵ Warke et al.⁵⁶ demonstrated that the aerodynamic particle size range for pharmaceutical aerosols appears between 0.3 and 25 μm . However, for dry powder inhalation, drug particles are formulated with a size range below 5 μm .⁵⁷ This size may be equivalent to the particles with higher geometric size depending on their shape and density.⁵¹

Shape and morphology

In general, particles with non-spherical shape have an irregular drag force and terminal settling velocity for their irregular shape effect. This requires a dynamic shape correction factor in Stokes's law to correct the effect of shape irregularity²⁹:

$$V_{TS} = \frac{\rho_p g d_q^2}{18S\mu} \quad (3)$$

where d_q is the equivalent volume diameter and S is the dynamic shape correction factor. Then the expression for the aerodynamic diameter corrected for shape would be:

$$d_a = d_q \left(\frac{\rho_p}{S} \right)^{\frac{1}{2}} \quad (4)$$

The shape factor denotes the deviation of shape from a uniform sphere^{29,58}. It can be indicated by the ratio of the drag force experienced by the concerned particle and the drag force for a sphere having the same volume.

Elongated fibers and other rod-like particles have unusual airborne characteristics because of their irregular shapes.^{29,59} The aerodynamic properties of individual fibers may be determined in terms of length(l), diameter(d) and aspect ratio(l/d) of the particles.²⁹

Deposition does not vary significantly with a particle length. The main reason is that the fibrous shape elongated particles can follow the direction of gas flow.⁶⁰ Aerodynamic diameter of fibers are independent of length, proportional to $(l)^{1/6}$, therefore it is convenient to transport and deliver large quantity of elongated shape fibers of drug deeply into the human lungs without adversely affecting its airborne behavior.^{29,61} More efficacious therapeutic effect in the deep lung was noticed for elongated crystals.^{29,62} However, opposite result is also found where nearly spherical particles is more favourable in deposition efficiency in comparison with the needle shape long particles.⁶³ Hence, the effect of sphericity and shape irregularity of particles was not stated clearly. Elongated particles can also deposit by interception mechanism.⁶⁴ When the centre of mass of a particle stays on a fluid streamline but a part of it comes into physical contact with an airway surface, then particles deposit by interception.^{29,64} In most of these studies, nebulization procedure was involved where drug particles are suspended in a solution. Hence, extensive studies are not available for dry particles of different shapes.

Adhesion and cohesion hamper the dry particle dispersion and aerosolization. These attractive forces reduce the dispersibility of the particles. Some techniques were adopted to reduce the attractive forces in the study of deposition of dry particles. The use of spacer molecules is known as a good solution. Spacers can be small molecules attached onto the interacting particles with dimension larger than the interparticle voids. Apart from this, surface characteristics can also influence the particle aggregation behavior due to the screening effect.⁶⁵ Surface coatings reduce particle interaction to increase dispersibility which is an important factor for better delivery efficiency.²⁹ Formulation technique also influences particle morphology which has a greater effect on

adhesive force. Spray dried particles, having a smaller adhesive force between primary particles, show a high deposition percentage. On the other hand jet mill particles are not so spherical and because of point and surface contact the adherence with each other is higher whereas spray dried particles get adhered mainly by point contact.⁶⁶

Density

Particle density influences their aerodynamic diameter. From equation (4) it can be seen that the aerodynamic diameter is proportional to the square root of the particle density. Therefore, different aerodynamic diameter can be found for a constant geometric diameter particle by changing its density. It is found that large geometric diameter, low-density particles is an effective way to improve inhaled therapeutics delivery.

Large porous particles

Large porous particles of required aerodynamic diameter have high efficiency of particle deposition in the lung because of their better dispersion quality. For instance, porous particles of respirable size range can be achieved with a density much lower than 1 g/cm^3 and a geometric particle size around $10 \text{ }\mu\text{m}$.^{67,68} Porosity of the particles are helpful for better dispersion because of the larger geometric size to the particles. This property eventually reduces the van der Waals force which is responsible for particle interaction, aggregation and coagulation. As a result, deposition is improved in the alveolar region. This kind of lung deposition reduces loss of valuable drug particles and side effects caused by extrathoracic and tracheobronchial airway deposition. Moreover, the increased particle size reduces the effect of diffusion on deposition. Therefore, their improved delivery efficiency is due to the dispersion properties and minimal deposition occurring in the oropharyngeal region.⁶⁹ The large porous particles also assured the

advantages of targeted deposition, reduced macrophage clearance and enhanced bioavailability. Different formulation techniques have been used to produce porous particles for improved delivery, e.g. supercritical fluid technology⁷⁰, spray drying^{71,72} etc. Generally, porous particles produced by spray drying are formed in thin walled hollow morphology which would collapse in drying.^{67,73} A stable porous structure was also produced by modified spray drying named as PulmoSphere[®]. PulmoSphere[®] particles are with a hollow and porous morphology with a physical diameter <5 μm and low densities. The particles showed improved targeting of drugs to the lower respiratory tract from MDIs which is suitable for the delivery of systemically acting molecules absorbed via the lung. It is reported that this PulmoSphere[®] can have a very high respirable fraction with different kinds of drugs like cromolyn sodium,⁷⁴ budesonide,⁷⁵ albuterol sulfate⁷⁶ etc.

Hollow nanoparticle aggregates

Nanoparticles have good drug release and delivery potential. However, nanoparticles exhibit severe problem in powder flowability, dispersion and handling. The potential difficulties are tried to solve by implying better aerosolization and deposition behavior of large porous particles. The novel engineered aggregated particles exhibit much better flow and aerosolization properties than nanoparticles. The large aggregates dissolved into nanoparticles in physiological conditions. A new technique is proposed recently to formulate hollow nanoparticle aggregate for inhaled delivery by considering their potential as therapeutic carriers. In this formulation technique, large hollow carrier particles are fabricated by spray drying technique using nanoparticle aggregate, where this nanoparticle aggregates are acting as the shell of the large particles.⁷⁷⁻⁸¹ However, due to the hollowness, the particles have low density and aerodynamic diameter. The

physical properties would make the particles viable option for effective lung deposition. These carrier particles are formulated with a geometric diameter about 10 μm and effective density around 0.1 gm/cm^3 which have high flowability and better therapeutic efficacy. Different important drugs for lung and systemic diseases are formed into nanoparticle aggregates for pulmonary delivery, e.g. rifampicin with PLGA (poly (lactic-co-glycolide)),⁸² chitosan with lactose and mannitol,^{83,84} insulin,⁸⁵ ovalbumin,⁸⁶ doxorubicin⁸⁷ etc. Similar approach is addressed by producing flocculates from nanoparticles with required characteristics suitable for dry powder inhalation in some recent studies.⁸⁸

Hygroscopicity

Another important property of the particles for lung deposition is the hygroscopicity. Under high relative humidity hygroscopic particles exhibit physical growth or shrinkage and that would affect particles aggregation and dispersion as well as the aerodynamic behavior of the particles.^{89,90} Hygroscopic growth due to the moisture would increase particle interaction, therefore they would make aggregates and deposit early in the lung. The environmental condition of human lung is vulnerable to hygroscopic growth and it is a common problem encountered for most pharmaceutical aerosols. However, the hygroscopic effect of the particles can be reduced by using hydrophobic additives in the formulation which can repel the hygroscopic growth of the particles.^{91,92} the main significance of particle hygroscopicity lies in its potential to enhance or reduce deposition for intermediate-sized and fine particles, respectively.⁹³

2.3.3.2 Carrier particle characteristics

For effective dry powder inhalation, drug particles need to be micronized to a size between 1-10 μm .⁹⁴ Nonetheless, the inter-particle forces are considerable for particles between 1-10 μm .^{10,65} They form aggregates and difficult to disperse by air flow. To dissolve this problem, the micronized drug particles are physically mixed with large sized carrier particles.

In the formulation, the micronized particles could form aggregates with the carriers, like Turbuhaler[®] (Astra Zeneca) uses aggregates of budesonide, lactose and formoterol and agglomerates of the mometasone furoate and lactose is found in Twisthaler[®] (Schering-Plough) with appropriate size and hardness. The entire delivery process is comprised of some steps. Firstly, the dispersion of the powder from the inhaler; secondly, the detachment of the drug particles from the carrier and finally, the eventual deposition of the drug particles in the lower airways.⁹⁵ Consequently, any factor that effect these steps would change the therapeutic activity.

The carrier particles are generally of large size. Hence, they deposit early in the airways and usually swallowed. For deep lung deposition, drug particles must be detached from the carriers during inhalation to avoid early deposition.⁵¹ Detachment of drug particles from the carrier particles is mainly influenced by the size of the carrier. Adhesion and friction characteristics of the drug and carrier particles are also important factor in this phenomenon. Drug detachment was found to be easier for smaller carrier particles.⁹⁶ It has been modeled with lateral detachment of the drug particle from the carrier surface due to the drag force⁹⁷. Drug particles detach from the carrier by sliding and rolling along the surface of the carrier particles. This phenomenon could be hindered

by the adhesion and friction forces which need to be overcome to ensure the movement of the drug particles.

Carrier particles facilitate the handling of the powder and the filling of the powder in capsule. They also increase dispersibility of the drug particles during the aerosolization from inhaler which improves the total emission of the drugs. The volume of the formulation is high after mixing with the carrier and it is easy to meter a larger volume than smaller volume. Carrier particles also help in improving dose uniformity.⁹⁸ The choice of carrier demands some materialistic considerations. The carrier materials need to be available in pharmaceutical grade, non-toxic, no harmful impacts on the respiratory tract, bio-compatible, have high chemical and physical stability, inert with the drugs, biodegradable and can be cleared from the airways easily.⁹⁹ The physical properties of carrier that influence the inhalation performance of dry powder formulation are described here.

Size

The size of carrier particles for dry powder inhalation studies used in the literature covers a wide range of 10-220 μm .^{40,51,95,96,98-116} However, 30-90 μm is found as the most commonly used size range. There are mixed reports on the most efficient size range for carrier particles. French et al.⁴⁰ showed that large (90-125 μm) carrier particle exhibit better emission and deposition behavior than smaller particles (38-75 μm). On the other hand, Steckel and Muller⁹⁶ found that carrier particles with small size (<32 μm) have higher respirable fraction (also known as Fine particle fraction, FPF, which is described in the Experimental chapter) than larger particles (63-90 μm and 125-180 μm). In some other studies, it is also found that FPF increases with the decrease of carrier particle

size.^{110,115-117} However, if the particles are very small, inter-particle interaction among the particle would be prominent and there would be difficulty in dispersing them. Bell et al.¹¹¹ found the 30-90 as the most suitable size range for the carrier particles. Nevertheless, it is accepted that particle size distribution of the carrier has a significant impact on the drug delivery efficiency.

Morphology and Surface roughness

Aerodynamic behavior of carrier particles can be improved by changing their morphology. Thus, the use of carrier particles with different morphology has attracted great interest in aerosol science. Aerodynamic diameter of elongated shape particles, like needle and fiber like particles, is approximately independent of their length and depends on their shortest dimension. Therefore, the aerodynamic diameter of these particles would be low and they can travel a long way in the lung airways and allow a longer time for drug detachment which increases the probability of deep lung deposition. Elongated carrier particles with different shape, like tomahawk,¹⁰⁷ needle¹⁰⁰ etc. exhibit high FPF of the drug particles. However, large elongated particles also suffer from deposition due to interception. Moreover, because of their large contact surface area, dry elongated particles experience high interaction which would affect their dispersion quality. Hence, an increase in the elongation ratio of carrier particles can reduce drug emission.¹⁰⁰

Surface properties of the carrier particles also influence drug attachment and liberation in the lung. Most of pharmaceutical grade available carrier particles for dry powder formulation have crevices on their surfaces. Specially the carriers generated from natural sources would have irregularities (microscopic amorphous region on the surface) due to their production and processing method.¹¹² These irregularities are characterized

by surface roughness of the particles. They create more active binding sites on the surface of the carriers which facilitate the attachment of the drug particles.^{98,101} Therefore, particles with high roughness can accommodate large number of drug particles and they also improve the blend homogeneity and stability.^{104,107} However, if the crevices are large than the size of the drug particles, than the drug particles might be trapped. The liberation of these entrapped drug particles would be low by the inhalation force applied by the patient. Hence, drug particles would be deposited early with the carrier and the probability of lower airway deposition would be severely reduced. By investigating different grade of lactose⁹⁸ and their surface modification,¹⁰⁶ it is also reported that a small amount of surface roughness would increase respirable fraction.

On the other hand, smooth surface has larger contact area with the drug particles than rough surfaces. Therefore, a smooth carrier surface has stronger adhesion force on the drug particles and shows a better attachment. They can carry the drug particles properly into the lung and increase the emitted dose.⁹⁸ However, due to the limited detachment of the drug from the carrier surface, a low FPF may be observed,^{104,106,108} though a high FPF with smooth carriers produced by crystal engineering is also observed by some researchers.^{107,118}

Fine particle mixing

Recently the using of fine carriers in dry formulation is approached by the researchers to improve drug delivery to the airways. In this method, a small amount of fine particles are added to the binary mixture of the carrier and drug.^{95,103,109,119-124} Fine carrier particles influence the magnitude of interaction between drug and carrier particles to improve their liberation. After the attachment with the fines, the carriers would be a

binary carrier system. It is hypothesized that fine particles attach to the high energy active sites on the surface of the carrier particles, they saturate the active sites and thus the drug particles attach to the low energy sites of the surface.¹²² Therefore, the adhesion forces between the carrier and drug particles would be low and they can be liberated from the surface easily by the inhalation forces.

Fine particles in the dry powder formulation help to improve the liberation as well as the respirable fraction (FPF) of the drug. Addition of the fine magnesium stearate, leucine and glucose increase the deposition of salbutamol sulfate.¹²¹ Micronized fine lactose particles can also improve the dispersion and deposition of different drugs, e.g. salbutamol sulfate,^{115,122} beclomethasone dipropionate¹²⁴ etc.

The concentration and size of the fines and their mixing sequence need to be optimized sophisticatedly to achieve the satisfactory drug delivery result. FPF increases with the increase of the concentration of the fines in the mixture.¹¹⁰ However, it is also reported that fines up to a concentration of 5% can improve the FPF; further addition of the fine has no effect on FPF¹²³. The mixing of the large carrier and the fine at first and then blending the drug with the binary mixture was found to be as the optimum mixing sequence.¹⁰⁹

Though the inclusion of fine particles has a high potential to improve the delivery performance of carrier particles, there could also be a possible trouble with this technique. A large portion of the fine carrier particles exist in amorphous form and at the exposure in moisture they transformed into crystalline form. The transformation of the amorphous fine particles could reduce the flowability and dispersion of the powder formulation.¹⁰⁵

2.4 Summary

From this review, it can be summarized that pulmonary drug delivery can be influenced by different factors. However, drug and carrier particle characteristics are the most feasible way to improve deep lung deposition. Hence, the knowledge of the effect of these characteristics on delivery efficiency is essential. It is found that limited studies of the shape and morphology of the particle formulation has done yet. Though elongated shape particles in irregular shape are addressed in some studies, other shape particles remain unstudied and particle surface irregularities are also reported with considerable inconsistency. The existing studies are also not enough to explain the effect of carrier shape and morphology on their attachment and detachment with the drug particles. Therefore, the insights of the particle shape effect on interaction with other particles and their flow, dispersion and deposition behavior would be an area of great interest. This knowledge would be helpful in developing novel formulation with engineered drug and carrier particles to improve the efficiency of pulmonary drug delivery.

2.5 Objective and scope

To improve the inhalation performance of aerosol therapeutics, understanding of the fundamental influencing factors is essential. The objective of the study is to understand particle shape and morphological effect on inhalation performance. Hence, aerodynamic flow behavior, dispersion and deposition performance of different shape particles are investigated experimentally by applying macro and micro particles. Based on this investigation, the knowledge of optimum shape of drug and carrier particles for efficient inhalation is obtained. The scope of this research includes a number of aspects in the following,

i) Particles in pulmonary drug delivery need to flow through airways of different diameter. In these airways, they could experience wall effects in the confined medium. Therefore, the study is initiated with a preliminary investigation of the irregular particle shape effect on wall effect during gravitational settling in a macro-particle system. Though the wall effect studies are mostly dealt with spherical particles, a gap in the knowledge for the wall effect for other shape particles is found. Therefore, influence of irregular shape particles on wall effect need to be studied.

The study involves settling of different shape particles in cylindrical columns with different wall diameters. This macro level observation is helpful for the understanding of micro-particle flow and deposition phenomena in the lung airways.

ii) For a comprehensive shape study, investigation with micro-particle system is essential. Shape of the micron size particles is one of the influential factors. Previous studies cover mostly spherical particles and a few cases of elongated particles. The behavior of other non-spherical particles remains unseen. Moreover, shape factor is not sufficient to account the shape effect on the aerodynamic behavior. The behavior is also influenced by particle orientation and the contact area with other particles. Therefore, the understanding of the effect of different shape particles on flow behavior, aerosolization and depositions is necessary.

In this study, different shape micro-particles are synthesized with a similar micron size range. The size and shape of these particles are characterized and their flowability, dispersion and deposition behavior are assayed through *in vitro* inhalation experiments. Shape effects on the inhalation performance of these particles are analyzed. Optimum

shape to improve dispersion and flowability are addressed for further studies in dry powder formulation.

iii) Similar limitation in the shape study is observed for carrier particles in dry powder formulation. Though carrier shape and morphology have great impact on the flow behavior and delivery performance, not many shapes are investigated for improved results. Therefore, particles with the potential of good flow behavior need to be investigated physically before use them in *in vitro* experimentation in a blend with drug particles.

In this part of the study, particles with the preferable shape and morphology are synthesized with different size ranges. The flow behavior, velocity field and turbulence occurrence of the particle laden gas flow in an idealized inhalation path model are investigated physically using proper measurement technique.

iv) Large rock-shape lactose particles are the most commonly used carrier in dry formulation where a few studies are also found for elongated particles. After observing the physical flow behavior of the large carrier particles with preferable shape, the *in vitro* flow, aerosolization and deposition behavior as a blend formulation with drug particles need to be studied.

The study involves large particles of the shape with preferable flowability and dispersion behavior employed in a drug formulation as carrier. The dry powder formulation is produced using a model drug. The drug loading capacity of the particles is also assessed. The performance of the binary mixture is studied *in vitro* to assess the feasibility of those carriers in dry powder inhalation.

v) Drug loading and delivery performance of the carrier particles are highly influenced by the drug mixing ratio and inhalation flow rate. However, the dependence of drug loading and detachment mechanism on carrier shape and morphology has not been studied before.

In this study drug loading capability of the carrier particles are determined by applying the binary mixtures with different drug mixing ratio. The effect of the drug loading of the binary mixtures and inhalation flow rate is assessed. The performance of the experimental shape of the particles is compared with traditional carrier. The impact of the physical characteristics of the particles on delivery efficiency is also assessed.

Chapter 3 EXPERIMENTAL

3.1 Settling experiment of different shape macro-particles

3.1.1 Particle fabrication

Different shape particles are fabricated to investigate their wall effect on settling in a confined medium for the preliminary study. Simple technique is used to obtain the particles. Metal (copper) wire with different diameters is cut into different lengths to get cylindrical particles with a range of aspect ratio. Commercially available glass beads with bicone and cube shape is used. Polyisoprene is cut into specific dimensions to get flat plate particles with different aspect ratios. The schematic of different shape particles can be seen in Figure 3.1. Settling velocities of these various shape particles are investigated in cylindrical columns of various diameters. In general, different settling medium, e.g. water, oil etc. is used to get separate regime of settling.

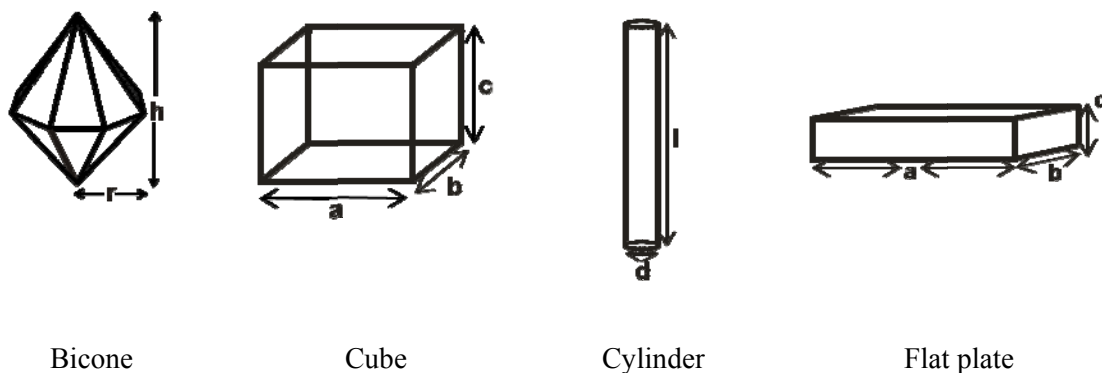


Figure 3.1 Schematic of the different shape macro-particles for the settling experiment. r and h are radius and height of the bicone, l and d are the length and diameter of the cylinder, and a , b , c are the three dimensions (width, length and height) of cube and flat

plate particles. For the flat plate particles dimension c is much lower than a and b where for cube shape particles all three dimensions are similar.

3.1.2 Velocity measurement: High speed camera



Figure 3.2 High speed camera with view monitor.

In Figure 3.2, the high speed camera (Olympus, i-Speed) used in the settling experiment can be seen. The camera uses a CMOS sensor with a resolution 800×600 and pixel size $14 \mu\text{m}$. It has a maximum frame speed of 33000 fps. For a high frame speed, an intense light source is needed for better illumination. A C-mount lens is coupled with the camera. The captured video or a required fraction of that is recorded in a compact flash (Type II Sandisk, 2 GB) memory card. The camera is connected with a monitor. The monitor comes with several buttons to control the operation of the camera easily. An i-

Speed software (Olympus) is provided with the camera to analyze the captured videos and measure the settling velocity of the particles. The settling orientation and behavior of the particles also can be analysed precisely using a high frame speed even for the particles settling with a high velocity.

3.2 Sample preparation

3.2.1 Preparation of hydroxyapatite (HA)

Hydroxyapatite (HA, $\text{Ca}_5(\text{PO}_4)_3(\text{OH})$) particles are synthesized by hydrothermal reaction using potassium dihydrogen phosphate (KH_2PO_4 , Merck, Singapore), Calcium nitrate tetrahydrate ($\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, Sigma-Aldrich, Singapore), poly(sodium-4-styrene-sulfonate) (PSS, Aldrich, Singapore), and urea (1st Base, Singapore).¹²⁵ 30 ml of KH_2PO_4 (0.02 M) solution is added with 50 ml of $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ (0.02 M) solution. PSS and urea are then added to the mixture to get a required concentration. The mixture is stirred gently for half an hour to dissolve the urea properly. The solution mixture is then kept in an autoclave and put into an oven at a higher temperature. For the synthesis a reaction time of 6 hrs is used. Finally, the precipitated product is collected by centrifugation, washed several times and then dried at 70°C.

To control the morphology and size of the produced HA particles, the PSS concentration, urea concentration and reaction temperature are varied. Different types of HA particles are assigned with distinct names in the following studies and their required reaction parameters are presented in Table 3.1.

Table 3.1 Different reaction parameters for the produced HA particles

No	PSS conc. (g/L)	Urea conc. (M)	Temp. (°C)	Time(hr)	Notation
1	40	7.5	200	6	Pollen-1 (Ch.-5)
2	40	3	200	6	Pollen-2 (Ch.-5)
3	80	7.5	200	6	Spherical-2 (Ch.-5)
4	40	0.5	150	6	HA-3 (Ch.-7)
5	40	0.5	120	6	HA-1 (Ch.-6, 7, 8, 9)
6	30	0.5	200	6	HA-2 (Ch.-6, 7, 9)

3.2.2 Preparation of other different shape particles

Syntheses of other different shape particles are carried out by precipitation reaction. In the precipitation reaction, fixed amount of aqueous reactant solutions are mixed in a conical flask. The surfactants are added in required amount where needed. Then, the pH of the mixture is maintained at a certain level by using dilute HCL or NaOH. The mixture is kept at a constant temperature under static or stirred condition for required time. After the reaction, the product is collected, washed and dried at 70 °C. Depending on the size and morphology of the particles, the parameters are varied.

Spherical calcium carbonate (CaCO_3) particles are produced by using calcium nitrate tetrahydrate $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, sodium carbonate (Na_2CO_3 , Kanto Chemical, Singapore) and PSS using precipitation reaction.¹²⁶ Plate-shape calcium oxalate (CaC_2O_4) particles are produced by precipitation reaction of sodium oxalate ($\text{Na}_2\text{C}_2\text{O}_4$, Kanto Chemical, Singapore) and calcium chloride (CaCl_2 , Kanto Chemical, Singapore) using PSMA (poly-(styrene-alt-maleic acid), Sigma-Aldrich, Singapore) as the surfactant.¹²⁷ Cube-shape CaCO_3 is produced by precipitation reaction of Na_2CO_3 and CaCl_2 using

CTAB (Cetyl trimethylammonium bromide) as the surfactant.¹²⁸ Needle-shape CaCO_3 particles are synthesized by precipitation reaction of potassium hydrogen carbonate (KHCO_3 , Merck, Singapore) and CaCl_2 .¹²⁹ The material and reactants of the different shape particles with their references are listed in Table 3.2.

Table 3.2 Reactants for the synthesis of different shape particles

Product	Shape	Reactants	Ref.
calcium carbonate (CaCO_3)	Spherical	$\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, Na_2CO_3 , PSS	126
calcium oxalate (CaC_2O_4)	Plate-shape	$\text{Na}_2\text{C}_2\text{O}_4$, CaCl_2 , PSMA	127
CaCO_3	Cube-shape	Na_2CO_3 , CaCl_2 , CTAB	128
CaCO_3	Needle-shape	KHCO_3 , CaCl_2	129

3.2.3 Preparation of LA

α -Lactose monohydrate (LA, Sigma-Aldrich, Singapore) is used as the control carrier for comparison with other carrier particles. The LA particles are separated into different size range by sieving with a mechanical shaker (Retsch GmbH, Haan, Germany). 20 g of the LA sample is sieved using test sieves. The test sieves are of 100 mm diameter with different apertures. The maximum vibration power of the shaker is used for the separation for several hours. To avoid the loss of the sieving particles, the test sieves are covered tightly with the clamp. The mechanical shaker used for the separation can be seen in Figure 3.3.

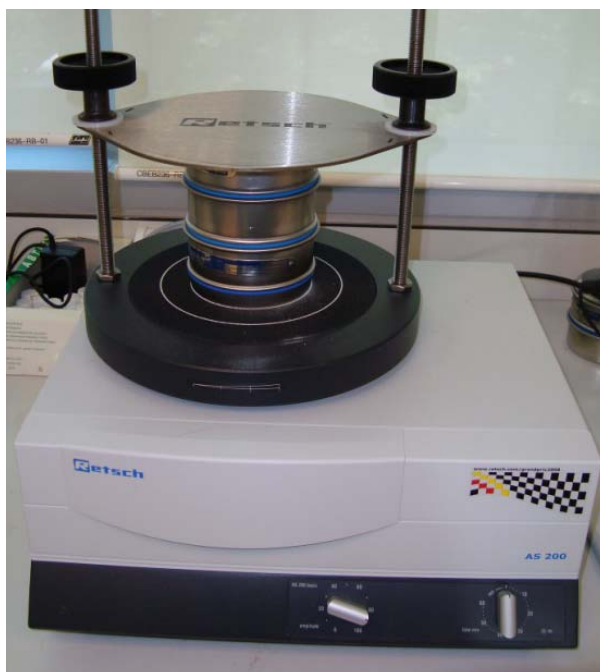


Figure 3.3 Vibrating sieve shaker with the sieves.

3.2.4 Blend preparation

Blends are prepared physically by mixing drug and carriers particles at different weight ratio. At first the fixed amount of carrier and drug samples for a certain mixing ratio are weighted on a balance. The measured samples are poured in a clear sample vial. Then the sample in vial is mixed properly using a REAX top mixer (Heidolph, Kelheim, Germany) for 15 min at 1000 rpm. After mixing, the attachment of the drug particles with the carrier surfaces are characterized with scanning electron microscopy (SEM) and the blend homogeneity is measured with UV spectrophotometer.

3.3 Sample characterization

3.3.1 Size measurement

The particle sizes are measured by laser diffraction using a Mastersizer 2000 (Malvern Instruments Ltd, Malvern, Worcestershire, UK). It is a simple and straight forward way of particle sizing that can offer very high accuracy. Particles with a size

range of 0.2 to 2000 μm can be measured by the particle sizer. It is suitable for the measurement of emulsions, suspensions and dry powders. Before each measurement, the sampling chamber of the particle sizer is cleaned with deionised (DI) water three times. This would help to avoid any contamination from the previously examined particles. Then the chamber is filled with DI water and the particle sample is dispersed in the liquid. For some samples, 1% IPA solution in DI water is used for better dispersion. The sonicator inside the sampling chamber of the particle sizer ensures the homogeneity of the particles in the suspension. Then the particle sizer is run three times to get the size distribution of the sample. The average of the three runs is used as the actual size distribution. A sample particle size distribution obtained from the sizer is shown in Figure 3.4. The laser diffraction data measured are also provided in terms of particle diameter at 10%, 50% and 90% of the volume distribution.

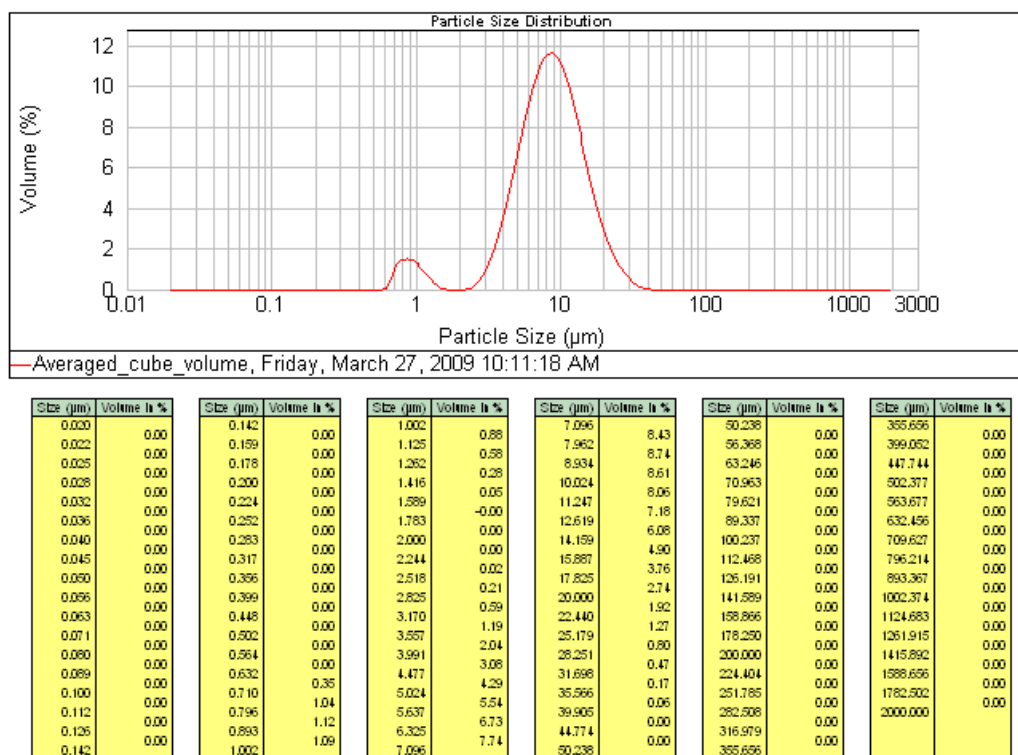


Figure 3.4 Size distribution of a sample produced from the Mastersizer 2000.

3.3.2 Size, shape and morphology

The size and morphology of the experimental particles are also characterized by the SEM (JSM-5600, JEOL, Tokyo, Japan). The possible accelerating voltage of the electron beam is 0.5-30 kV. Pre-centered W hairpin filament with continuous auto bias is used for the SEM. Super conical lens is used as the objective lens. Maximum area with a diameter of 32 mm can be covered with the SEM.

In general, for high magnification high accelerating voltage is used. The dry particles are dispersed on a double sided carbon tape attached on a stub. Then the particles are coated with platinum under an argon atmosphere (JFC-1600, JEOL, Auto Fine Coater, Tokyo, Japan) for 60 sec with a current 20 mA. Then the coated particles are examined under SEM. The SEM images are taken randomly from different areas of the samples. The images are analyzed to get the distribution of the geometrical size of the particles. A sample image taken by the SEM and the geometric diameter of the captured particle is shown in Figure 3.5. Dimensions of large number of particles are measured for each sample to get the size distribution.

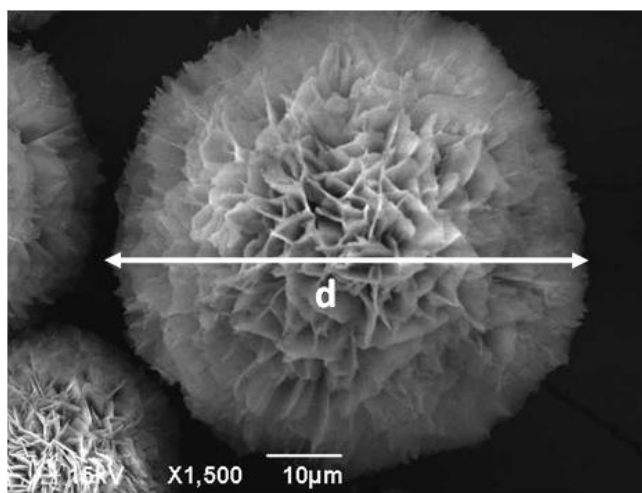


Figure 3.5 SEM image of a sample particle with $\times 1500$ magnification.

3.3.3 Powder density

The powder samples used in this study are also characterized with their bulk (ρ_{bulk}) and tap (ρ_{tap}) densities. When a powder is poured in a cylinder, the measured density of the powder at loose packing is called the bulk density. With a tighter packing, the powder volume reduces and density increases. Density measured at the tight packing is called tap density. The densities are measured manually by following the procedure from Shi et al.⁸⁸ Firstly, 100 mg of a powder sample is measured using an analytical balance. Then the measured powder sample is poured in a one mL micro-syringe tube. The volume of the powder with this normal packing without any tapping is measured. The tube containing the powder is then manually tapped on a tabletop at a constant rate of about 4 times per second. The tapping is continued for 2000-2500 times until no reduction of the volume of the powder is found. The measured density before the tapping at $n=0$ is ρ_{bulk} and at $n=2000-2500$ is ρ_{tap} .

3.3.4 Crystalline structure

The crystalline structure of experimental samples is analyzed using X-ray diffraction (XRD). XRD analysis are performed by LabX-Shimadzu XRD6000 (Shimadzu Corporation, Kyoto, Japan) diffractometer with Cu-K α as X-ray source ($\lambda = 1.5406^\circ\text{\AA}$). The maximum power output for the X-ray generator is 3kW and output stability is $\pm 0.01\%$ (for 10% power fluctuation). Maximum tube voltage and current are 60 kV and 80 mA, respectively. The goniometer is vertical type with a scanning radius of 185 mm. It can provide minimum step angle 0.002° (2θ) with an angle reproducibility of $\pm 0.001^\circ$ (2θ). The scanning angle range is $-6^\circ \sim 163^\circ$ (2θ) and speed is $0.1^\circ \sim 50^\circ/\text{min}$ (2θ), Scintillation counter detector is used in this diffractometer.

Experimental samples are shaped into a thin layer in the sample holder and then inserted in the path of X-ray in the diffractometer. The parameters used for the XRD analysis for the samples of this study are mentioned in Table 3.3.

Table 3.3 Parameters for the XRD analysis

Parameter name	value
Scan rate	0.5/min
Voltage	40 kV
Current	40 mA
Step size	0.02

3.3.5 Moisture content

Thermogravimetric analysis (TGA) of the samples is also conducted using Diamond TG/DTA.(Perkin Elmer, MA, USA). This instrument determines water and ash content in a sample. Reaction velocity and acceleration degradation can also be tested with this. In this study, moisture adsorbed on the surface of the particles is measured by this analysis. In this technique, weight change of the experimental sample is measured as a function of temperature and/or time.

The temperature range is from ambient to 1500 °C. A gas-cooling unit cools the furnace after each measurement to improve productivity. A base pan and a sample pan are used for the analysis. At first the empty base pan is calibrated with the empty sample pan. Then 1-2 mg of the sample is put in the sample pan. The aluminium crucible pan is heated up in the furnace to required temperature with a continuous heating rate of 10 °C min⁻¹. A nitrogen environment is kept in the system by maintaining a continuous flow of that. After the analysis the furnace is cooled with a automatic gas cooling unit to enhance productivity for the next analysis.

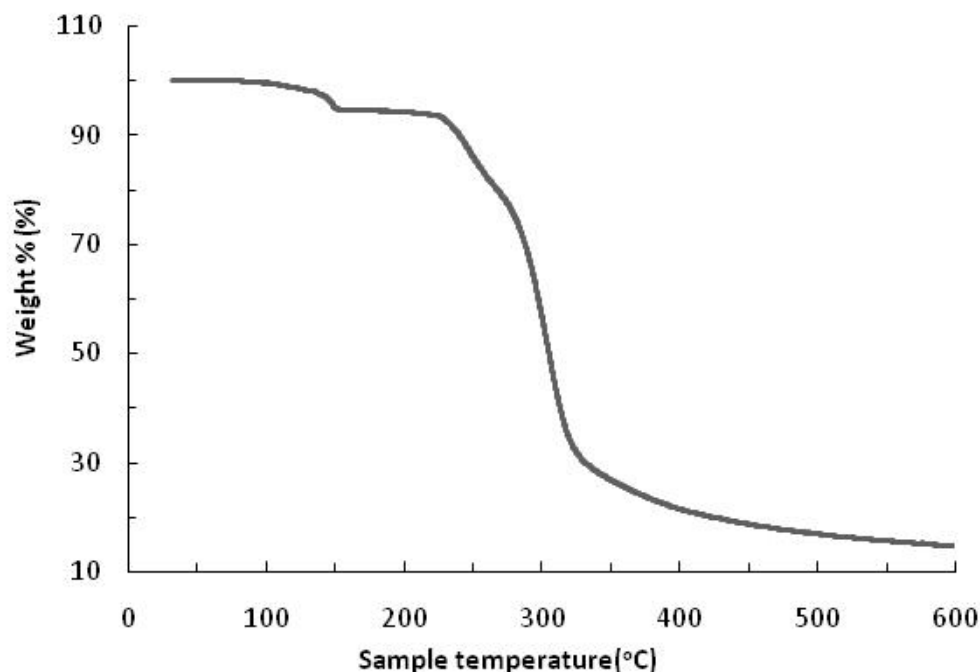


Figure 3.6 TGA thermogram of a sample.

A plot of the weight loss of the sample versus increasing temperature is produced by the TGA software. A sample of the TGA thermogram is shown in Figure 3.6. The decrease of the sample weight around 150 °C shows the loss of due to the evaporation of the water molecules in the sample. The sharp decrease in the sample weight around 250 °C is caused by the hydrolysis of the sample.

3.3.6 Specific surface area

BET surface area is a very important property for many types of advanced and pharmaceutical materials. BET stands for Brunauer, Emmett and Teller, the three researchers who optimized the theory for measuring surface area. The BET surface area of the prepared samples is measured using Autosorb[®] 6B (Quantachrome Instruments, Florida, USA) surface analyzer with nitrogen adsorption method. The pressure range for the equipment is 0-10 torr. It can provide high accuracy of 0.0015% (reading). Nitrogen

or any non-corrosive gas with appropriate coolant can be used as adsorbate. In N₂ environment, measured surface area ranges from 0.01 m²/g to an unknown upper limit. Pore size range is 3.5 to >4000 Å. Minimum pore volume can be measured 1×10⁻⁸ cc/g.

Sample preparation for the surface area analysis requires the elimination of surface contaminants. The contaminations are removed by heating the sample under vacuum in Autosorb[®] Degassar (Quantachrome Instruments, Florida, USA). It has six examination stations side-by-side. For independent analysis, each station can be controlled with separate controller. The temperature of the degassing can also be fixed for the individual samples. The heating can be terminated by a timer or manually. About 100 mg of the sample is weighted in a BET column. Then the column with the samples is dried in the degasser at 80 °C for 24 hrs under a flow of N₂. Then the sample is weighted again before putting under the BET analysis. This process needs to be done promptly to avoid any contact with the moisture in the air. After the BET analysis, this weight of the dried sample is used for the BET calculation for specific surface area.

3.3.7 Powder flowability

Powder flowability can be characterized by different measurement parameters. Carr's compressibility index (CI) and Hausner ratio (HR), are the main static flow measurements. Measurements conducted in rotating drum and vibrating spatula¹³⁰ are dynamic powder flowability. However, for small amount of samples the angle of slide¹⁰⁰ can be a representative dynamic flow measurement parameter. In this study, CI index and angle of slide are used as flowability parameters.

Carr's compressibility index

Powder flowability is characterized empirically using Carr's compressibility index (CI) based on the bulk and tap densities of the powder. CI is a common way to

characterize the flow behavior of particles. It is related with the compressibility of the powder particles, compressible powders have strong inter-particle forces and would exhibit poor flowability.⁹⁴ CI is calculated using tap density (ρ_{tap}) and bulk density (ρ_{bulk}) using the equation below:

$$CI = \frac{\rho_{tap} - \rho_{bulk}}{\rho_{tap}} \times 100\% \quad (3)$$

Lower CI values are indicative of better flowability.^{94,113} In general, CI value lower than 20% indicates good flow of powder.¹³¹ However, as CI is based on empirical understanding, it may pose discrepancies in result interpretation.

Angle of slide

The angle of slide method is a simple and effective dynamic measurement of powder flowability, especially for small amount of sample powder.¹⁰⁰ The measurement principle is based on the gravitational shear. A fixed amount of particle samples is placed on a dry glass plate. A lab jack is used to tilt one side of the plate vertically upwards until the major portion of the powder slides. The angle between the tilted and horizontal planes (angle of slide, θ) is then measured. The lower the angle, the better the flowability.

3.3.8 Blend homogeneity

Content uniformity of the blended samples (carrier and drug) is examined by analyzing the quantity of drug in the blend. Three 5 mg samples are collected randomly from each blend and kept in three separate sample vials. The dry powder samples are dissolved with fixed amount of solvent mixture of 2% Nitric acid (Fluka, Singapore) solution with ethanol (Merck, Singapore) in a ratio of 3:1(v/v). After complete dissolution, the drug content of the solutions is assessed using a UV spectrophotometer (Shimadzu Corporation, Kyoto, Japan). The wavelength at which the UV absorbance is

maximum for the drug solutions is measured. By using that wavelength, a calibration plot for that drug component is created. A number of standard solutions with known drug concentration are needed for the calibration curve.

At the beginning, a standard drug sample in the mentioned solvent is prepared with a known concentration of 100 ppm. Then this solution is diluted to different known samples like 1 ppm, 5 ppm, 10 ppm, 20 ppm, and 50 ppm. These standard solutions are used to get a calibration curve. The concentration of the unknown samples is measured using the calibration curve. Each solution is analyzed three times to obtain the average drug content. The content uniformity for the mixtures is estimated from the variation of the drug content results and expressed as the coefficient of variance (CV). CV is defined as the ratio of the standard deviation of the drug content to the average drug content in the samples as a percentage.

3.4 Determination of particle flow behavior in a idealized path model:

Particle image velocimetry (PIV)

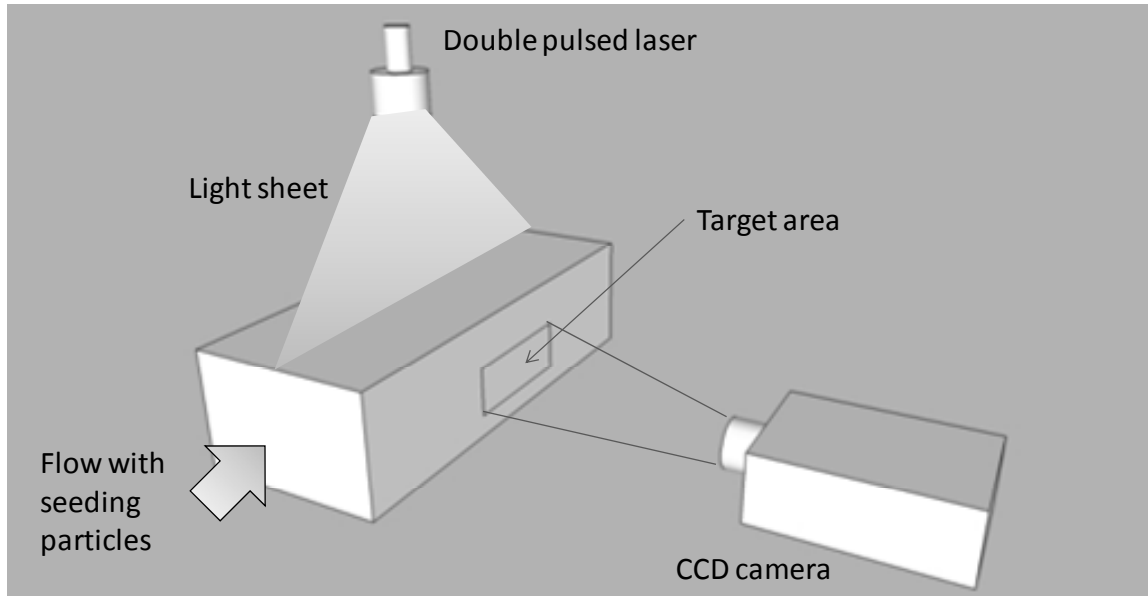


Figure 3.7 Schematic diagram of the PIV system

The flow behavior of the experimentally carrier particles flowing through an idealized inhalation path model is determined physically using particle image velocimetry (PIV). Figure 3.7 shows the PIV system comprising the CCD camera and laser system. The camera and laser system are connected with a computer to control the system and store the data. The principle of PIV is based on determining the movement of a number of particles into the flow. Two consecutive images of the flowing particles are taken by the camera and the average velocity profile of the flow is measured by analysing the positions of the imaged particles.

A Nd:YAG double pulse laser (Solo PIV, New Wave Research Inc.) is used as the light source to illuminate the particle flowing through the target area. This laser is capable of producing 30 mJ/pulse at 15 Hz. A CCD camera (TSI Incorporated) is placed perpendicular to the light source to capture the laser illuminated flow fields. The Δt is

optimized to 20 μs . The whole capturing process is controlled by a PIV computer together with a synchronized box. The capturing process is adjusted using the PIV through 3G software. The capturing process of the instantaneous flow field is time match the flow where the majority of the particles are passing through the region of interest. This allows the radial distribution of instantaneous particle velocity to be determined. The Nd:YAG laser fire two pulses at known time interval, where the CCD camera capturing a pair of instantaneous particle-induce image.

Once the camera system and laser sheet has been set up and aligned for a particular location, the working distance between the camera and image plane is measured. A series of preliminary PIV measurements are then carried out to find the optimum values of laser pulse delay (duration of the light pulses) and Δt (time interval between the two consecutive pulses of the laser). The optimum values of the laser powers are also determined for better imaging.

The image data are analyzed using the insight 3G software package from TSI. The vector fields are generated using Fast Fourier transform (FFT) correlator. PIV data is always arranged in a rectangular grid. The interrogation region is decreased from 64×64 pixel to 32×32 pixel with an overlap of 50% on the final pass. Recursive Nyquist Grid is selected as the grid engine. For most of the measurements the field of view is $25 \text{ mm} \times 35 \text{ mm}$. A range validation and a filter are used to avoid erroneous vectors. Flow characteristics are measured for the transitional flow of the samples. Therefore, the image pairs in which majority of the experimental area of the path model filled with vectors are selected for post processing.

Deposition efficiency

For deposition study, silicon oil is coated on the surface of the path model to prevent particle bounce. At the end of each deposition experiment, the particles deposited in the different regions of the path model are extracted separately. Regional deposition is quantified in different sections of the path model. Sample deposited different section of the path model is extracted using dilute acidic solution (2% HNO₃). Then the samples are quantified with inductively coupled plasma (ICP, Optica 2000 DV). For ICP measurement, a set of standard solutions with known concentrations are prepared, like 0 ppm, 5 ppm, 10 ppm, 20 ppm, 50 ppm and 100 ppm. Standard solutions are used to format a calibration curve of the measured sample. On the basis of the calibration curve, the concentration of the unknown sample is determined.

The deposition efficiency is defined as the amount of the sample deposited in a section per the total amounts of sample fed into the path model.

$$\% \eta_a = \frac{\text{deposition at region 'a'}}{\text{total deposition in the path model}} \times 100$$

3.4 *In vitro* aerosolization and deposition study: Andersen cascade impactor (ACI)

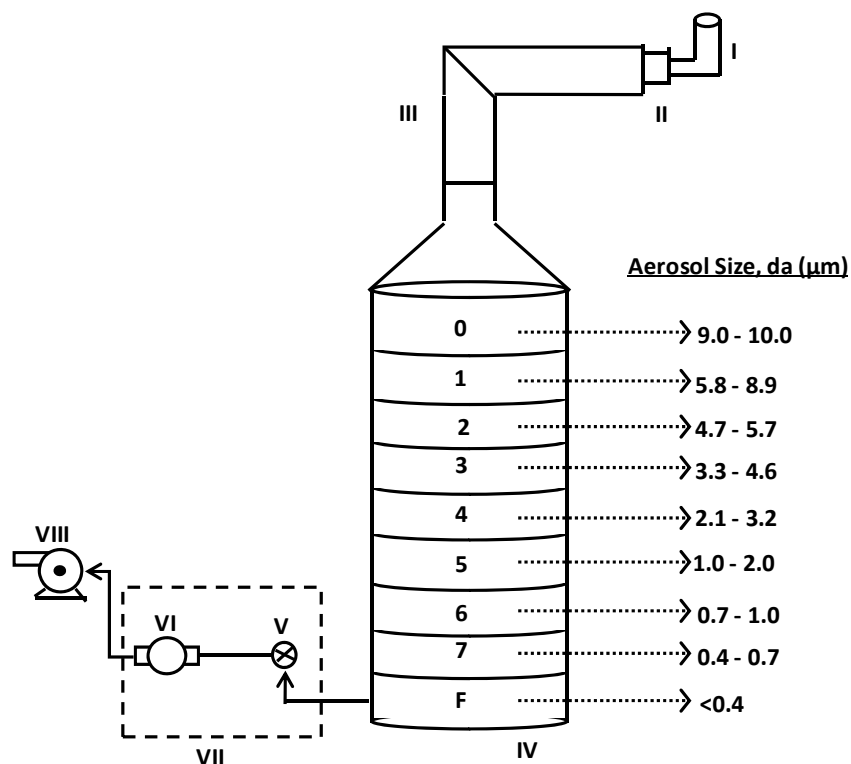


Figure 3.8 The particle size distribution in different stages of cascade impactor for a flow rate of 30 L/min along with different parts, I) dry powder inhaler, II) mouthpiece, III) throat, IV) stages of impactor, V) flow control valve, VI) two way solenoid valve, VII) critical flow controller, VIII) vacuum pump.



Figure 3.9 Experimental set up for the *in vitro* inhalation experiments using ACI.

The cascade impaction technique is commonly used for the dynamic characterization of pharmaceutical aerosol and currently it is in pharmacopeia requirements. The collection of particles in the cascade impactor is based on the inertial impaction of the particles on the collection plate. Large particles with sufficient inertia will impact on the collection plate of a particular stage. When the particles have insufficient inertia, they remain entrained in the airflow and move to the next stage. Literature confirms that in human lung normal impingement causes the major fraction of deposition.

The principle of the operation of cascade impactors is the aerodynamic behavior of aerosol particles. In this study, *in vitro* aerosolization and deposition experiments have been conducted using an eight-staged Andersen cascade impactor (ACI) (Copley Scientific Limited, Nottingham, UK). Figure 3.8 shows the schematic of the ACI along with the cut-off diameter of the stages of the impactor. The original experimental set up including the vacuum pump and the critical flow controller can be seen in Figure 3.9. The ACI is designed in such a way that the classification of particles would closely simulate

the biological classification of particles in human respiratory tract. Particles collected in different stages of the cascade impactor can be resembled to different regions of human lung.

The *in vitro* aerosolization and deposition properties of the experimental powder sample are determined using a Rotahaler[®] (Glaxo, UK) device. Experiments are carried out with a pre-separator at fixed flow rate. In each experiment, 8 ml of the extracting solvent is poured inside the pre-separator. A coating of 1% w/v solution of silicon oil in hexane is used on the impaction plates to prevent particle bounce and re-entrainment. A fixed amount of sample is loaded into a hard gelatin capsule (Gelatin Embedding Capsules, size 4, 0.25 cc, Polysciences, Inc., PA, USA) before putting into the Rotahaler[®]. Rotahaler[®] is used as the inhaler to aerosolize the powder inside the ACI. A consistent actuation time is used according to the flow rate for each capsule to completely disperse all the particles. Experimental runs are conducted in triplicate.

Particles remaining in the capsule, inhaler and different parts of the ACI are extracted. For the blend samples with Bd, the same solvent for the drug content measurement is used to extract the particles. The Bd concentration of the solutions is examined using UV spectrophotometer. For the extraction of single powder formulation, 2% nitric acid is used as the solvent. The extracted solutions from different regions are then quantified with ICP.

The *in vitro* aerosolization and deposition properties of the blends are commonly characterized by two parameters, namely emitted dose (ED) and fine particle fraction (FPF). ED is defined as the mass percentage of particles delivered from the inhaler (i.e., total amount excluding those in the inhaler and capsule). FPF is defined as the amount of

the particles deposited in stage 2 or lower in the cascade impactor for 30 L/min and stage -0 or lower for 60 L/min (particles <5.8 µm) as a percentage of the particles collected from all the parts of the ACI including those in the inhaler and capsule.

$$ED (\%) = \frac{\text{total amount excluding inhaler and capsule}}{\text{total amount deposit in ACI including inhaler \& capsule.}} \times 100$$

$$FPF_{30 \text{ L/min}} (\%) = \frac{\text{Particle deposit in stage 2 and lower}}{\text{total amount deposit in ACI including inhaler \& capsule.}} \times 100$$

$$FPF_{60 \text{ L/min}} (\%) = \frac{\text{Particle deposit in stage - 0 and lower}}{\text{total amount deposit in ACI including inhaler \& capsule.}} \times 100$$

Chapter 4 THE INFLUENCE OF COLUMN WALL ON SETTLING OF CYLINDRICAL PARTICLES WITH DIFFERENT ASPECT RATIO IN INERTIAL REGIME

4.1 Introduction

Gravitational sedimentation is one of the most important mechanisms for particle deposition in respiratory tract. In this preliminary study, effect of particle shape on settling in a confined medium is assessed using a macro-particle system. Settling column has several dissimilarities compare to human lung airways. The elastic, flexible surface of the lung channel is idealized to constant cross section with rigid wall to remove the complexities. In spite of these dissimilarities, the preliminary study is conducted because the settling under gravitational sedimentation is relatively simple to control. In this system, it would be easier to identify the effect of the different shape particles on the key mechanism of deposition. As the system involves single particle settling, surface deposition due to particle collision with the wall is not involved. The aim of the study is to investigate the movement of different shape particles in a settling medium under intermediate to inertia flow regime.

Particles settling in the presence of walls will experience a retarding effect. The presence of other particles and finite wall can cause a reduction in the settling velocity. The effect is more significant when the particle is close to the wall. Wall effects on

spherical particles under different flow conditions have been studied in the literature. However, studies for non-spherical particles are limited and cover only viscous flow regime.

The presence of walls or finite boundaries exerts a retarding effect on the settling of particles. The wall effect on a settling particle is defined as the ratio of the particle terminal settling velocity in a column with finite dimension, u_t to that in an infinite diameter column, $u_{t\infty}$. Initially researchers considered the terminal settling velocity of spherical particles to be independent of the Reynolds number in both the inertial and viscous regimes. Newton¹³⁶ first developed a theoretical relationship based on the forces acting on a settling particle in a vessel with finite dimensions in the inertial flow regime. The wall effect can be estimated by:

$$\frac{u_t}{u_{t\infty}} = (1 - \lambda^2) \sqrt{1 - .5\lambda^2} \quad (4.1)$$

where λ is the particle diameter to column diameter ratio, u_t is the particle terminal settling velocity in a column with finite dimension and $u_{t\infty}$ is the particle terminal settling velocity in an infinite diameter column. For viscous regime, the model proposed by Francis¹³⁷ is:

$$\frac{u_t}{u_{t\infty}} = \left(\frac{1 - \lambda}{1 - 0.475\lambda} \right)^4 \quad (4.2)$$

In later studies, some researchers revealed that the wall effect depends not only on the particle diameter to column diameter ratio, λ but also Reynolds number evaluated at $u_{t\infty}$, $Re_{t\infty}$ ^{1,2,138-143}. In general, the wall effect for spherical particle can be expressed as:

$$\frac{u_t}{u_{t\infty}} = f(Re_{t\infty}, \lambda) \quad (4.3)$$

$$\text{where} \quad Re_{t\infty} = \frac{d_e \rho u_{t\infty}}{\mu} \quad (4.4)$$

where d_e is the volume equivalent diameter of the particle, ρ is the density of the fluid and μ is the viscosity of the fluid. When λ approaches zero, the terminal settling velocity in a finite column, u_t will be the same as that in an infinite diameter column, $u_{t\infty}$. On the other hand, when $\lambda = 1$, the settling velocity becomes zero. More specifically, Fidleris and Withmore¹³⁸ showed that the wall effect is higher at low $Re_{t\infty}$ and high λ . For λ less than 0.1, and $Re_{t\infty}$ larger than 100, the effect becomes negligible. One semi-empirical equation to predict the wall effect on settling of spheres in Newtonian fluid was proposed by Munroe¹⁴¹:

$$\frac{u_t}{u_{t\infty}} = 1 - (\lambda)^{\frac{3}{2}} \quad (4.5)$$

Another relationship to predict the wall effect for any flow regime was proposed by Di Felice²:

$$\frac{u_t}{u_{t\infty}} = \left(\frac{1 - \lambda}{1 - 0.33\lambda} \right)^\alpha \quad (4.6)$$

where the exponential constant, α is a function of Reynolds number and can be expressed by:

$$\frac{0.33 - \alpha}{\alpha - 0.85} = 0.1 Re_{t\infty} \quad (4.7)$$

Kehlenbeck and Di Felice¹ later proposed a relationship for the wall effect, $u_t/u_{t\infty}$ based on their experiments in a Reynolds number range $2 \leq Re_{t\infty} \leq 185$ and diameter ratio $\lambda \leq 0.9$.

Two empirical parameters p and λ_o are adopted to fit the experimental data.

$$\frac{u_t}{u_{t\infty}} = \frac{1 - \lambda^p}{1 + \left(\frac{\lambda}{\lambda_o}\right)^p} \quad (4.8)$$

Both of the empirical parameters, p and λ_o are found to be dependent on Reynolds number, $Re_{t\infty}$ through the following relations:

$$\frac{\lambda_o - .283}{1.2 - \lambda_o} = 0.041 Re_{t\infty}^{0.524} \quad (4.9)$$

$$\text{for } Re_{t\infty} \leq 35, \quad p = 1.44 + 0.5466 Re_{t\infty}^{0.434} \quad (4.10)$$

$$\text{for } Re_{t\infty} \geq 35, \quad p = 2.3 + 37.3 Re_{t\infty}^{-0.8686} \quad (4.11)$$

Compare to studies on spherical particles, there are only a handful of studies available for the settling of non-spherical particles. Most researchers either do not consider the wall effects¹⁴⁴⁻¹⁴⁶ or simply use the same corrections developed for spherical particles for the settling of non-spherical particles^{133,147,148}. However, Kasper et. al.¹⁴⁹ found that models for spherical particles are unable to predict the settling of cylindrical particles with aspect ratios larger than 2 with a Reynolds number less than 0.015. Chhabra^{132,134} later developed an empirical correlation for the settling of cylindrical particles along with some other shapes in Newtonian liquids and non-Newtonian polymer solutions in viscous flow regime which can be represented as,

$$\frac{u_t}{u_{t\infty}} = 1 - A'\lambda \quad (4.12)$$

where A' is an empirical coefficient related to the particle shapes. A' was determined empirically as 1.33 and 3.58 for cylindrical particles with aspect ratio ≤ 10 and for aspect

ratio ≥ 10 , respectively in the viscous regime. A different set of values were found to predict the wall effects of different shape particles in these fluids¹³².

The available studies of wall effect on cylindrical particles only covered a very narrow range of Reynolds number, in which no dependence of wall effect on Reynolds number was found. In this work, the wall effect on the settling of cylindrical particles with an aspect ratio range of 3-20.7 is experimentally investigated in cylindrical columns in the inertial regime with Re_{∞} ranges from 700-2600 and for λ ranges from 0.01-0.54.

4.2 Experimental

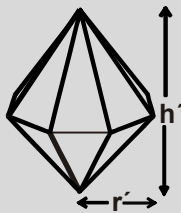
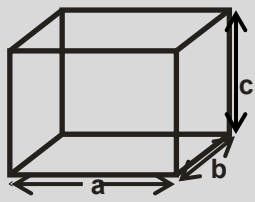
Settling velocities of various particles are investigated in cylindrical columns of various diameters. The schematic diagram of the experimental setup is shown in Figure 4.1. Each column is made of acrylic and 1 meter in height. The dimensions of the columns are listed in Table 4.1.

Table 4.1 Dimensions of the columns used in this study

Column No.	1	2	3	4	5	6	7	8
Inner dia.(mm)	6	10	16	20	26	42	52	70
Outer dia.(mm)	10	15	20	30	30	52	62	80

Most of the experiments are performed using cylindrical particles with an aspect ratio range of 3-20.7 while some bicone and cube shape particles are also used. The dimensions of particles used are listed in Table 4.2.

Table 4.2 Dimensions of the various particles employed in this experiment

Material	Shape	Density (gm/cm ³)	Aspect ratio, l/d(-)	Dimension (mm)		Volume equivalent diameter, d _e (mm)	Spheri city, ψ(-)
Copper	 Cylinder	7.82	4	d=0.8 7	l=3.5	1.58	0.729
			5		l=4.35	1.70	0.694
			7		l=6.09	1.90	0.636
			8		l=7	2.00	0.618
			11.5		l=10	2.24	0.579
			13.8		l=12	2.38	0.524
			17.2		l=15	2.57	0.492
			20.7		l=18	2.73	0.465
			3	d=2	l=6	3.30	0.778
			3.5		l=7	3.48	0.757
			5		l=10	3.91	0.698
			6		l=12	4.16	0.666
			7		l=14	4.38	0.617
			8		l=16	4.58	0.581
			10		l=20	4.93	0.549
Glass	 Bicone	2.28	-	r=2.06, h=3.6		3.12	0.864
	 Cube			a=4, b=3.83, c=3.9		4.85	0.81

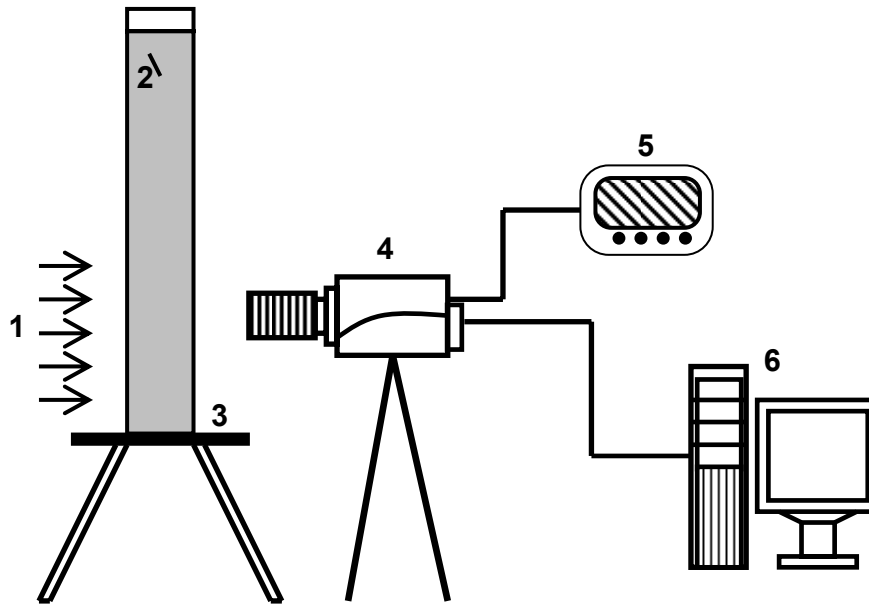


Figure 4.1 Schematic diagram of the experimental apparatus: (1) light source; (2) particle; (3) settling column; (4) high speed camera; (5) monitor; (6) computer.

Water is used as the settling medium. Particles are dropped at the centre of the column just under the liquid surface to create minimum disturbance of the liquid medium. The particle settling motion is recorded by a high-speed camera at a frame rate of 500 Hz. The settling velocity is determined based on the video captured at the bottom of the column to ensure that the steady settling of particle is reached. Experiments show that most particles attain the steady settling velocity within 200 mm.

Volume equivalent diameter, d_e of the non-spherical particles is commonly used for the calculation of particle to column diameter ratio.^{132,134} Particle settling velocity in an infinite column, $u_{t\infty}$ is obtained by extending the plot of measured velocity u_t and diameter ratio λ ($=d_e/D$, here D is the column diameter) line to $\lambda = 0$. This extrapolation method is completely valid for all the shape of particles because for large diameter columns when d_e/D tends to 0, the particle settling orientation does not change

significantly with further increase in column diameter. This procedure has been used to determine the settling velocity in the absence of wall effect^{132,134,142}. Multiple measurements of settling velocity for each particle are taken to ensure the repeatability of results.

4.3 Results and discussion

4.3.1 Experimental results

Cylindrical particles have the characteristics of having different sphericity with different aspect ratio, l/d . The sphericity of cylindrical particles can change from 0.465 to 0.778 when the aspect ratios change from 3-20.7. Figure 4.2 shows the $u_t/u_{t\infty}$ values for cylindrical particles with aspect ratio of 7, 11.5, 17.2 and 20.7 as a function of λ . It can be seen that at low λ , $u_t/u_{t\infty}$ decreases with an increase in λ . This behavior agrees well with the general understanding of wall effect. However, when λ is larger a critical value, λ_c , depending on the aspect ratio of the particle, an increase in $u_t/u_{t\infty}$ is observed with an increase in λ . With the change of λ , the settling velocity and Reynolds number changes for the particles. Experimental cylindrical particles covers the range of 700~2600.

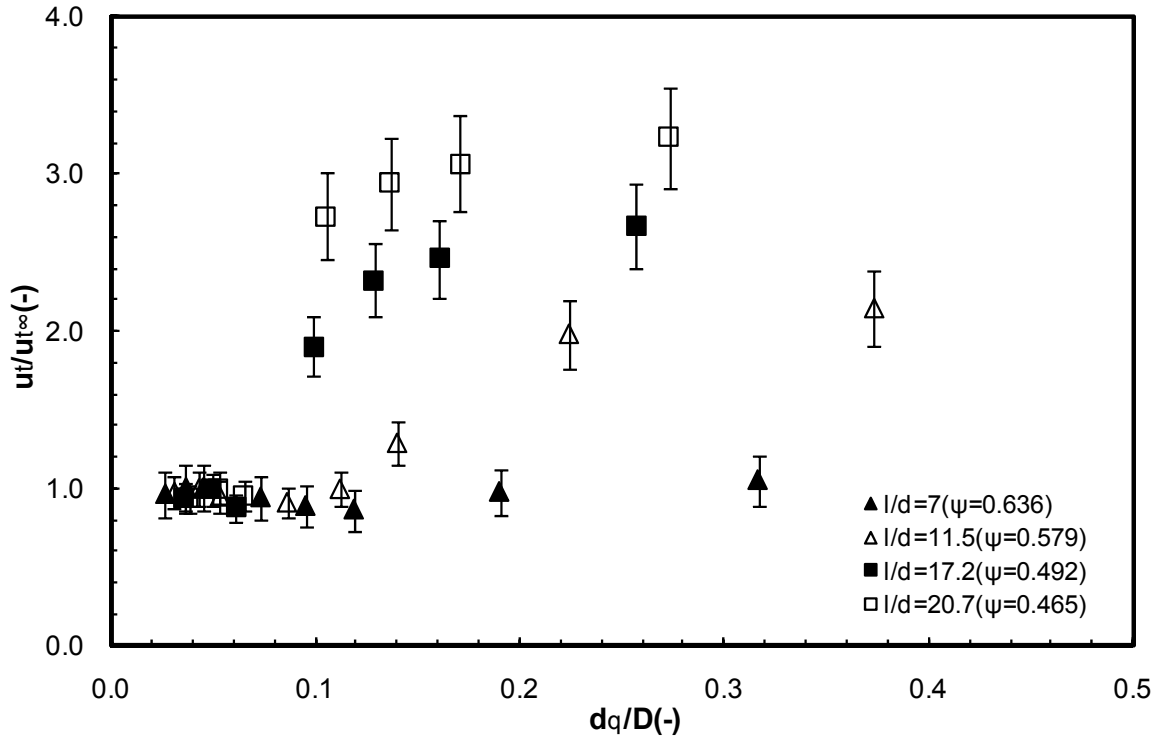


Figure 4.2 Experimental $u_t/u_{t\infty}$ results for copper cylindrical particles with different aspect ratio with a $Re_{t\infty}$ range 790-940. (Error bars include the percentage error)

It can also be seen that $u_t/u_{t\infty}$ is larger for particles with higher aspect ratios when λ is larger than λ_c . For example, at $\lambda = 0.2$, $u_t/u_{t\infty}$ of cylindrical particle with l/d of 20.7 is around 3 while $u_t/u_{t\infty}$ of cylindrical particle with aspect ratio of 11.5 is only around 1.8. The rate of increase in $u_t/u_{t\infty}$ at λ larger than λ_c is also more apparent for particles with higher aspect ratio. A comparison of λ_c for copper cylindrical particles with different aspect ratio is shown in Figure 4.3. Particles with a higher aspect ratio are found to have a smaller λ_c compared with those with a lower aspect ratio.

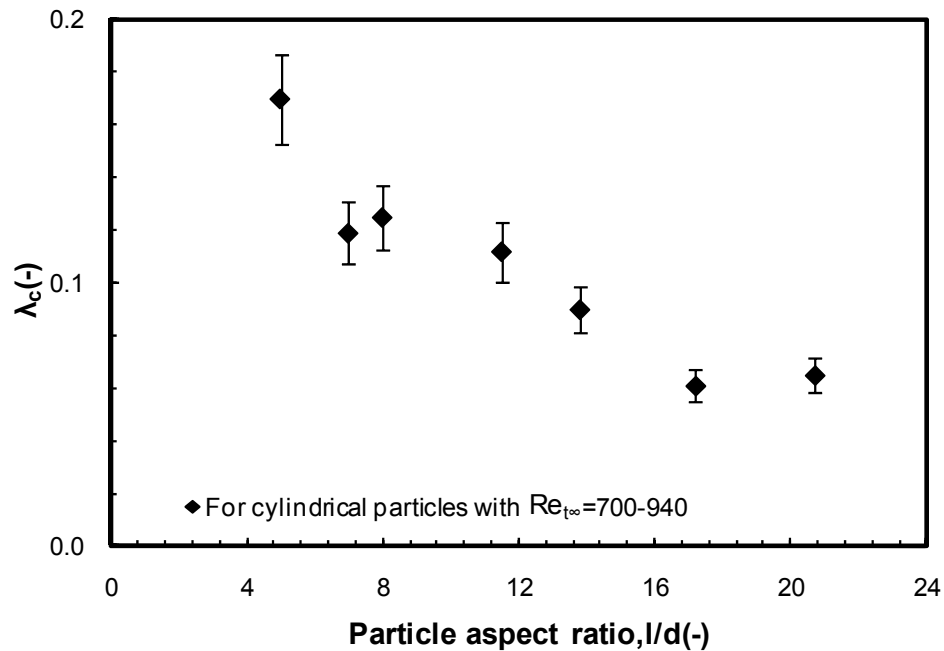


Figure 4.3 Critical λ values for particles with different aspect ratio in water with a $Re_{t\infty}$ range of 700-940. (Error bars include the percentage error)

Generally, cylindrical particles settle with a horizontal orientation as shown in Figure 4.4.¹⁵⁰ An experimental settling study of a cylindrical particle in a rectangular column with Length $\times$ Width $\times$ Height = 0.475 m $\times$ 0.48 m $\times$ 1.5 m (tube was half full of water) confirms the settling behaviour. However, even with negligible wall effect translational and rotational motions are tracked for the particles. When settling with this horizontal orientation, the terminal settling velocity decreases with an increase in λ . When a particle is settling in a small diameter column, it experiences higher viscous dissipation due to the wall compared to that in a large column. This increase causes a reduction in the particle terminal settling velocity. Substantial collisions of particles with the column wall also contribute to the reduction of particle settling velocity. As λ increases, the viscous dissipation increases. However, the viscous dissipation is not

uniform across a particle. Higher viscous dissipation is experienced on the end of the particle that is closer to the wall, resulting in a lower velocity than the end further from the wall. The difference in velocities between the two ends rotates the particle to an inclined orientation. The particle then settles with this inclined orientation in a helical motion as illustrated in Figure 4.5. The inclined orientation of the particles results in a lower drag due to a smaller projected area than the horizontal orientation. Therefore, the settling velocity of a particle is higher in an inclined orientation. Since the degree of inclination of the particle increases with λ , $u_t/u_{t\infty}$ also increases with λ as observed in the experiments.

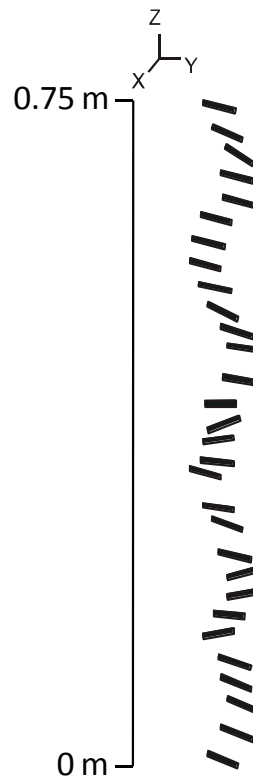


Figure 4.4 The trajectory and orientation of a settling cylindrical particle observed in stagnant water without any wall effect.¹⁵⁰

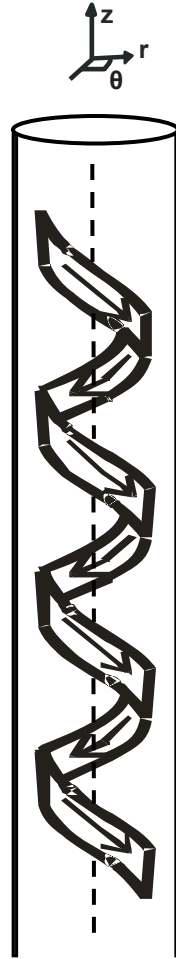


Figure 4.5 Schematic of the trajectory of cylindrical particles in helical motion in a cylindrical column.

The effect of initial particle orientation on settling is also investigated. When λ is smaller than λ_c , regardless of the initial dropping orientation, cylindrical particles always settle with a horizontal orientation with minor swing and rotation motion. However, when λ is larger than the critical λ_c , any initial dropping orientation will result in an inclined orientation and settle with a helical motion in the experimental Reynolds number range. The formation of the helical motion can be seen in Figure 4.6 where a cylindrical particle is dropped with an inclined orientation and is able to attain the helical motion within 0.29 sec.

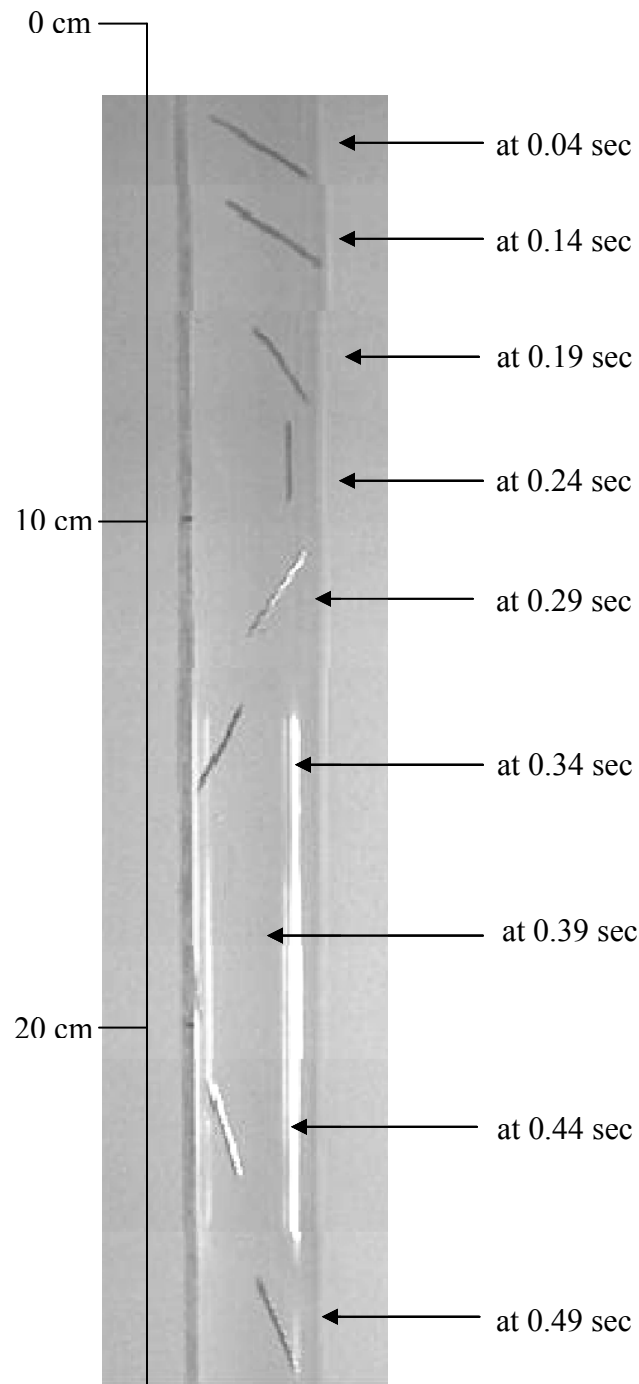
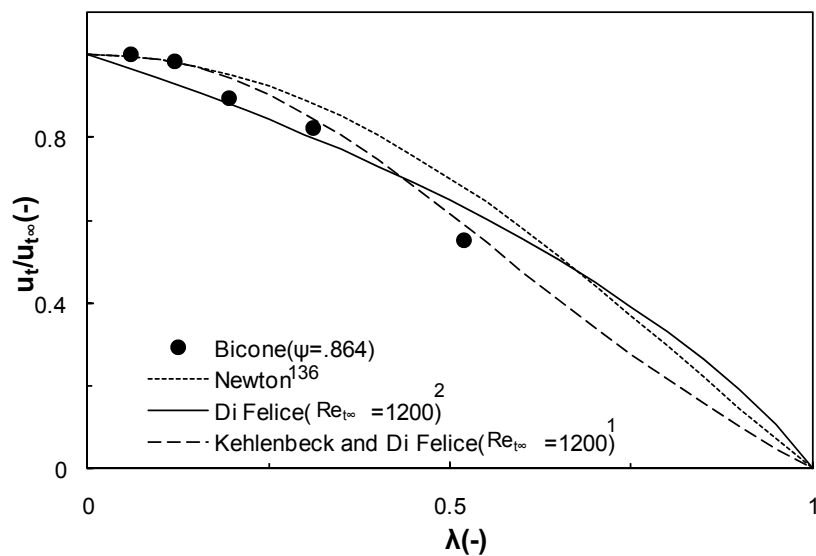


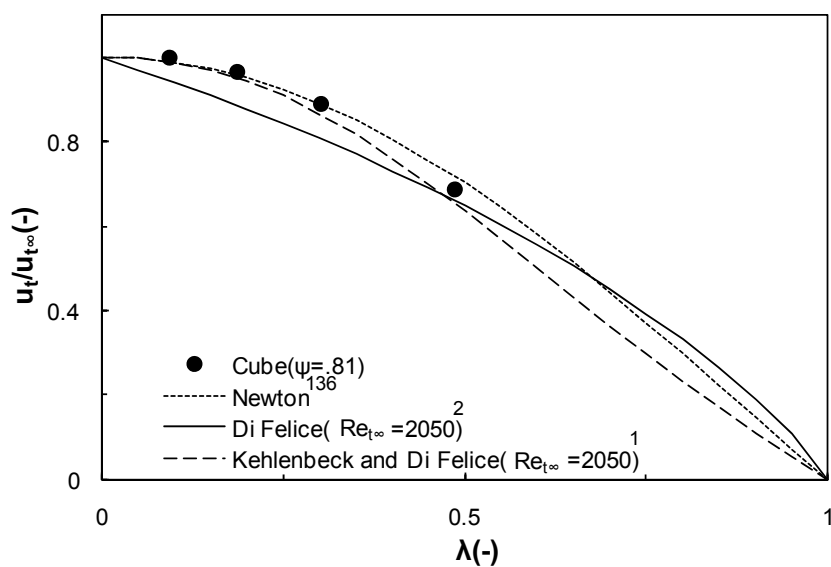
Figure 4.6 Formation of the vortical settling motion of a cylindrical particle.

Settling of bicone and cubic particles is also studied and the results are shown in Figure 4.7. No increasing trend of $u_t/u_{t\infty}$ with λ is observed for both the bicone and cubic shape particles, which have relatively high sphericity of 0.864 and 0.810 respectively.

Kehlenbeck and Di Felice¹ model, which was developed for spherical particles, is found to give good prediction for both bicone and cube particles used in this study.



(a)



(b)

Figure 4.7 Wall effect on the settling of non-spherical particles (a) bicone particle with a sphericity of 0.864 and Reynolds number of 1200. (b) cube particle with a sphericity of 0.81 and Reynolds number of 2050.

4.5 Conclusion

The settling of single non-spherical particle with various sphericities in cylindrical column is investigated in the inertial flow regime. It is observed that the presence of wall has strong impact on the settling velocities. The wall effect is quantified in terms of $u_t/u_{t\infty}$ ratio. The available models for spherical particles fail to predict $u_t/u_{t\infty}$ values for cylindrical particles with high aspect ratios in the experimental Reynolds number range. A decreasing trend of $u_t/u_{t\infty}$ is observed with an increase in the equivalent particle diameter to column diameter ratio, λ . However, when λ is greater than λ_c , an increasing trend of $u_t/u_{t\infty}$ is noticed for high elongated particles. At $\lambda > \lambda_c$, the settling particles change to an inclined orientation, resulting in a helical motion with a higher terminal settling velocity compare to the settling in the absence of wall effect.

Chapter 5 EFFECT OF PARTICLE SHAPE ON DRY PARTICLE INHALATION: STUDY OF FLOWABILITY, AEROSOLIZATION AND DEPOSITION PROPERTIES

5.1 Introduction

Good flowability and aerosolization capability are two basic needs for inhalation aerosols.¹³¹ Particles with an aerodynamic diameter (d_a) range of 1-5 μm were found to have the highest delivery efficiency in inhalation studies. Particles of this size range have severe effect on powder flow and aerosolization behavior. Theoretically, non-spherical particles can give rise to distinct drag forces and terminal settling velocities, which would in turn affect the aerodynamic behavior of the particles.^{29,58} However, previous studies were mostly focused on spherical particles in dry powder formulations. Therefore, shape effect of micronized particles on their flowability and *in vitro* aerosolization capacity and corresponding *in vitro* deposition behavior are investigated in this chapter.

A shape factor is normally used for non-spherical particles in the definition of aerodynamic diameter. Aerodynamic diameter is a general characterizing parameter for aerosol particles, defined as the diameter of a sphere of unit density that has the same terminal settling velocity as the particle under consideration, $d_a = d_g \sqrt{\frac{\rho_e}{S\rho_s}}$ where $\rho_s = 1 \text{ g/cm}^3$, d_g is the particle geometric diameter, ρ_e is the effective particle density in the same

unit as ρ_s and S is the dynamic shape factor of the particle.⁵⁸ This shape factor is defined as the ratio of the drag force of the particle to that of a sphere of equivalent volume.^{52,151} Increasing particle surface roughness would attribute to lower aerodynamic diameter.¹⁵² With a reduced aerodynamic diameter, particles would have a higher possibility of reaching the deep lung as compared with spherical particles. Indeed, researchers found that elongated particles exhibit better aerodynamic behavior than spherical particles.^{62,102,153} Elongated or needle-like particles are reported to have longer suspended time in the air and can travel further in the lung airway.¹⁰² However, high elongated particles also experience high interception deposition as demonstrated by Balásházy et al.⁶⁴

Particle aerosolization is another important factor for deep lung delivery. Aerosolization of particles from the inhaler depends on the particle-particle and particle-wall interaction. Particle interaction is closely related to the van der Waals force, which is a function of the particle surface morphology,^{65,154} size,¹⁵⁵ shape,^{65,156} electrostatic properties,¹⁵⁷ and hygroscopicity.¹⁵⁸ Particle shape that has low contact area and van der Waals force will have lower aggregation tendency and can be dispersed well in the air.¹⁰⁷ Elongated particles are not suitable for aerosolization due to their large attractive forces.⁶⁵

Most of the studies are focused on spherical particles and a few on elongated particles. It has been shown that particle shape has an important effect on dry powder inhalation.¹⁵⁹ However, The use of shape factor is also not sufficient to represent the effect of shape on the particle aerodynamic behavior as it also depends on the particle orientation and the contact area with other particles. Therefore, the understanding of the shape effect on these properties needs to be investigated properly to improve particle

aerosolization and minimize early depositions due to interception, inertial impaction and gravitational deposition. A preliminary study found that a pollen-shape characteristic of particles can enhance the flowability, aerosolization and deposition properties compared to particles having similar volume equivalent diameter.^{159,160} However, the particles' characteristic size and aerodynamic diameter are also an important factor for the aerodynamic behavior. Therefore, the particle shape effect on the flowability, aerosolization and deposition behavior is investigated in this study with the introduction of additional particles with different size and shapes. The particle shapes studied include sphere, plate, cube, needle, and pollen.

5.2 Experimental

5.2.1 Preparation

Pollen-shape and spherical hydroxyapatite (HA, $\text{Ca}_5(\text{PO}_4)_3(\text{OH})$) particles are synthesized using potassium dihydrogen phosphate (KH_2PO_4 , Sigma-Aldrich), calcium nitrate tetrahydrate ($\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, Sigma-Aldrich), poly(sodium-4-styrene-sulfonate) (PSS, Sigma-Aldrich), and urea (Sigma-Aldrich).¹²⁵ 30 ml of KH_2PO_4 (0.02 M) solution is mixed with 50 ml of $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ (0.02 M) solution. PSS and urea are then added to the mixture. The amount of PSS and urea added govern the shape and size of the product HA particles. In this study, the PSS concentration used ranges from 40-80 g/L and urea concentration used ranges from 3-7.5 M. The final mixture is stirred to completely dissolve the urea. The solution mixture is then kept in an oven at 200°C for 3-6 hours to allow sufficient time for hydrothermal reaction. Finally, the precipitated product is filtered, washed and then dried at 70°C.¹⁵⁹

Spherical calcium carbonate (CaCO_3) particles are produced by using $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, sodium carbonate (Na_2CO_3 , Kanto Chemical) and PSS.¹²⁶ Equal volume of 0.1 M $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ and 0.1 M Na_2CO_3 solution are mixed in a solution with PSS concentration of 50 g/L at room temperature. The CaCO_3 precipitate is then filtered, washed, and dried at 70 °C.

Plate-shape calcium oxalate (CaC_2O_4) particles are produced by precipitation reaction of sodium oxalate ($\text{Na}_2\text{C}_2\text{O}_4$, Kanto Chemical, Singapore) and calcium chloride (CaCl_2 , Kanto Chemical, Singapore) using PSMA (poly-(styrene-alt-maleic acid), Sigma-Aldrich, Singapore) as the surfactant.¹²⁷ 0.8 ml of $\text{Na}_2\text{C}_2\text{O}_4$ (0.1 M) solution is added to 80 ml aqueous solution of PSMA (0.5 g/L). Dilute HCl (Fluka, Singapore) is added to achieve a pH of 4. 0.8 ml of CaCl_2 (0.1 M) is then added and the mixture is kept at 25 °C under static condition for 24 hours. The product is then collected, washed and dried at 70 °C.

Cube-shape CaCO_3 is produced by precipitation reaction of Na_2CO_3 and CaCl_2 using CTAB (Cetyl trimethylammonium bromide) as the surfactant.¹²⁸ 1.28 ml of Na_2CO_3 (0.5 M) solution is added to 80 ml aqueous solution of CTAB (1 g/L). NaOH (Fluka, Singapore) is added to obtain a pH of 10. 1.28 ml of CaCl_2 (0.5 M) solution is added and the mixture is kept at 80 °C under static condition for 24 hour before the final product is collected. After washing the product is dried at 70 °C.

Needle-shape CaCO_3 particles are synthesized by precipitation reaction of potassium hydrogen carbonate (KHCO_3 , Merck, Singapore) and CaCl_2 .¹²⁹ Equal volume of 0.1 M KHCO_3 and CaCl_2 are heated till boiling separately. The two solutions are then

mixed together to obtain the carbonate precipitate. The precipitate is then filtered, washed, and dried at 110 °C.

Particles of different materials could have difference in flowability, aerosolization and deposition properties. These properties of particles highly depend on van der Waals forces between the particles. The van der Waals force between two equal size spherical particles can be calculated by the equation:⁶⁵ $F_{\text{tot}} = \frac{AR}{12H^2} + \frac{A}{6\pi H^3} \pi r^2$ where R is the diameter of the spherical particle, A is the Hamaker constant, H is the separation distance between two particle surfaces, and r is the radius of the contact area. Based on equation mentioned above, the van der Waals force, F is proportional to the first order of the Hamaker constant, A but inversely proportional to between second and third order of the separation distance, H . Vissar⁶⁵ also showed that the separation distance have a stronger effect on the van der Waals force than the Hamaker constant. Therefore, even though particles of different materials are used in this study, the difference in flowability, aerosolization and deposition properties can be considered to be solely dependent on the particle physical properties (size, shape, and density).

5.2.2 Particle Characterization

Synthesized powder samples are characterized by measuring their mean particle diameter, shape, bulk (ρ_{bulk}) & tap (ρ_{tap}) densities and powder flowability. The flowability of the powder samples analyzed by Carr's compressibility index (CI) and angle of slide (θ).

5.2.3 *In vitro* aerosolization and deposition properties

The Andersen cascade experiment is carried out at an air flow rate of 30 L/min. 20 mg of each sample is loaded into a hard gelatin capsule (Gelatin Embedding Capsules,

size 4, 0.25 cc, Polysciences Inc.) manually. A capsule filled with particles is loaded into a Rotahaler® (Glaxo) which is used as the inhaler to aerosolize the particles. An actuation time of 20 seconds is allowed for each capsule to completely disperse all the particles. The extracted solutions with deposited samples from different regions are then quantified with inductively coupled plasma (ICP, Optica 2000 DV) (for Ca^{2+} element). Each experiment is repeated at least three times.

5.3 Results and discussion

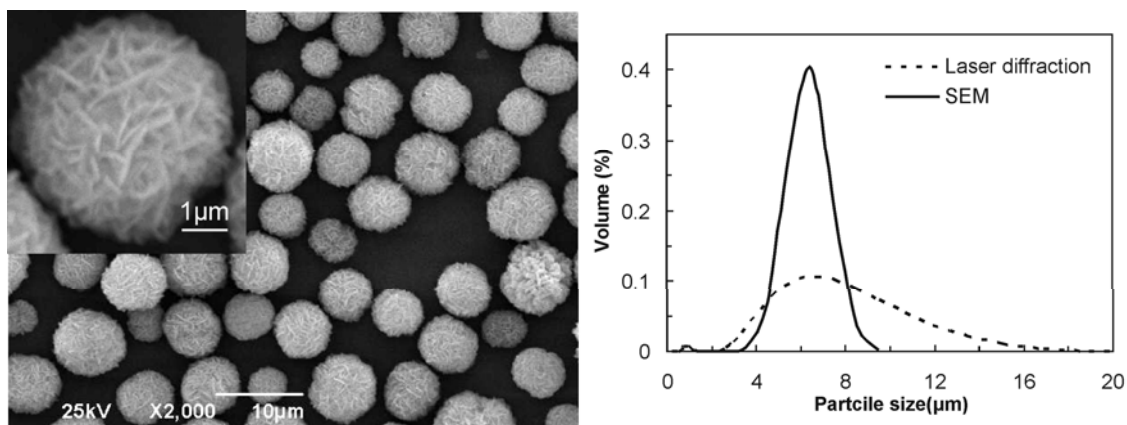
5.3.1 Particle characteristics

The SEM images and size distributions of the different particle samples used in this study are shown in Figure 5.1. Particle size measurement of the non-spherical particles is always a challenge especially with the use of one single parameter. Two most common methods for particle size measurement, laser diffraction measurement and direct visualization from microscopy images are used in this study. The laser diffraction measurement takes into account the lights diffracted from the dispersed particles to estimate the particle size. However, the diffracted lights can change with the orientation of non-spherical particles and the results may not be a good representation of the actual size. Therefore particle size obtained from SEM images is also used for comparison with laser diffraction measurement.

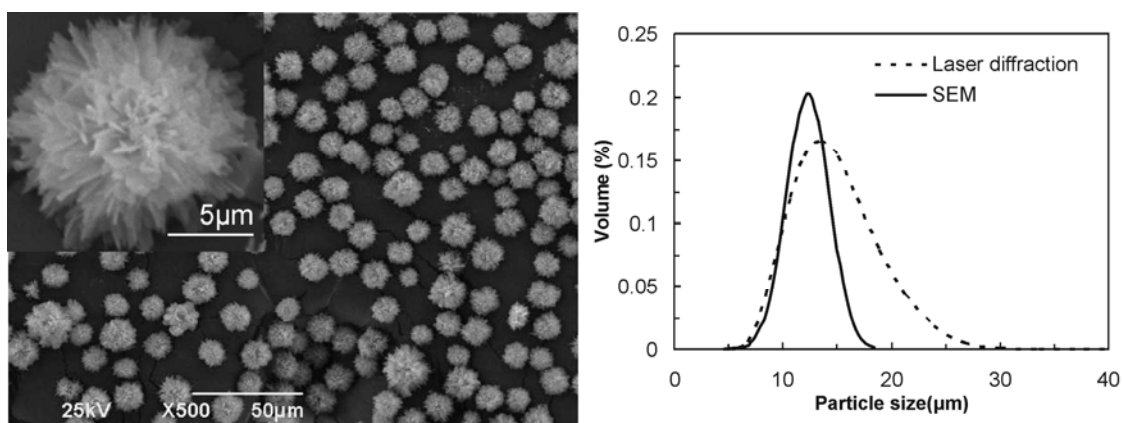
It is to note that the size distributions obtained from laser diffraction measurement are volume-weighted while the size distributions from the SEM images are number-weighted. Therefore, the SEM size distributions are converted to volume-weighted distribution before comparisons are made. The conversion of number-weighted size distribution to volume-weighted size distribution requires the knowledge of particle

volume. Therefore, pollen-shape particle volume is assumed to be the same as a sphere with the same diameter. Plate-shape particle volume is assumed to be the same as a particle that has the average width and thickness from SEM images. Volume of cube-shape particle is calculated as the cube of the side-length. Needle-shape particle volume is assumed to be the same as a cylinder with the same length and average diameter from SEM images. The physical properties of the particles are listed in the Table 5.1.

Pollen-shape particles are selected for the surface morphology that increase the separation distance and may lower interaction force and eventually may have high potential for better dispersion and aerosolization. Two types of pollen-shape HA particles are produced with distinct size to investigate whether the shape effect applies for different size range. The HA particles synthesized with PSS concentration of 40 g/L and urea concentration of 7.5M (Figure 5.1(a), pollen-shape I) has a petal like surface structure and a $d(50\%, \text{SEM})$ of 6.3 μm . The HA particles synthesized with PSS concentration of 40 g/L and urea concentration of 3M (Figure 5.1(b), pollen-shape II) has a fibrous surface structure and a $d(50\%, \text{SEM})$ of 12.5 μm . As shown in Figure 5.1(a) & 5.1(b), both laser diffraction and SEM measured size distributions yield similar mode but the laser diffraction measurement consistently gives a larger size distribution than the SEM measurement. This maybe because the surface irregularity of both pollen-shape HA particles cause additional diffraction of the light beam in laser diffraction measurement and yield a larger measurement size than the actual size. Therefore, the SEM image measurement results are used as the characteristic size.

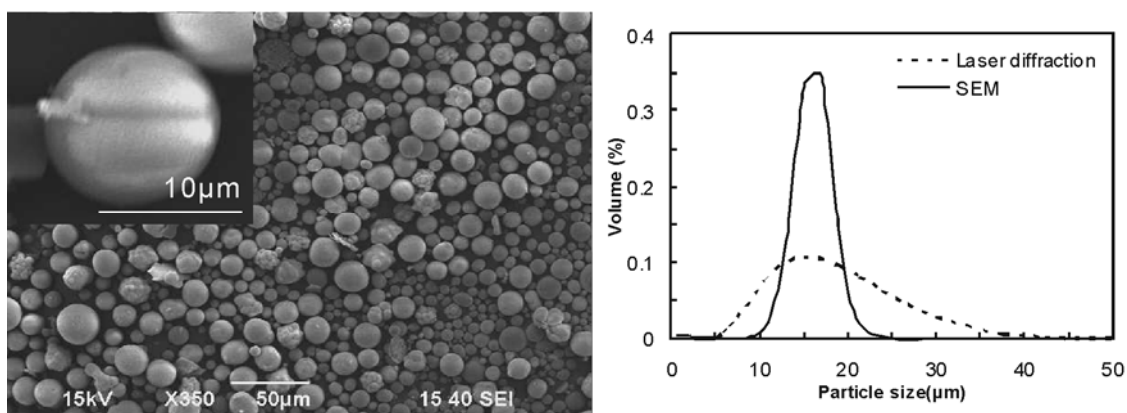


(a)

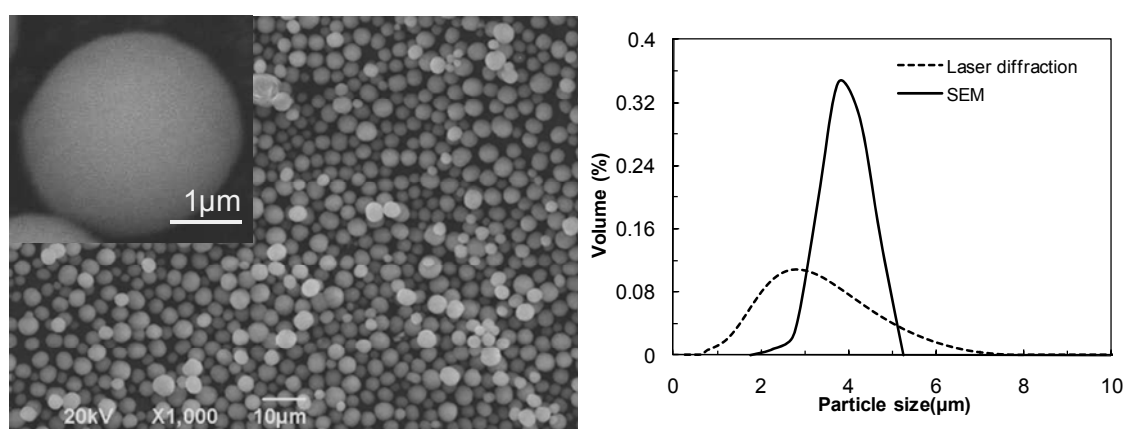


(b)

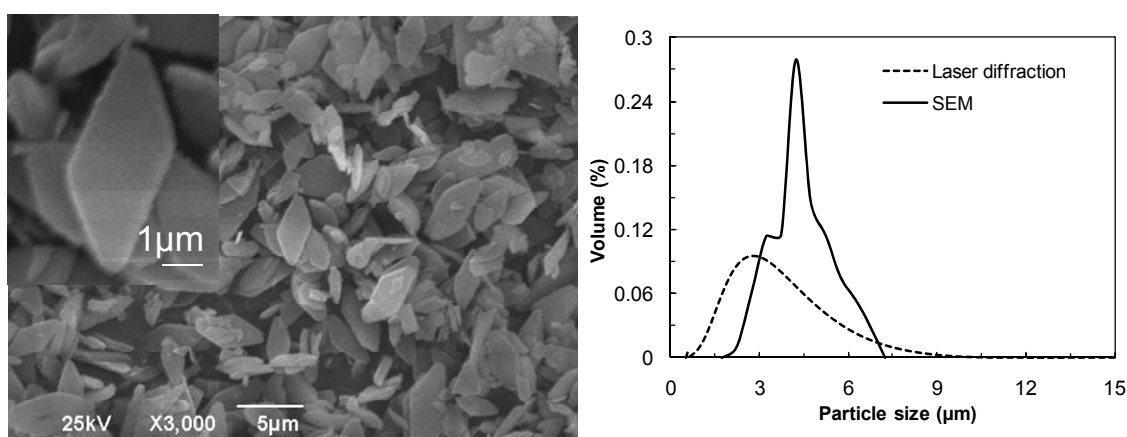
Figure 5.1 SEM image and size distribution of (a) pollen-shape I HA particles, (b) pollen-shape II HA particles; (c) spherical I CaCO_3 particles; (d) spherical II HA particles; (e) plate-shape CaC_2O_4 particles; (f) cube-shape CaCO_3 particles; (g) needle-shape CaCO_3 particles.



(c)

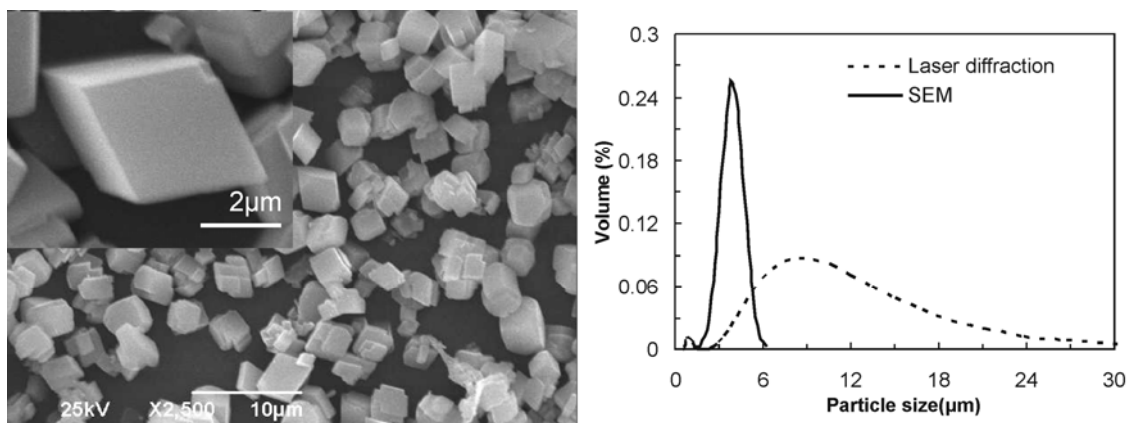


(d)

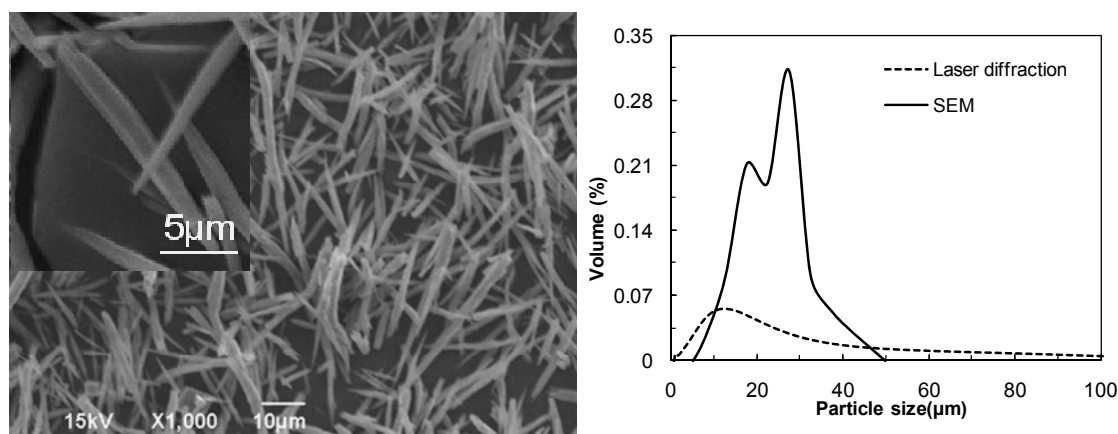


(e)

Figure 5.1 Cont.



(f)



(g)

Figure 5.1 Cont.

The size of spherical particles does not vary with the particle orientation. The size distribution of the spherical particles obtained by laser diffraction is indeed similar to the size obtained from SEM images shown in Figure 5.1(c) & 5.1(d). Spherical CaCO_3 (spherical I) sample in Figure 5.1(c) has a $d(50\%, \text{SEM})$ of 3.7 μm and a $d(50\%, \text{LD})$ of 3.9. The larger spherical HA sample (spherical II), produced with PSS concentration of 80 g/L and urea concentration of 7.5 M, has a $d(50\%, \text{SEM})$ of 15 μm and a $d(50\%, \text{LD})$ of 14.9 μm. Both SEM and LD measurements give similar $d(50\%)$ for the spherical

particles. Since LD takes a larger sample size into consideration, LD measurement size is taken as the characteristic size.

The plate-shape particles can be described by three dimensions, length (maximum dimension), width (intermediate dimension), and height (minimum dimension). The SEM image and size distributions of the plate-shape particles are shown in Figure 5.1(e). Since the laser diffraction measurement depends on the particle orientation, it would be difficult to judge the true representation of the laser diffraction measured size distribution. Therefore, it is more appropriate to use the length as the characteristic size of plate-shape particles. The characteristic size distribution is mastered based on the volume of the particles. The average values of the width and height obtained from the SEM images are used to calculate the volume of the particles with various lengths. The median of this distribution volume against the length of the particles are termed as $d(50\%, \text{SEM})$ of the length as the characteristic size of plate-shape particles. The calculation of the aerodynamic diameter requires the volume equivalent diameter which is estimated from the dimensions obtained from SEM measurements.

The SEM image and size distributions of the cube-shape CaCO_3 particles are shown in Figure 5.1(f). The laser diffraction measurement yields a $d(50\%)$ of $8.5 \mu\text{m}$ while SEM measurement shows that the cubes has a side length of $3.7 \mu\text{m}$. The SEM image shows that the cube-shape particles are clustered together. In this case, the laser diffraction would give a better measurement of the average cluster size than the side length obtained from SEM measurement. Therefore, the size obtained from the laser diffraction is used as the characteristics size and volume equivalent diameter for cube-shape particles.

The SEM image and size distributions of needle-shape CaCO_3 particles are shown in Figure 5.1(g). $d(50\%)$ of the needle-shape CaCO_3 particles from laser diffraction is $11.6\ \mu\text{m}$. It is obvious from the SEM image that most of the particles have a length of over $20\ \mu\text{m}$ and a diameter of around $2\ \mu\text{m}$. In this case, it is quite possible that the laser diffraction measurement result comprises measurement of both the length and diameter of the needle-shape particle. Similar technique of the plate-shape particles is adopted for needle-shape particles to measure the characteristic length size distribution. The $d(50\%, \text{SEM})$ of $24.1\ \mu\text{m}$ is found as the characteristics size of the needle-shape particles. Aerodynamic diameter is estimated from the dimensions from SEM measurements. The characteristic sizes of different shape particles are listed in Table 5.2.

Table 5.1 Properties of different shape particles

Shape	$d(50\%, \text{LD})$ (μm)	$d(50\%, \text{SEM})$ (μm)	ρ_{bulk} (g/cm^3) ($n=0$)	ρ_{tap} (g/cm^3) ($n=2500$)	CI Index (%)	Angle of slide, θ ($^\circ$)
Pollen-shape (I)	6.7	6.3	0.35	0.72	51.8 ± 3.6	35.3 ± 2.3
Pollen-shape (II)	13.4	12.5	0.23	0.40	42.2 ± 2.9	34.7 ± 0.6
Spherical I	3.9	3.7	0.58	1.04	44.0 ± 3.1	65.0 ± 3.0
Spherical II	14.9	15	1.05	1.13	19.2 ± 1.3	49.3 ± 2.6
Plate-shape	2.8	4.2 (length) 2.0 (width) 0.5(thickness)	0.39	0.66	40.6 ± 2.8	51.0 ± 6.1
Cube-shape	8.5	3.7	0.29	0.62	53.6 ± 3.8	55.7 ± 5.5
Needle-shape	11.6	24.1(length) 1.9(diameter)	0.28	0.46	39.9 ± 2.8	40.7 ± 1.5

Table 5.2 Characteristic size, shape factor and aerodynamic diameter of different shape particles.

Shape	Characteristics size (μm)	Shape factor, S (-)	Aerodynamic diameter, d_a (μm)
Pollen-shape I	6.3	1.2	4.1
Pollen-shape II	12.5	1.2	5.9
Spherical I	3.9	1.0	3.8
Spherical II	14.9	1.0	15.9
Plate-shape	4.2	1.5	1.4
Cube-shape	8.5	1.3	3.2
Needle-shape	24.1	1.7	2.7

5.3.2 Shape factor of the different shape particles

The determination of the aerodynamic diameter requires the dynamic shape factor of the particles, which is a function of the drag force experienced by the moving particles. The dynamic shape factor for sphere is defined as 1. However, for irregular shape particles, it is difficult to measure the dynamic shape factor. Therefore, the shape factors of the particles are estimated based on Davis,¹⁶¹ in which shape factors for different shape particles extensively reviewed.

A needle shape can be viewed as a prolate spheroid (polar diameter greater than equatorial diameter) with a very high aspect ratio. The shape factors of prolate spheroids were reported to be between 1-15.43 for aspect ratios range from 1-1000.¹⁶¹ An increase in the aspect ratio would increase the shape factor. The needle-shape particles used in this study has an average aspect ratio of 12.7 (average length 24.1 μm and average width 1.9 μm). Thus, the average dynamic shape factor is estimated to be 1.7 by interpolation.

The plate-shape particles, on the other hand, can be considered as an oblate spheroid (polar diameter shorter than equatorial diameter). The dynamic shape factor of an oblate spheroid with an aspect ratio of 10 is reported to be 1.49.¹⁶¹ Since the aspect ratio of the plate-shape particles used is around 10, a dynamic shape factor of 1.5 can be estimated.

For the cube-shape particles, SEM images indicate that the particles are clustered together and form rock-shape clusters. The dynamic shape factor of rock-like quartz particles are found to have a shape factor of 1.36.¹⁶¹ From the SEM image it can be seen that these particles are closer to spherical shape than the rock-like quartz particles. Thus, these cube-shape clusters are estimated to have a shape factor of 1.3.

The pollen-shape particles closely resemble a spherical shape. The rough surface would cause a slightly higher shape factor than spherical particles. Thus, a dynamic shape factor of 1.2 is estimated for the pollen-shape particles.

It is also to note that the dynamic shape factors are to be used in the calculation of aerodynamic diameter. While the aerodynamic diameter is proportional to the square root of the dynamic shape factor, the error in the estimation of dynamic shape factor should have a relatively small impact on the accuracy of the aerodynamic diameter calculated. The calculated aerodynamic diameters for the particles are also shown in Table 5.2.

5.3.3 Flowability, aerosolization and deposition properties

CI, θ , ED and FPF results of the different shape particles are plotted as a function of their aerodynamic diameter and characteristic size in Figure 5.2(a) and 5.2(b) respectively. The aerodynamic diameter of the particles ranges from 1.4-15.8 μm with a majority of particles in the range of 1.4-5.9 μm . The different shape particles have their

characteristic size ranging from 3.9-24.1 μm . The flowability, aerosolization and deposition properties show no evidence of direct relation with the aerodynamic diameter as well as the particle characteristic size. It can be seen that the increase in ED does not follow any trend. Particle with large d_a showed poor FPF, but FPF does not increase regularly with the decrease of d_a . Similar to the aerodynamic diameter, characteristic size also cannot correlate the particles' behavior. The results signify that the flow, aerosolization and deposition characteristics are function of another factor which is

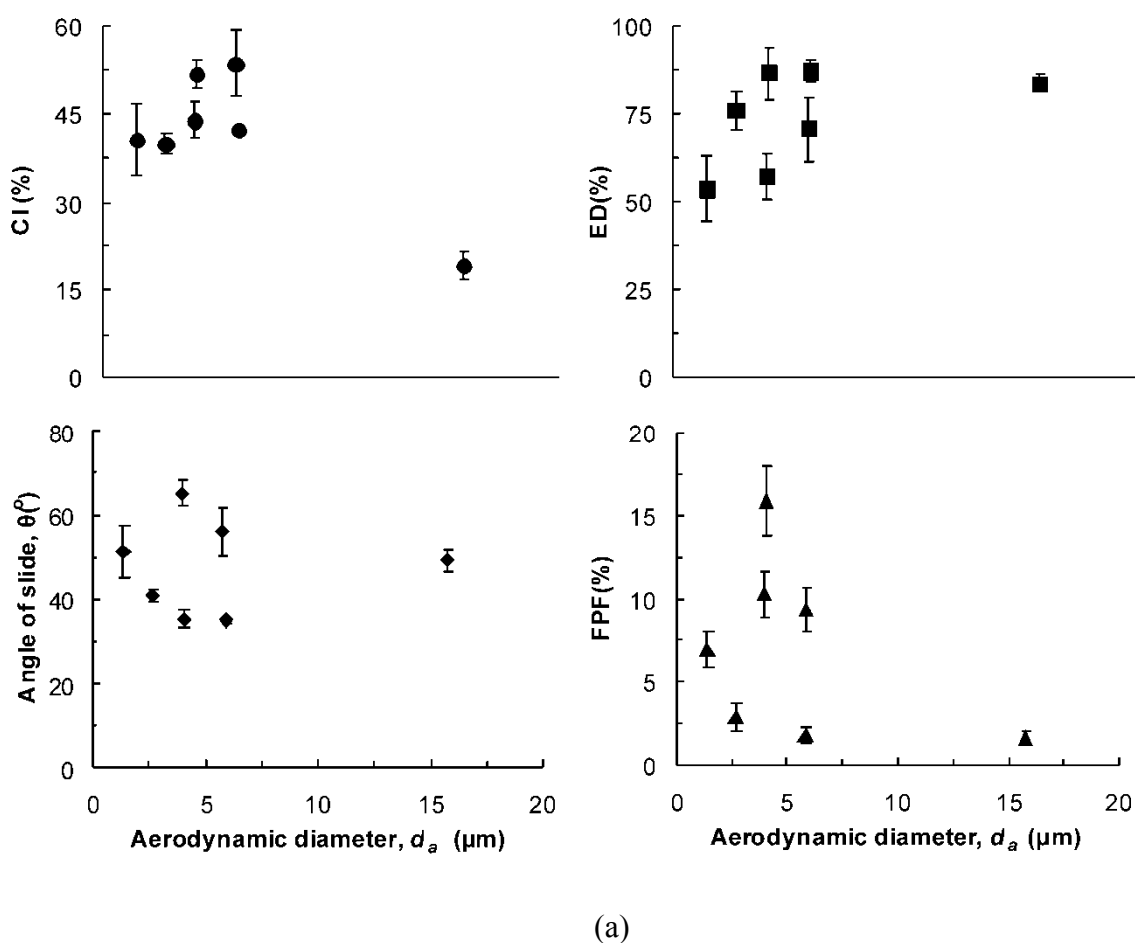
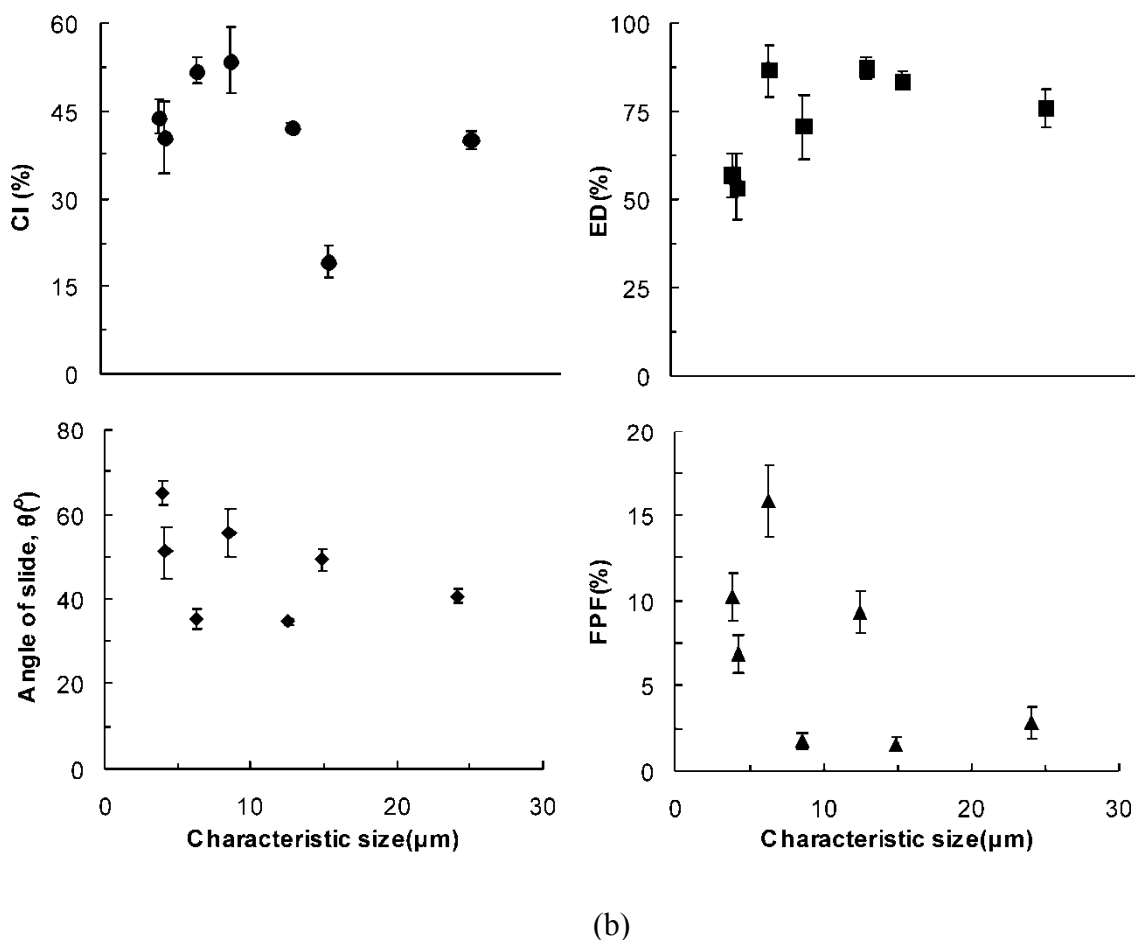


Figure 5.2 CI, θ , ED and FPF results of different shape particles as a function of their (a) aerodynamic diameter and (b) characteristic size. (Error bars indicate standard deviation, $n=3$)

**Figure 5.3 Cont.**

possibly the distinct shape of the particle samples. Therefore, the inhalation properties of the particles will need to be analyzed separately according to either the aerodynamic diameter or the characteristic size.

Aerodynamic diameter takes into account the difference in particle size, density and shape. It is also a commonly accepted way of categorizing aerosol particles. Therefore, the particles used in this study are grouped into three aerodynamic diameter ranges. The CI, θ , ED and FPF results of the different shape particles in the respective aerodynamic diameter ranges are shown in Figure 5.3. Other than spherical II particles, the aerodynamic diameter of all the particles used are within a narrow range of about 1.4-

5.9 μm . The p values of the statistical analysis within each size group are also presented in the Figure. The superscripts like '#', '*' and 'o' are used to notify the p values for a particular group marked with that sign. The difference among the experimental results within each size group can be considered statistically significant for $p < 0.05$.⁸²

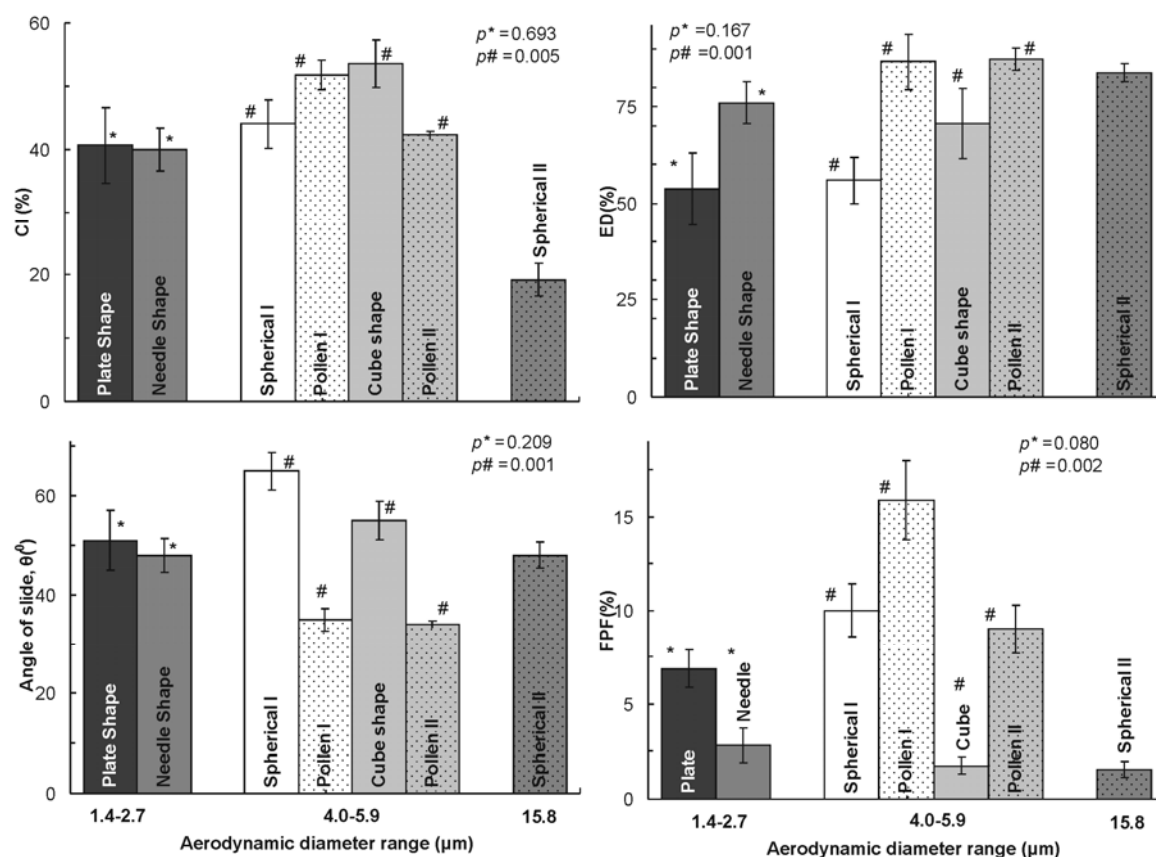


Figure 5.4 CI, θ , ED and FPF results of different shape particles in similar aerodynamic diameter range. (Error bars indicate standard deviation, $n=3$)

Needle-shape and plate-shape particles are in the similar d_a range of 1.4-2.7 μm . In this size range, both plate-shape and needle-shape particles show comparable CI and θ . Generally, CI of 25 and above indicates poor flow and 15 and below indicates good flow.¹⁶² In this study, most of the particles used are observed to have high CI. Both needle-shape and plate-shape particles has high CI of around 40. Because of the flat

surfaces, the contact surface areas of these particles are large. The particles would experience high interparticle forces and form aggregates. Their high CI would be a reflection of the breakage of the aggregates and rearrangement of particles.⁹⁴ The aggregation tendency of the particles causes high resistance to flow, thus high θ is also observed. However, though the particles show similar flowability, needle-shape particles have a higher ED than the plate-shape particles. Even though both particles have similar aerodynamic diameter, in fact the needle-shape particles have much higher characteristic size than the plate-shape particles. Therefore, the needle-shape particles should have lower inter-particle interaction and showed lower cohesiveness than the plate-shape particles. As shown from the regional deposition results in Figure 5.4, larger amount of the plate-shape particles would be deposited in the inhaler region compared to needle-shape particles. Despite the fact that plate-shape particles have lower ED than needle-shape particles, they exhibit higher FPF. It indicates that even though the needle-shape particles are easier to be aerosolized than the plate-shape particles, the relatively large major dimension of the needle-shape particles make it more susceptible to early deposition because of interception deposition mechanism.⁶⁴

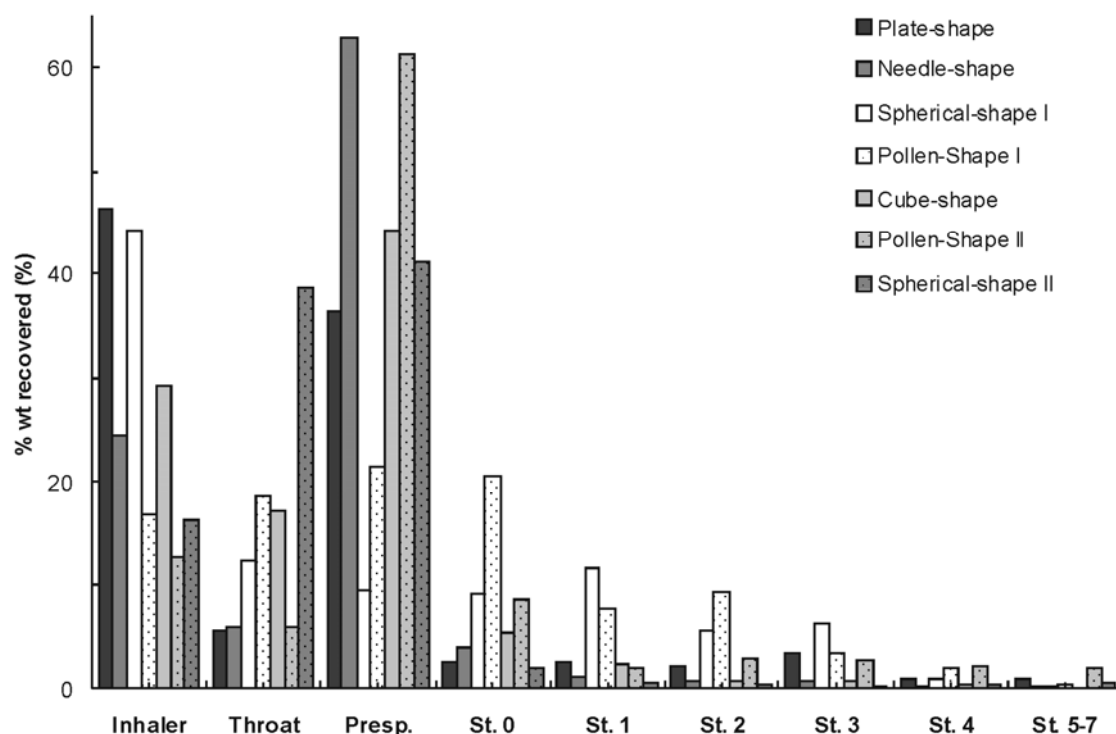


Figure 5.5 Regional deposition in the Andersen cascade impactor for different shape particles.

Spherical I, pollen-shape I, cube-shape, and pollen-shape II particles have comparable d_a in the range of 4.0-5.9 μm . CI results indicate that spherical I and pollen-shape II would have better flowability than cube-shape and pollen-shape I particles. However, θ results show that both pollen-shape I and pollen-shape II particles can flow better than spherical I and cube-shape particles. Since CI makes use of the difference in tap density and bulk density, high CI can indicate weak interparticle forces being overcome by tapping. The weak interparticle forces may lead to good flowability, as indicated by the contradicting behavior in CI and θ of pollen-shape I particles. It is possible that the surface features of the pollen-shape particles increase the distance between two interacting particles and reduce their contact surface. Thus, the interparticle

forces would be lower. The low interparticle forces of pollen-shape particles also lead to better ED than cube-shape and spherical I particles. Both pollen-shape particles have an excellent ED of 86-88%. However, because of the large characteristic size, pollen-shape II particles have higher deposition in the preseparator and lower FPF is observed compared to pollen-shape I particles. On the other hand, cube-shape and spherical I particles show poor flowability in both CI and θ . Therefore, it is believed that the interparticle forces are stronger for cube-shape than the spherical I particles. The strong interparticle forces also result in low ED of both particles. However, a big difference between the spherical I and the cube-shape particles is that despite a low ED, spherical I particles can still give a relatively high FPF. Since the cube shape particle possesses large characteristic size, most of them are deposited in the preseparator and the initial stages of the impactor and a less amount can go to the lower stages.

The spherical II particles have an average d_a of 15.8 μm , much higher than other particles. As expected, the large size favours better CI, θ and ED. However, deposition due to inertial impaction and interception in the throat and preseparator are also increased. Therefore, the low FPF is observed for the spherical II particles.

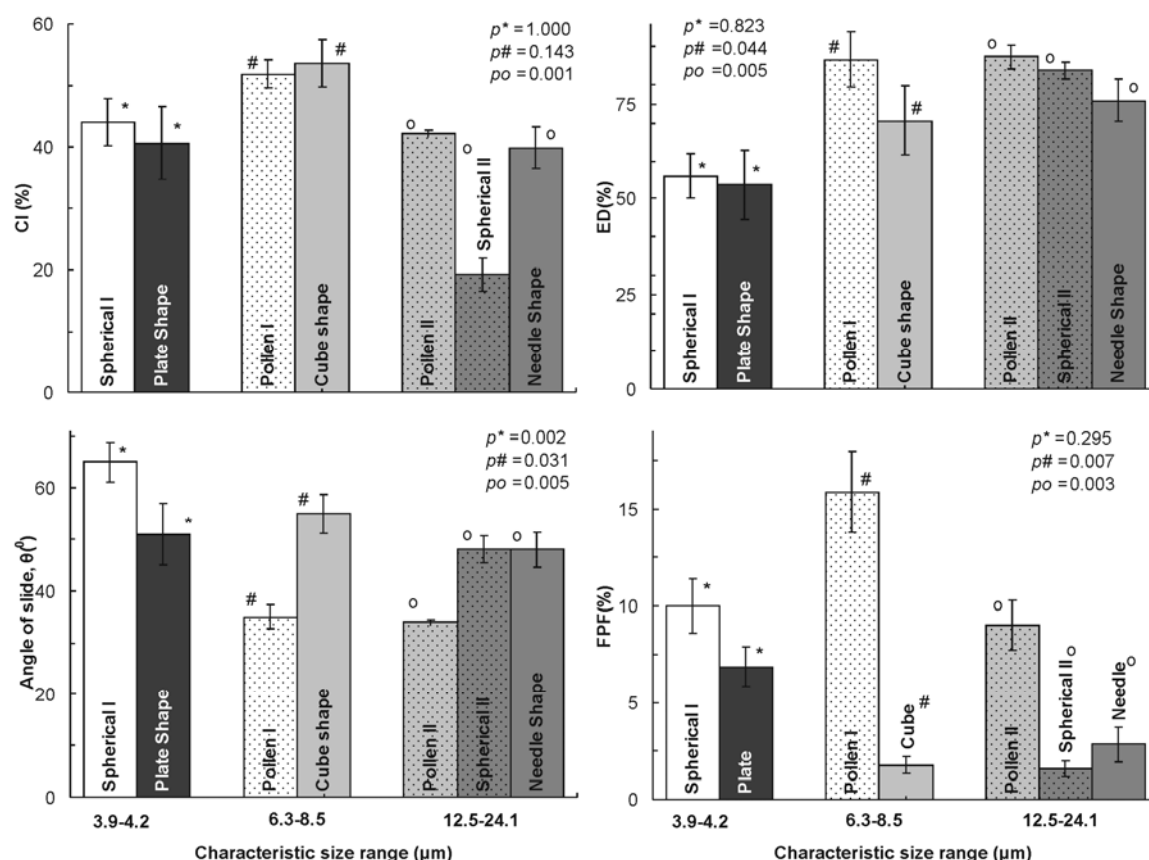
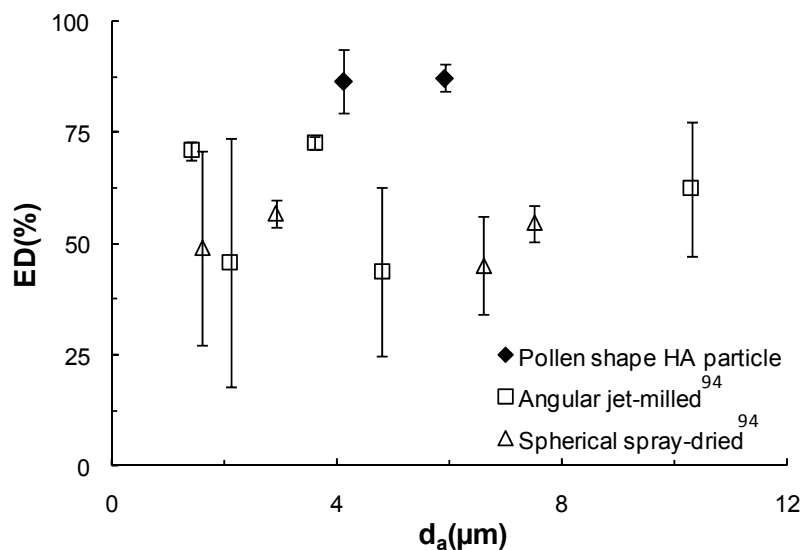


Figure 5.6 CI, θ , ED and FPF results of different shape particles in similar characteristic size range. (Error bars indicate standard deviation, $n=3$)

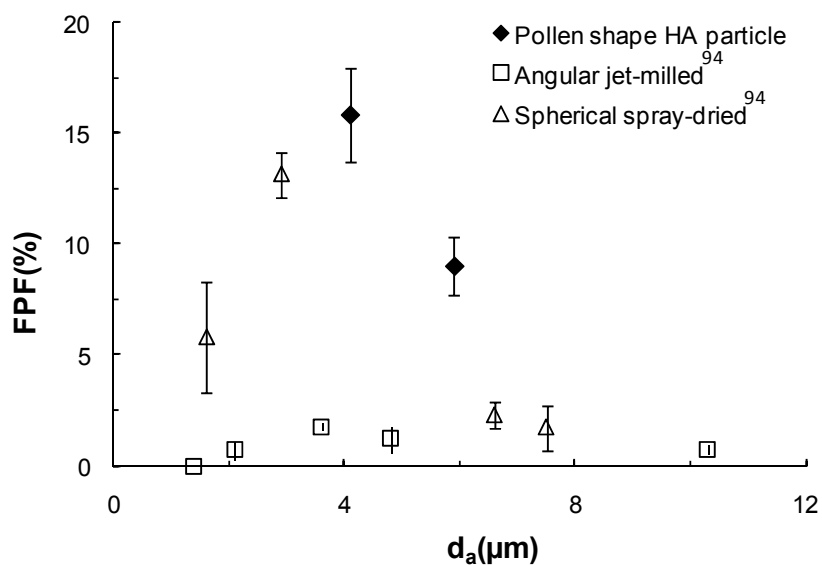
The physical size of particles also plays a vital role in the flow, aerosolization and dispersion behavior. Figure 5.5 shows the CI, θ , ED and FPF results of the different shape particles grouped in similar characteristic size range. It can be seen that the grouping of particles based on characteristic size is different from that based on aerodynamic diameter. Spherical I and plate-shape particles are in smaller size range of 3.9-4.2 μm , pollen-shape I and cube-shape particles are in medium size range of 6.3-8.5 μm and pollen-shape II, spherical II and needle-shape particles are in large size range of 12.5-24.1 μm .

The physical size of the particles would be closely related to the interparticle forces as well as the deposition mechanism in the cascade impactor. In general, small physical size would have stronger interparticle forces than inertial forces within the size range of 1-10 μm .⁹⁴ The strong interparticle forces would reduce the particle flowability. Therefore, large particles generally have higher ED. On the other hand, large particles are more susceptible to early deposition due to inertial impaction and interception and low FPF is observed. As shown in Figure 5.5, both flowability (in terms of θ) and ED follow a general increase with an increase in particle characteristic size while FPF follows a general decrease with an increase in particle characteristic size with an exception of both pollen-shape particles. In both medium and large characteristic size range, both pollen-shape particles show good θ , ED and FPF results. The FPF of pollen-shape II is even comparable to the much smaller sized spherical I particles. As discussed in the earlier section regarding aerodynamic diameter, the pollen-shape structure minimizes the interparticle forces and aggregation tendency. It would be easier to disperse and deliver pollen-shape particles in dry powder inhalation.

5.3.4 Comparison with literature results



(a)



(b)

Figure 5.7 Comparison of (a) ED and (b) FPF of pollen-shape HA particles with angular jet-milled and spherical spray-dried particles. (Error bars indicate standard deviation)

There are limited ED and FPF results in the literature using single particle formulation in cascade impactor. The ED and FPF of both pollen-shape I and pollen-

shape II particles are compared with angular jet milled and spherical spray dried mannitol particles with aerodynamic diameter lower than 10 μm .⁹⁴ The mannitol particles were aerosolized at an air flow rate of 60 L/min with an actuation time of 20 seconds whereas the flow rate used in this study is 30 L/min with the same actuation time. It can be seen in Figure 5.6 that despite the lower air flow rate, the pollen-shape I and pollen-shape II particles show higher ED and FPF than the angular and spherical particles of similar size (aerosolized by Rotahaler®). One of the spherical particles indeed showed higher FPF than the pollen-shape particles. However, the spherical particles have an average size of 2.9 μm , which is much smaller than the 6.3 μm and 12.5 μm pollen-shape particles. It is believed that the high FPF is not contributed from the spherical shape. Pollen-shape particles having similar size as the spherical particles are anticipated to have higher FPF based on the comparison of similar sized particles used in this study. An increase in flow rate can substantially increase the total particle emission from an inhaler.⁴⁰ It is expected that the pollen-shape I and pollen-shape II particles would exhibit a higher ED and FPF at a flow rate of 60 L/min. Based on the results in this study, it shows that the use of pollen-shape particles would be promising in improving the delivery efficiency of dry particle inhalations.

5.4 Concluding remarks

The flow and deposition properties of different shape particles are investigated. Since there is no good parameter to account for particle shape, the different particles are compared against both characteristic size and aerodynamic diameter. It can be seen in this study that even within the d_a range of 1-5 μm , the FPF can vary substantially from 2-16% depending on the shape of the particles. It is found that particle shape has a strong effect

on particle behavior. Careful control of the shape can improve the flowability, aerosolization and deposition performance of the particles. Pollen-shape particles are found to exhibit better flowability, aerosolization and deposition properties compared with other particle shapes. The pollen-shape particles exhibited ED values over 80% where the ED range for the other shape particles 50-75%. The FPF results in 9-16% where for other shape particles it varies in the range 2-10%. Therefore, the use of pollen-shape drugs can be a good solution to improve the delivery efficiency in dry particle inhalation.

Chapter 6 FLOW BEHAVIOR AND DEPOSITION

STUDY OF POLLEN-SHAPE CARRIER

PARTICLES IN AN IDEALIZED INHALATION

PATH MODEL

6.1 Introduction

In the previous chapter, flow, aerosolization and deposition behavior of different shape particles are investigated. Pollen-shape particles exhibit better flow and aerosolization behavior. In this chapter, pollen-shape particles with large carrier size are addressed to examine their inhalation behavior as dry powder formulation. The flow properties, velocity field and turbulence occurrence in an idealized inhalation path model is investigated using particle image velocimetry.

Morphology of the carrier particles has important effects on deposition. Zeng, et al.¹⁰⁷ showed that increasing the elongation ratio of carrier LA particles can increase the respirable fraction of drug from dry powder formulation for inhalation. More elongated particles would be more aerodynamic and would help the particles to go further into the lung.¹⁰⁰ There are contradicting results on the effect of particle surface roughness on the fine particle fraction. Most researchers^{104,106,108} found that increasing the surface roughness of the carrier particles can enhance the particle respirable fraction while Zeng et al.¹⁰⁷ found otherwise.

For pulmonary drug delivery, the oral cavity, pharynx, larynx and the bifurcation regions are the major obstacles to prevent efficient drug delivery to the lungs. Studies have been focused on the undesired depositions in these regions and are conducted either *in vivo* or *in vitro*. *In vivo* studies are conducted in oral passage using radioactive aerosols.¹⁶³⁻¹⁶⁹ *In vitro* measurements are conducted in either anatomical casts or idealized models based on actual dimensions of mouth and throat passageway.^{36,163,170-173} However, information on the flow characteristics of the particles in these regions is limited. Heenan et al.¹⁷⁴ and Grgic et al.^{175,176} used particle image velocimetry (PIV) technique in their specialized mouth and throat model and found a complicated flow structure with several separation and circulation regions. The presence of these separation and circulation regions may have strong impact on the deposition of particles.

The flow behavior and deposition of pollen-shape (spiked sphere) hydroxyapatite (HA) carrier particles is investigated in this study. High surface roughness of carrier particles favors the homogeneity and stability of the formulation blend.¹⁰⁴ Theoretically, the surface structure of pollen-shape particles can attribute the same characteristics. The flow field and the deposition of the pollen-shape particles are studied in an idealized inhalation path model with a bending and a bifurcation. The performance of the pollen-shape particles are compared against typical LA carrier particles with comparable size range. This study also reveals the mechanisms and parameters responsible for aerosolization, flow and deposition of particles.

6.2 Experimental

6.2.1 Preparation of HA & LA

Particles with a surface morphology that increase the separation distance may have lower interaction force and eventually would have high potential for better dispersion and aerosolization. After the preliminary study with different shape particles, pollen-shape particles become more interesting. Large pollen-shape particles are involved as model carrier in the flow behavior study. On the other hand LA particles are the most common in use as the carrier for dry formulation. They mostly comprise rock-like shape. Therefore, the physical flow behavior of the HA model carrier particles are compared of rock-like LA particles with similar size range.

The preparation of HA particles has been described in the Experimental chapter (Section 3.2.1). HA-2 with a PSS concentration 40 g/L & at 120°C, and HA-3 with PSS concentration 30 g/L at 200°C are utilized in this study. The LA particles with size range of 20-38 μm (LA-1) and 38-75 μm (LA-2) are used.

6.2.2 Particle characterization

Synthesized particles are characterized with their size, shape & morphology, powder density, and powder flowability.

6.2.7 Experimental setup and PIV measurement

The schematic diagram of the experimental setup is shown in Figure 6.1. Air is supplied from a compressed air source through a flow meter using Teflon tubing. In dry powder inhalation, it is a common practice to use a fixed amount of the formulation in a single inhalation. Therefore, 20 mg of each sample is used for each run to simulate the powder inhalation process. Test particles are introduced in a single pulse also to minimize

particle deposition on the wall of the inhalation path model, which would hamper the visualization of the flowing particles. As the size and density of the particles are different, the concentration of particles in the experimental region would be different. However, particle concentration in the system has little effect on the flow field measurement technique, that is, particle image velocimetry (PIV). The working principle of PIV system is described in the 'Experimental' chapter. Since the focus of this study is to investigate the velocity profile of different powders in the inhalation path model, the effect of particle concentration in the system is neglected. A needle valve is used to control the flow. Two filters are used to collect the particles at the two bifurcation outlets of the inhalation path model.

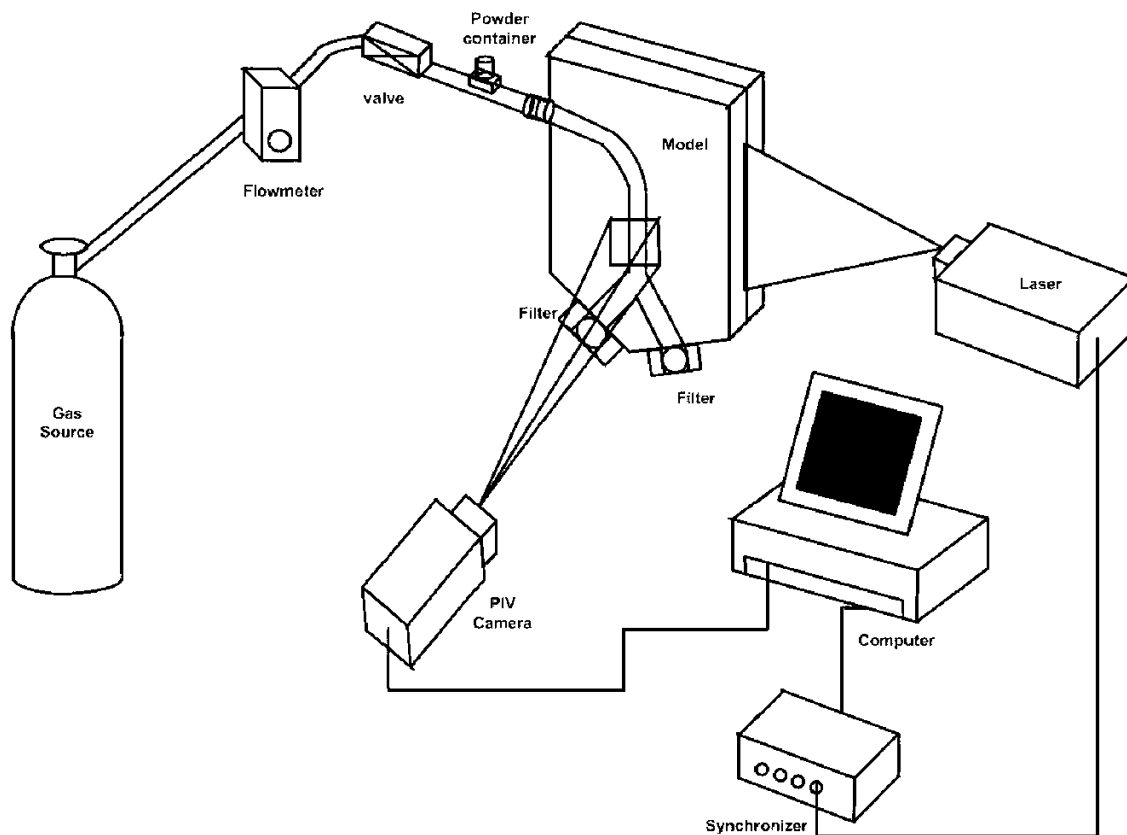


Figure 6.1 Schematic of the experimental setup.

The inhalation path model is made of acrylic with an idealized bending and bifurcations. The dimensions of the bending and bifurcations of the inhalation path model are shown in Figure 6.2. The model comprises a front and back piece for the ease of cleaning. The bending and bifurcations are machined on two rectangular acrylic blocks so that the outside surface is flat when the two pieces are joined together. The initial horizontal section with the bending can be considered as oral cavity, the vertical section comprised of pharynx, larynx and trachea and the two branches after the bifurcation resembles generation 1 of the lung airway. The diameter and length dimensions of the model are equivalent to the lung dimensions proposed in literature.^{8,177} However, for simplicity, the diameter of the cast is kept constant up to the bifurcation and a 60° bifurcation angle is used. The two bifurcations are of different lengths to study the effect of distance. Though the path model has the bending and bifurcation section casted on the same plane, experimental results show that the particles can achieve a uniform distribution before reaching the bifurcation section. Silicon oil is coated on the surface of the path model to prevent particle bounce.

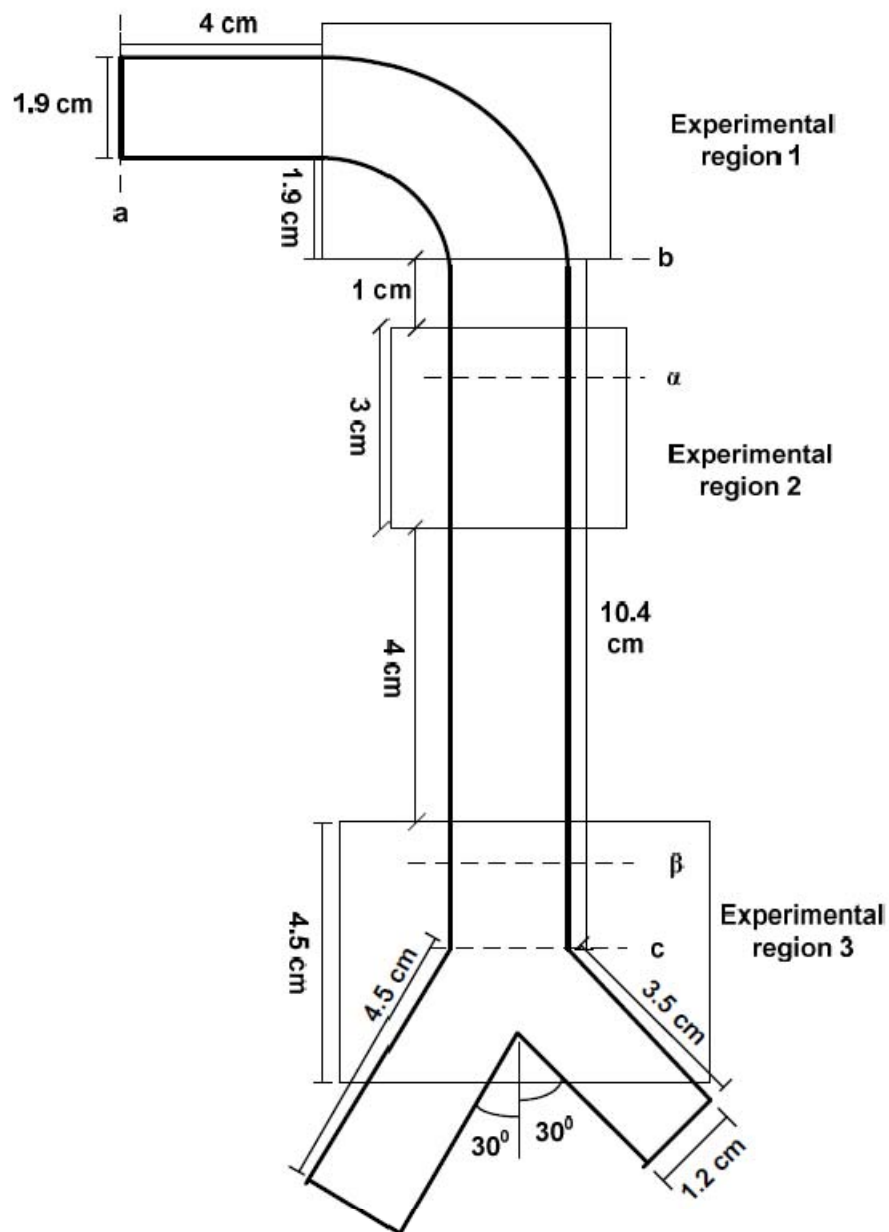


Figure 6.2 Dimensions of the idealized bending and bifurcations of the inhalation path model.

Instantaneous flow field measurements are conducted by using particle image velocimetry (PIV). PIV measurement is focused on three regions denoted as experimental region 1, 2 and 3 in Figure 6.2. Instantaneous flow field of the particles is measured at the

instant when the majority of the samples are passing through experimental region of interests. Radial distribution of instantaneous particle velocity is also determined at point α and β . A Solo PIV laser system (New Wave Research, Inc.) is used to illuminate the particles flowing through the path model. The laser system consists of a pair of 30 mJ/pulse Nd:YAG lasers capable of producing 5–7 ns pulses at 10 Hz. The particle images are recorded at a resolution of 1280×1024 pixels and 15 Hz frame rate using a PowerView Plus Camera (TSI Incorporated). Once the camera system and laser sheet has been set up and aligned for a particular location, the working distance between the camera and image plane is measured.

The time difference between the two consecutive images for vector analysis, Δt will need to be optimized for accurate velocity measurement. The optimum Δt value is determined to be 20 μ s by trial and error. The PIV exposure and laser pulse delay is also optimized for better image. The image data are analyzed using the insight 3G software package from TSI. The vector fields are generated using Fast Fourier Transform (FFT) correlator. PIV data is always arranged in a rectangular grid. The interrogation region is decreased from 64×64 pixel to 32×32 pixel with an overlap of 50%. Recursive Nyquist Grid is selected as the grid engine. The field of view is 25 mm×35 mm for most of the measurements. A range validation and a filter are used to avoid erroneous vectors.

For deposition study, at the end of each deposition experiment, the particles deposited in the different regions of the path model are extracted separately. Regional deposition is quantified in four sections of the path model, namely the bending section, the vertical section, the bifurcation section and the filter. As shown in Figure 6.2, the bending section is taken from position “a” to position “b”, the vertical section is taken

from position “b” to position “c”, and from position “c” to just before the filter outlets is considered as the bifurcation section. For the HA particles dilute acidic solution (2% HNO_3) is used as the extraction solvent. Then the samples are quantified with inductively coupled plasma (ICP). The deposition efficiency is defined as the amount of the particles deposited in the bending, vertical and bifurcation sections per the total amounts of particles fed into the path model.

6.3 Results and discussion

6.3.1 Particle characteristics

Precipitation of HA powders is achieved in aqueous solution at an elevated temperature under hydrothermal conditions. Two HA samples are synthesized at 100°C with PSS concentration of 40 g/L and at 200°C with PSS concentrations of 30 g/L, respectively. The SEM images of the two samples are shown in Figure 6.3. It can be seen from the SEM images that the first sample (HA-1) has petal like surface morphology while the second sample (HA-2) has a needle like surface morphology.

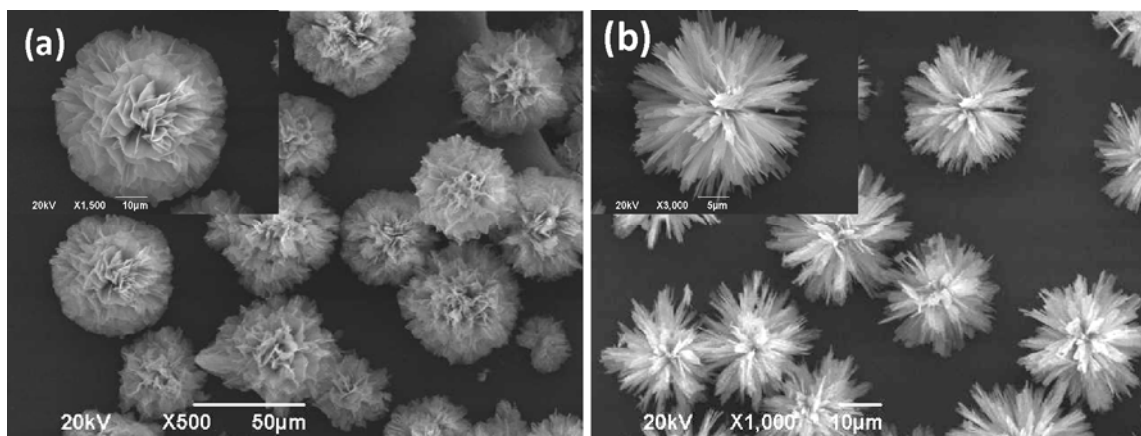


Figure 6.3 SEM images of HA particles synthesized at (a) 100°C, 40 g/L PSS concentration (HA-1) and (b) 200°C, 30 g/L PSS concentration (HA-2).

The LA particles are sieved into two different size ranges. It is to note that sieving alone is not possible to completely separate the fine particles from the larger particles. A fraction of fine particles is present in both LA samples shown in the SEM images in Figure 6.4. The particle size distribution is measured based on the SEM images. The presence of fine particles would result in a lower mean particle size and higher standard deviation of the particle size distribution.

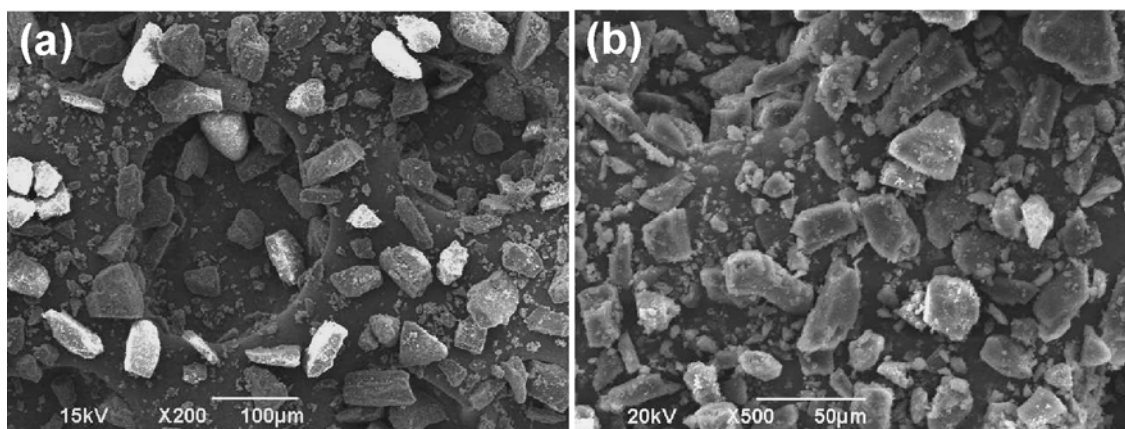


Figure 6.4 SEM images of α -lactose monohydrate with a size range of (a) 38–75 μm (LA-1) and (b) 20–38 μm (LA-2).

A comparison of the physical properties of the HA and the LA particles are shown in Table 6.1. Aerodynamic diameter is used as the characterizing property of aerosol particles. The aerodynamic diameter is defined as the diameter of a sphere of unit density that has the same terminal settling velocity as the particle under consideration,

$$d_a = d_g \sqrt{\frac{\rho_e}{S\rho_s}}, \text{ where } \rho_s = 1 \text{ g/cm}^3, d_g \text{ is the particle geometric diameter, } \rho_e \text{ is the}$$

effective particle density and S is the particle shape factor.⁵⁸ The effective density can be approximated by the tap density, ρ_{tap} . The shape factor is defined as the ratio of the particle surface area to the surface area of a sphere with the same volume as the particle. The shape factor of a sphere is 1 while any shape deviating from a sphere will have a shape factor larger than 1. The pollen-shape closely resembles a spherical particle, and is assumed to have a shape factor of 1. The LA particles are assumed to be rectangular shape for the estimation of the shape factor. Rectangular prism with an aspect ratio of 1 to 4 has a shape factor of 1.24 to 1.49. Therefore an average shape factor of 1.3 is assumed for the LA particles. It can be seen in Table 6.1 that HA particles have lower difference between ρ_{bulk} and ρ_{tap} than LA particles. One possible reason is that due to the petal-like or needle-like surface morphology HA particles have high separation distance. Thus, the tapped packing of the HA particles is loose. On the other hand, flat surfaces of the LA particles lower the separation distance among the particles and enhance tight packing. Therefore, higher tap density values are observed with LA particles. Another reason could be the size distribution of the LA samples. LA samples comprise some fine particles and those particles can easily be organized among the large particles in packing bed. This could also make the ρ_{tap} much higher than the ρ_{bulk} for the LA particles.

Table 6.1 Physical properties of experimental HA and LA particles

Sample	Mean particle size (μm)	Standard deviation (μm)	ρ_{bulk} (g/cm^3) ($n=0$)	ρ_{tap} (g/cm^3) ($n=2500$)	$d_{\text{a,theoretical}}$ (μm)
HA-1	48.5	10.21	0.215	0.289	26.05
HA-2	28.2	4.02	0.105	0.218	13.18
LA-1	42.7	25.30	0.586	0.874	35.01
LA-2	25.1	11.80	0.316	0.760	19.19

6.3.2 Powder flow

The Carr's compressibility index (CI) is commonly used to characterize powder flow. CI value quantifies the powder flow based on the concept that compressible powders are more cohesive and exhibit poor flow behavior. The CIs for the HA and LA samples used in this study are shown in Figure 6.5. A comparison of the HA and LA samples with similar geometric size range shows that the pollen-shape HA exhibits better flow behavior than the irregular shape LA particles. The surface structure of the HA particles increases the distance between the particles and reduces the contact surface. Thus, it is anticipated that the van der Waals forces would be smaller and allow better dispersion of particles.

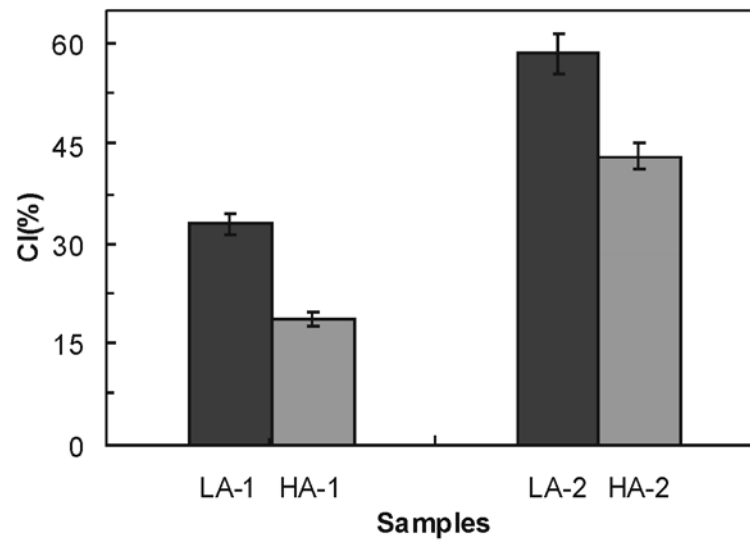


Figure 6.5 Compressibility index of the HA and LA samples with similar size range.

6.3.3 Flow behavior in the inhalation path model

A flow rate of 30 L/min, which is similar to the typical adult inhalation rate, is used for the experiments. The gas flow rate is equivalent to an average superficial gas velocity of 1.76 m/s in the path model. The PIV measurements of the instantaneous velocity field and the 2D streamlines of the LA-1 particles at experimental region 1 are shown in Figure 6.6. It is to note that particle velocity is not equivalent to particle concentration in the PIV results. It can be seen that the large LA-1 particles with high inertia are flowing through the lower region of the horizontal section. This is due to large size of the LA-1 particles. Most of the particles are flowing close to the bottom wall of the channel due to the dominance of gravitational force, which would enhance the particle deposition in the region.

As the particles pass through the bending, an empty region is observed at the upper left wall of the vertical section. When a fluid is flowing through a channel under a turbulent or transition regime, due to the disturbance or change in direction of the

geometry, fluid streamlines may be interrupted or separated. The phenomena results in an empty region in the flow which is termed as the separation region. The separation regions occur and vary with the geometry of the flow channel and the Reynolds number^{174,175}. There is less or no flow in the empty region. Therefore, PIV cannot detect many vectors in the separation region. The presence of the separation region affects the flow of aerosol particles. Separation region reduces the effective flow area for the particles. Therefore, particle concentration would increase as the particles pass through an area with flow separation. The increase in particle concentration may results in a higher rate of particle collision with each other or with the wall which enhance particle deposition. Therefore, large LA-1 particles show high possibility of inertial impaction in the bending section of the path model.

Streamlines in Figure 6.6(b) are concentrated at the bottom of the horizontal portion and at the posterior wall of the bending section. Concentrated streamlines at the horizontal section indicate possible deposition due to gravitation. The concentrated streamlines at the posterior wall would indicate deposition due to inertial impaction. Generally, gravitation deposition and inertial impaction are the two most important deposition mechanisms for large particles.¹⁰ PIV measurement results of smaller sized LA-2 sample are shown in Figure 6.7. It can be seen in Figure 6.7(a) that the high velocity vectors are evenly distributed in the center of the geometry and smaller separation region is present compared with the LA-1 sample. This indicates that a reduction in particle size is effective in reducing the possibility of inertial impaction and gravitational deposition. The smaller LA-2 particle size also results in a higher particle velocity than that of the LA-1 particles. The streamlines of LA-2 in Figure 6.7(b) show

that LA-2 particles are able to follow the geometry of the bending. Therefore, a lower particle deposition for LA-2 particles can be expected in this region.

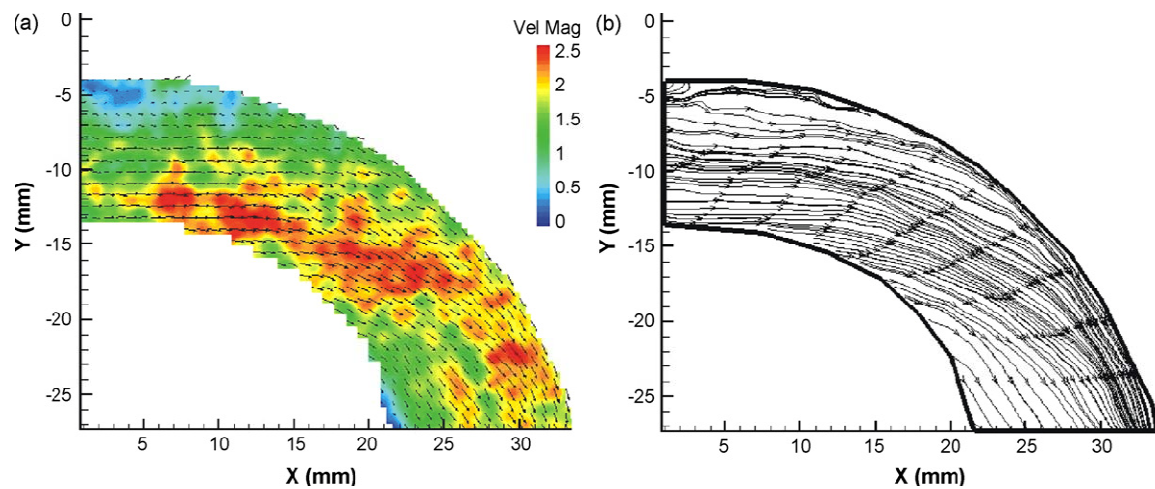


Figure 6.6 PIV results for LA-1 with an inhalation flow rate of 30 L/min, (a) instantaneous velocity field; (b) 2D streamlines at the bending section.

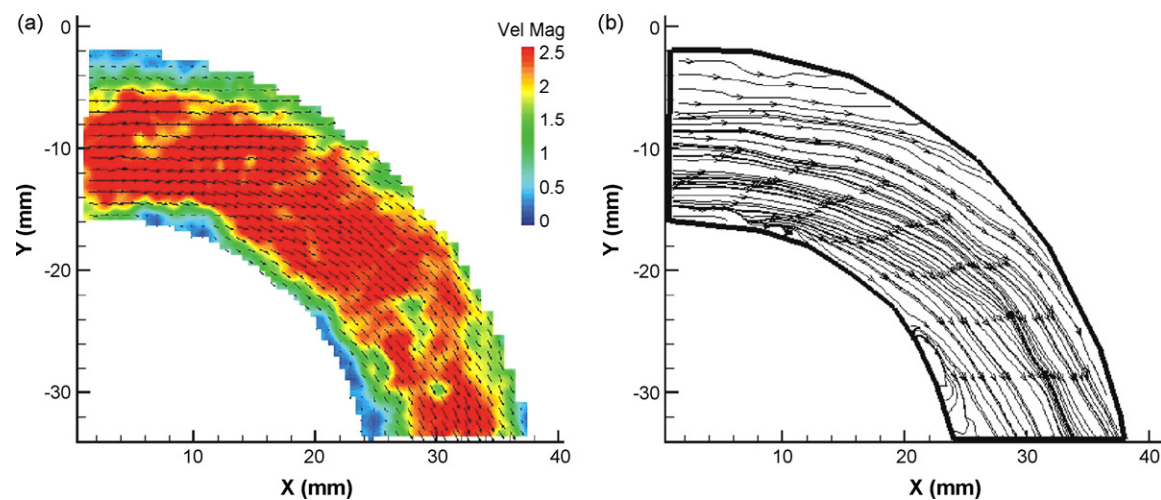


Figure 6.7 PIV results for LA-2 with an inhalation flow rate of 30 L/min, (a) instantaneous velocity field; (b) 2D streamlines at the bending section.

The flow field and streamlines of the HA-1 sample at experimental region 1 are shown in Figure 6.8. It shows that even though the HA-1 sample has comparable size to the LA-1 sample, an evenly distributed flow field can still be present. The distribution of

the vectors shows that the particles are flowing in the air without declining to the bottom section like the LA-1 particles due to gravitation. Particles can pass to the bending section without any severe deposition by sedimentation in the horizontal section. While the HA-1 particles passes through the bending section, a smaller separation region is observed compare to the LA-1 sample. The smaller separation region let the HA-1 particles to flow through a larger volume that makes the HA-1 particles less susceptible for deposition at the bending section by inertial impaction. The streamlines of the HA-1 sample are also following the bending geometry properly while the LA-1 streamlines are concentrated to the right wall after the bending section. It makes the HA-1 particles to have higher possibility to pass through the bending section and reduce the inertial impaction with the wall. The flow streamlines of Figures. 6.7 and 6.8 show that the flow characteristics of HA-1 sample are comparable to the LA-2 sample that has a lower aerodynamic diameter. The flow characteristics of the pollen-shape HA-1 particles can be attributed to both the pollen-shape structure as well as the low particle density. The pollen-shape structure allows the HA-1 particles to flow as discrete particles. The discrete particle flow together with the low particle density allows the HA-1 sample to flow with lower inertia and follow the fluid stream. Therefore, the HA-2 sample that has an even lower aerodynamic diameter exhibits more favorable flow characteristics around the bending showed in Figure 6.9. Due to the low aerodynamic diameter, the HA particles indeed shows that the HA-2 sample has the small separation region and flow through the bending section with low inertia posing less possibility of inertial impaction to the opposite wall. The measured flow behaviors show that pollen-shape HA particles

potentially reduce the deposition in the upper bending section of the inhalation flow path and continue to flow to the lower regions of the human airways.

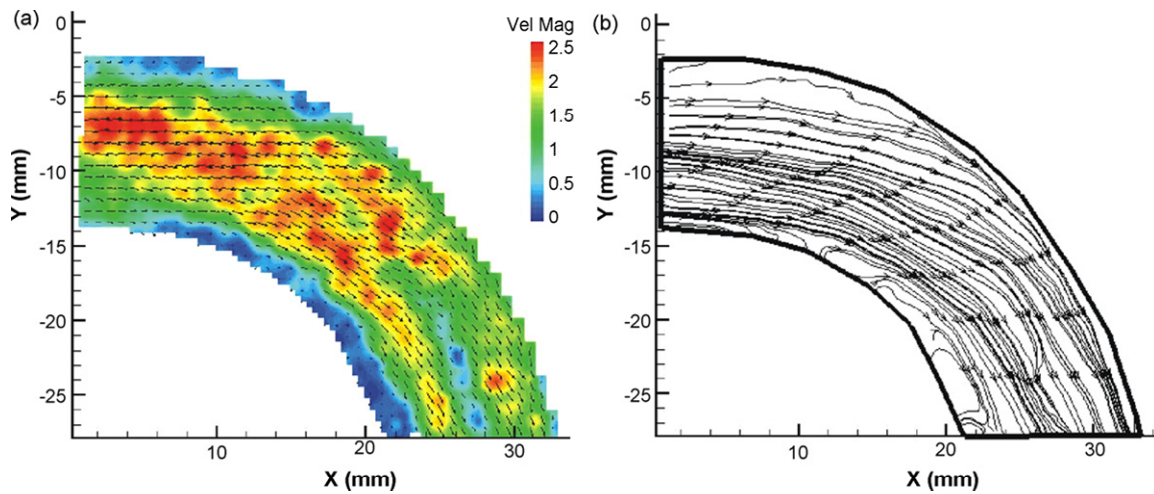


Figure 6.8 PIV results for HA-1 with an inhalation flow rate of 30 L/min, (a) instantaneous velocity field; (b) 2D streamlines at the bending section.

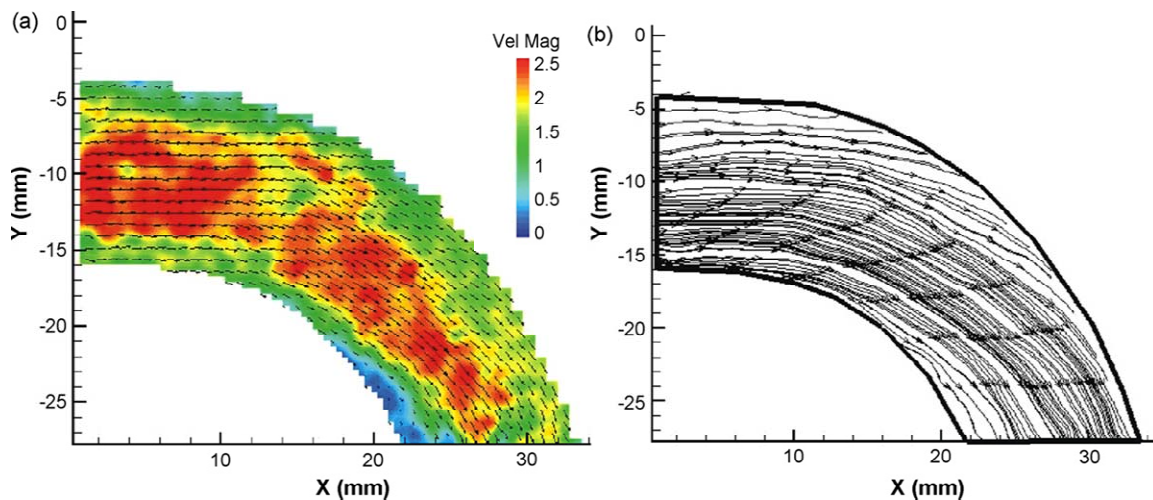


Figure 6.9 PIV results for HA-2 with an inhalation flow rate of 30 L/min, (a) instantaneous velocity field; (b) 2D streamlines at the bending section.

The flow fields of LA-1 and HA-2 samples at experimental region 2 are shown in Figure 6.10. Both LA-1 and HA-2 particles have a high velocity field near the anterior wall region of the upper vertical section as the particles travel past the bending section. This is a result of the particle flow inertia over the bending section. Similar flow structure

is also observed by other researchers in their models.¹⁷⁸⁻¹⁸⁰ Comparing Figures. 6.10(a) and (b) it can be seen that, LA-1 has higher particle velocity than HA-2 because the larger sized LA-1 particles are subject to higher gravitational force in the vertical section. For both LA-1 and HA-2, the higher particle velocity can be found near the anterior wall due to inertia (indicated by the high particle velocity on the right hand side of the vertical section in Figures. 6.10(a) & (b)). In addition, it should be noted that due to the reflection from the circular wall, PIV is unable to measure the velocity at the wall. Therefore, the edge of Figures. 6.10(a) & (b) is slightly off the wall. For a particle laden flow, particles with higher velocity would have a thicker boundary layer.¹⁸¹ Thus, the highest particle velocity for LA-1 is located 0.5 mm away from the wall while the highest particle velocity for HA-2 is located near the wall.

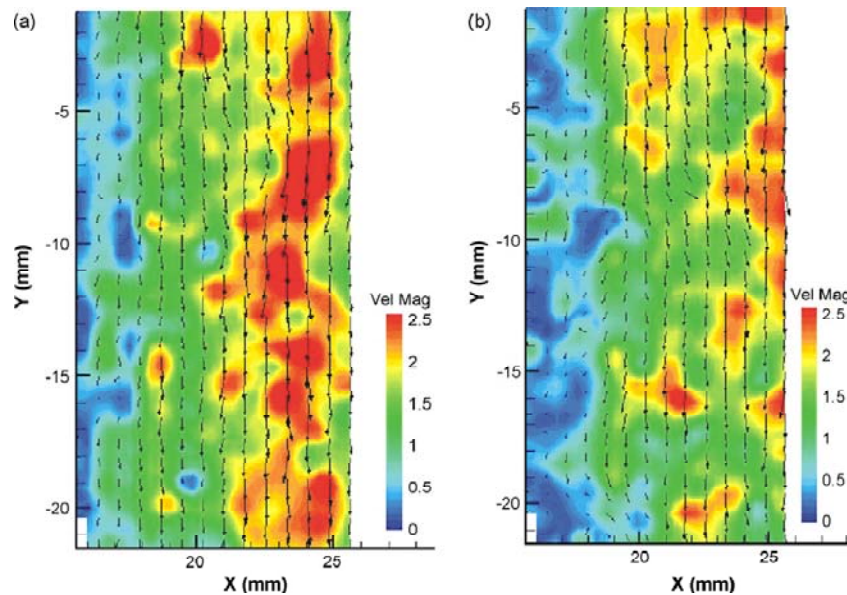


Figure 6.10 PIV results for instantaneous velocity fields of (a) LA-1, (b) HA-2 particles with an inhalation flow rate of 30 L/min at the experimental region 2.

The flow fields of LA-1 and HA-2 particles at the experimental region 3 are shown in Figure 6.11. The PIV measurements are performed with the laser placed on the

left side of the bifurcation. The laser sheet is unable to reach the right leg of the bifurcation due to the model structure. Therefore, the left leg has a clearer velocity field than the right leg as shown in the Figure 6.11. Opposite results are found when the laser is placed on the right side. It can be seen from Figure 6.11 that both LA-1 and HA-2 samples show a developed flow field right before the bifurcation section as opposed to the higher flow velocity at the anterior wall observed at the top of the vertical section. The radial distribution of the instantaneous particle velocity of LA-1 sample at the top (point α in Figure 6.2) and bottom (point β in Figure 6.2) of the vertical section is measured. From Figure 6.12, it can be seen that the particle velocity is higher near the anterior wall at the upper vertical section. However, a developed flow structure can be observed at the lower vertical section. The gas flow rate of 30 L/min gives rise to a turbulent flow with a Reynolds number of 2200. The high dispersion at turbulent flow thus causes the appreciable flow development in the vertical section despite the relatively short vertical flow distance.

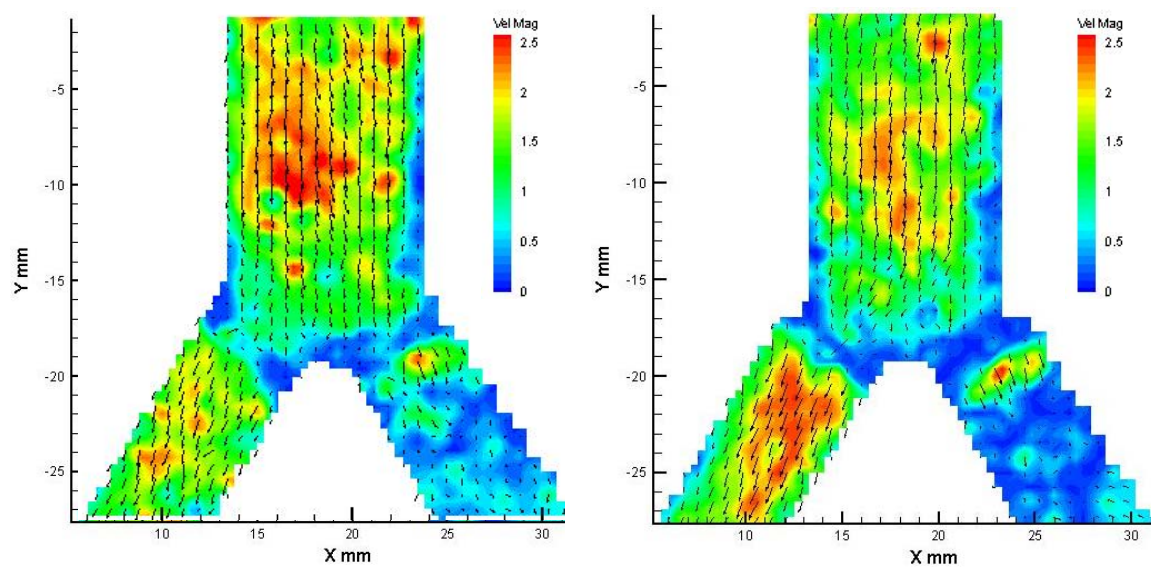


Figure 6.11 PIV results for instantaneous velocity fields of (a) LA-1, (b) HA-2 particles with an inhalation flow rate of 30 L/min at the experimental region 3.

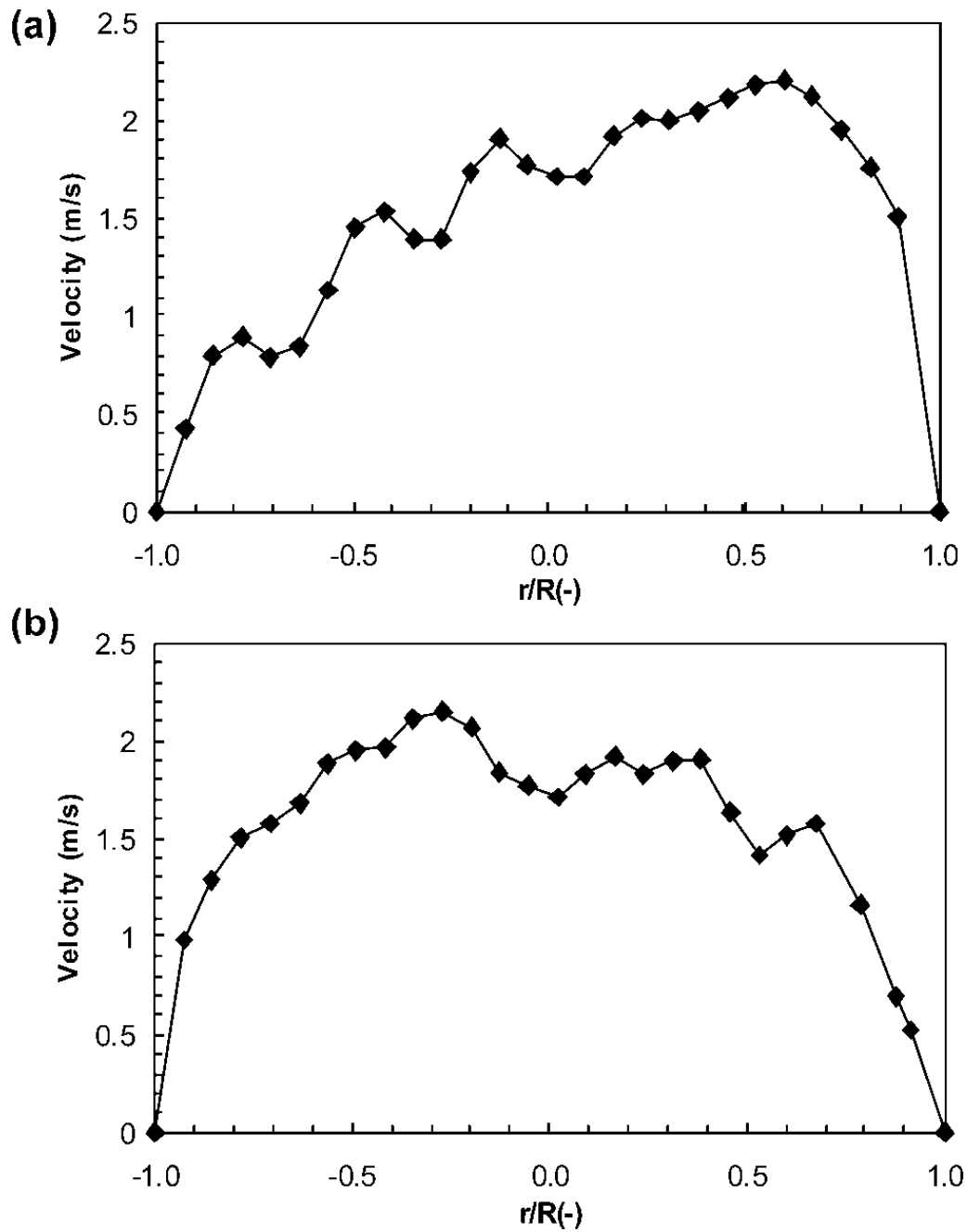


Figure 6.12 Radial distributions of instantaneous particle velocity of LA-1 sample at (a) upper vertical section (point α) and (b) lower vertical section (point β).

6.3.4 Deposition in the path model

The amount of particles deposited at the different regions of the path model is measured at two gas flow rates of 20 and 30 L/min. As shown in Figure 6.13, the bending section has the highest deposition for both pollen-shape HA samples. Deposition is found to be a strong function of particle properties while it is almost independent of the gas flow rate. Most of the depositions at the bending section are found at the bottom of the horizontal region as well as the anterior wall of bend. It agrees well with the literature that the two main mechanisms of the particle deposition are inertial impaction and gravitational deposition.¹⁰ Gravitational deposition causes deposition at the bottom of the horizontal section while inertial impaction causes deposition at the anterior wall. In the vertical section, most depositions are concentrated at the upper anterior wall due to the inertial impaction. The bifurcation section is found to have the lowest deposition among the four test sections. A comparison of the regional depositions of the two pollen-shape HA samples shows that the difference in the particle properties affects mainly the deposition at the bending section and the filter. The HA-2 sample with a lower aerodynamic diameter is effective in shifting early deposition from the bending to the bifurcation sections.

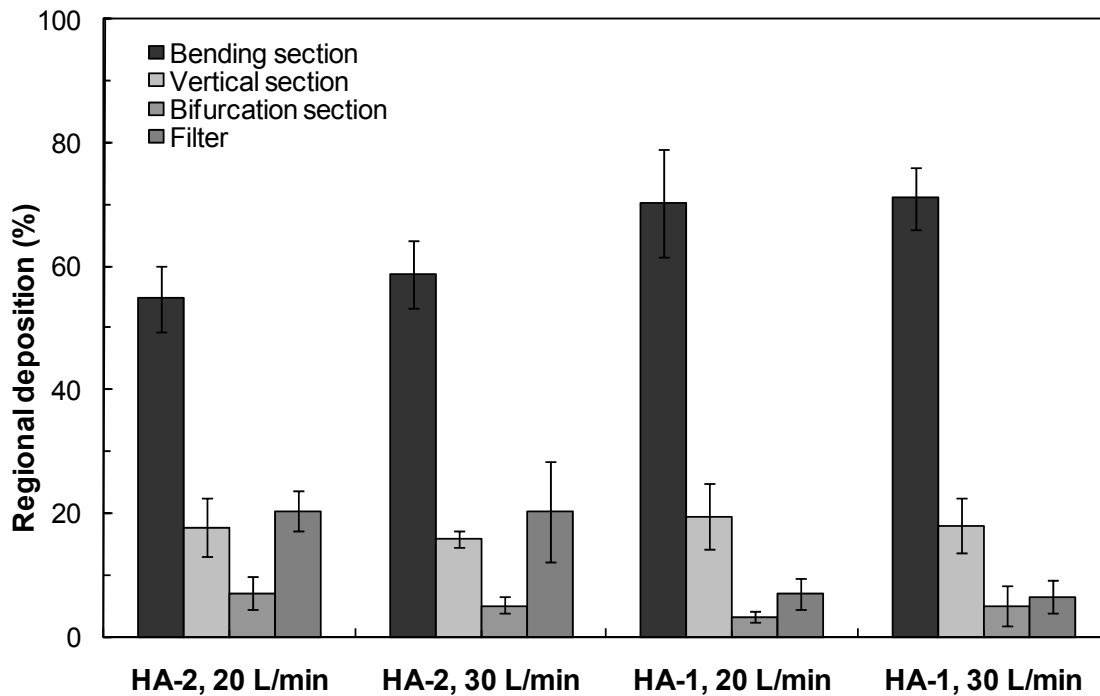


Figure 6.13 Regional deposition of the pollen-shape HA samples.

Deposition efficiency of the two pollen-shape HA particles are determined at gas flow rates of 20 and 30 L/min. It is defined as the ratio of the amount of the particles deposited in the bending, vertical and bifurcation sections to the total amounts of particles fed into the path model. The deposition efficiency is plotted against the particle Stokes number in Figure 6.14. Stokes number is correlated with the deposition efficiency, since it takes into account the relevant length and velocity scales. The particle Stokes number is defined as $Stk = \frac{\rho_e d_g^2 U}{18\mu D}$, where U is the mean velocity of the flow (m/s), μ is the fluid dynamic viscosity (kg/(m s)), D is the diameter of the flow channel (m), ρ_e is the effective particle density (kg/m³) and d_g is the mean geometric particle diameter (m). As shown in Figure 6.14, the deposition efficiency can be well correlated with a logarithmic function of the particle Stokes number,

$$\eta = 6.02 \times \ln(\text{Stk}) + 102.2 \quad (6.1)$$

where η is the deposition efficiency. The equation signifies that the efficiency reduces with the reduction of the Stokes number. Stokes number increases with the increase of the particle size. The increase in Stokes number eventually enhances inertial impaction deposition. The experimental Stokes number range is 0.03-0.33. At this Stokes number range, a high deposition fraction is found in the upper inhalation path model which can be supported by Grgic et al.¹⁷⁵. Grgic et al.¹⁷⁵ showed the deposition efficiency with a Stokes number range of 0.002-0.025 for smaller particles than the particles used in current study. A close correspondence is found between the efficiency trends and the reported result can be considered as an extrapolation of current results. This equation may not be validated for the particles much smaller than 1 μm for which diffusion would be the dominant deposition mechanism.

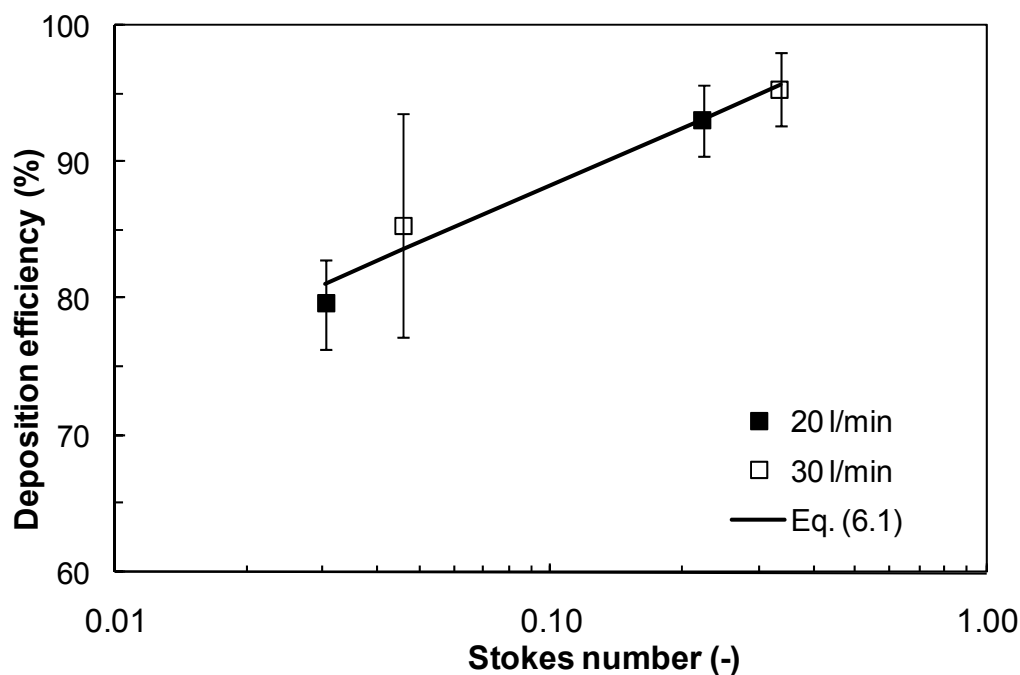


Figure 6.14 Deposition efficiency of the two pollen-shape HA particles as a function of Stokes number with error bars indicated standard deviation.

6.4 Conclusions

The flow behavior and deposition of pollen-shape carrier particles are investigated. Pollen-shape HA particles are synthesized to a comparable size range as typical carrier particles with mean diameter of 30–50 μm and effective density less than 0.30 g/cm^3 . The pollen-shape particles are found to have almost twofold better flow behavior compared with the traditional LA carrier particles in terms of CI index (for HA, CI=18-35%, for LA, CI=32-60%). PIV measurements of the particle flow in an idealized inhalation path model show that the pollen-shape HA particles are able to follow the geometry of the model and generate smaller separation regions compared with the LA particles. The low particle density and pollen-shape surface structure are the main contributors to the flow property of the HA particles. *In vitro* deposition study with gravimetric measurement is

performed with the pollen-shape HA particles at two different flow rates. Inertial impaction and gravitational deposition are the governing mechanisms of early deposition. Most of the particles are found to deposit in the bending section. The particle properties affect mainly the deposition at the bending section and the filter. The deposition efficiency is found to correlate well with the Stokes number of the particles. An empirical correlation is derived for the deposition efficiency of the pollen-shape HA particles as a function of particle Stokes number.

Chapter 7 FEASIBILITY STUDY OF POLLEN-SHAPE DRUG CARRIERS IN DRY POWDER INHALATION

7.1 Introduction

Flow behavior study of pollen-shape model carrier particles flowing with air in an idealized path model is presented in the previous chapter. *In vitro* aerosolization and deposition behavior are also essential to substantiate their performance as carriers in dry powder formulation. In this chapter, *in vitro* aerosolization and deposition behavior of the pollen-shape large carrier particles are assessed as a binary mixture blended with a model drug.

Small particle size allows the particles to travel with the air flow. Particles larger than 10 μm would experience high inertia and cause early deposition.¹⁸² Therefore, drug particles need to be micronized to a size between 1-10 μm , which is found to be the most suitable for dry powder inhalation.^{10,94} Nonetheless, the interparticle forces are considerable for particles between 1-10 μm .⁶⁵ Therefore, the micronized drug particles are normally mixed with larger sized carrier particles. An improvement in the flow and dispersion of carrier particles would improve the drug delivery efficiency.

The flow and dispersion of particles depend on the particle size, shape and surface properties.¹⁸³ The size of carrier particles for dry powder inhalation studies used in the literature covers a wide range of 10-220 μm .^{51,98,103,105,106,108,110} There are mixed reports

on the most efficient size range for carrier particles.^{40,95,96,111} Elongated particles have higher probability to travel further in the air stream and can help improve the overall respirable fraction of drug.^{100,102,107} The effect of carrier surface properties on drug inhalation efficiency is not clearly understood. A smooth carrier surface has stronger adhesion force on the drug particles and shows a better attachment. They can carry the drug particles properly into the lung and increase the emitted dose.⁹⁸ However, due to the limited detachment of the drug from the carrier surface, a low particle respirable fraction may be observed,^{104,106,108} though a high particle respirable fraction is also observed for smooth carriers by some researchers.^{107,118} In addition, carriers with high surface roughness and large surface area can attach higher amount of drug particles because of the large number of binding sites available.^{98,101} The drug content uniformity and stability are also better for particles with rough surfaces.^{104,107}

Crowder et al. suggested that particles with pollen-shape morphologies may have low van der Waals forces and excellent dispersion properties.²⁹ A recent study with hairy LA particles also shows better drug delivery than traditional lactose particle.¹⁰¹ The hairy surface allows the formulation to have longer time of flight (TOF) for drug liberation. Drug liberation happens due to the shear forces induced in the airways. Long TOF can improve drug liberation and deep lung deposition.¹⁰¹ Therefore, it is anticipated that particles with pollen-shape morphology would also give a better drug delivery efficiency and can be a good candidate for DPI. Moreover, carriers with larger surface area can carry larger amount of drug particles,^{98,101} which would be an added advantage of using pollen-shape drug carriers.

The objective of this study is to investigate the feasibility of using pollen-shape carriers for inhalation drug delivery. The conventionally used lactose (LA) is rock-shape and it is difficult to formulate it with large variations. Though, some studies have been focused on surface morphology (smoothness and roughness) and elongation of LA particles, the effect of other shapes (e.g. pollen-shape used in this study) was not studied.

Hydroxyapatite (HA) particles can be synthesized into a pollen-shape with a geometric size range 21.1-48.6 μm . Pollen-shape morphology is anticipated to allow the carriers to have a longer TOF for drug liberation because of the large particle size and low density. Pollen-shape HA particles are compared with traditional LA carriers of similar size range. Flowability of the particles is characterized with Carr's compressibility index (CI) and angle of slide (θ). The *in vitro* aerosolization and deposition properties of Budesonide (Bd) drug blended with pollen-shape carriers are compared to that blended with LA carriers. A relatively higher drug loading of 10:1 carrier: drug weight ratio is used in this study.

7.2 Experimental

7.2.1 Preparation of HA and LA

The preparation of HA particles has been described in the Experimental chapter (Section 3.2.1). Three types of pollen-shape HA particles are used. HA-1 is synthesized with a PSS concentration 40 g/L & at 120°C, HA-2 with PSS concentration 30 g/L at 200°C and HA-3 with a PSS concentration 40 L/min and at 150°C.

α -lactose monohydrate is used as the control carrier particles for comparison with the performance of pollen-shape HA carrier particles. In order to assess the feasibility of using a pollen-shape carrier for dry powder inhalation, the LA particles need to have

similar geometric size as the HA particles. The LA particles with size range of 38-75 μm (LA-1) and 20-38 μm (LA-2) are used.

7.2.2 Particle characterization

Synthesized particles are characterized with their size, shape & morphology, powder density, XRD diffraction, TGA isotherm and powder flowability.

7.2.3 Blending of carrier particles with Bd

Budesonide (Bd, Sigma) is used as the model drug in all through the studies to assess the behavior of the carriers in blending formulation. As the Bd particles are already in a smaller size range, they are used without any further treatment. Bd and different carriers are mixed at a 10:1 carrier: drug weight ratio. A lower drug mixing ratio of 45:1 is also prepared with LA for verification of the experimental procedure. The drug blends are also characterized with SEM.

7.2.4 Drug content, content uniformity, and drug attachment

The content uniformity of each blend is examined by analyzing the quantity of Bd in the blend. Drug attachment with the carriers and their stability are assessed by sieving.¹⁰⁴ Sieve with an opening of 20 μm is used with a mechanical shaker so that only the unattached drug particles are removed from the blends. 100 mg of each blend is placed on the sieve and shake for 30 minutes with a mechanical shaker. The sieved blends are collected from the sieve to measure the drug contents. Each assessment is performed in triplicate.

7.2.5 *In vitro* aerosolization and deposition properties

In vitro aerosolization and deposition experiments are carried out at an air flow rate of 30 L/min^{40,67,110,184,185} which is close to the normal inhalation rate of an adult. In

each experiment, 8 ml of the solvent is poured inside the pre-separator. An actuation time of 20 seconds¹¹⁰ is allowed for each capsule to completely disperse all the particles. Experimental runs are conducted in triplicate. The *in vitro* aerosolization and deposition properties of HA particles alone (without any drug) are also determined by following the same procedures.

7.3 Results and discussion

7.3.1 Particle characteristics of HA

Three types of pollen-shape HA particles are synthesized. Since the physical properties of the particles directly affect the flow characteristics during the inhalation process, it is important to have proper characterizations. The SEM images of the different HA particles produced in this study are shown in Figure 7.1. The HA particles are observed to exhibit two types of morphology, namely petal-like and needle-like. Bulk and tap densities are closely related to their morphology. Generally, a dense orientation of the surface petals or needles induces higher density (HA-1 & HA-3) and a spread orientation induces lower density (HA-2). The bulk and tap densities of the HA samples are listed in Table 7.2.

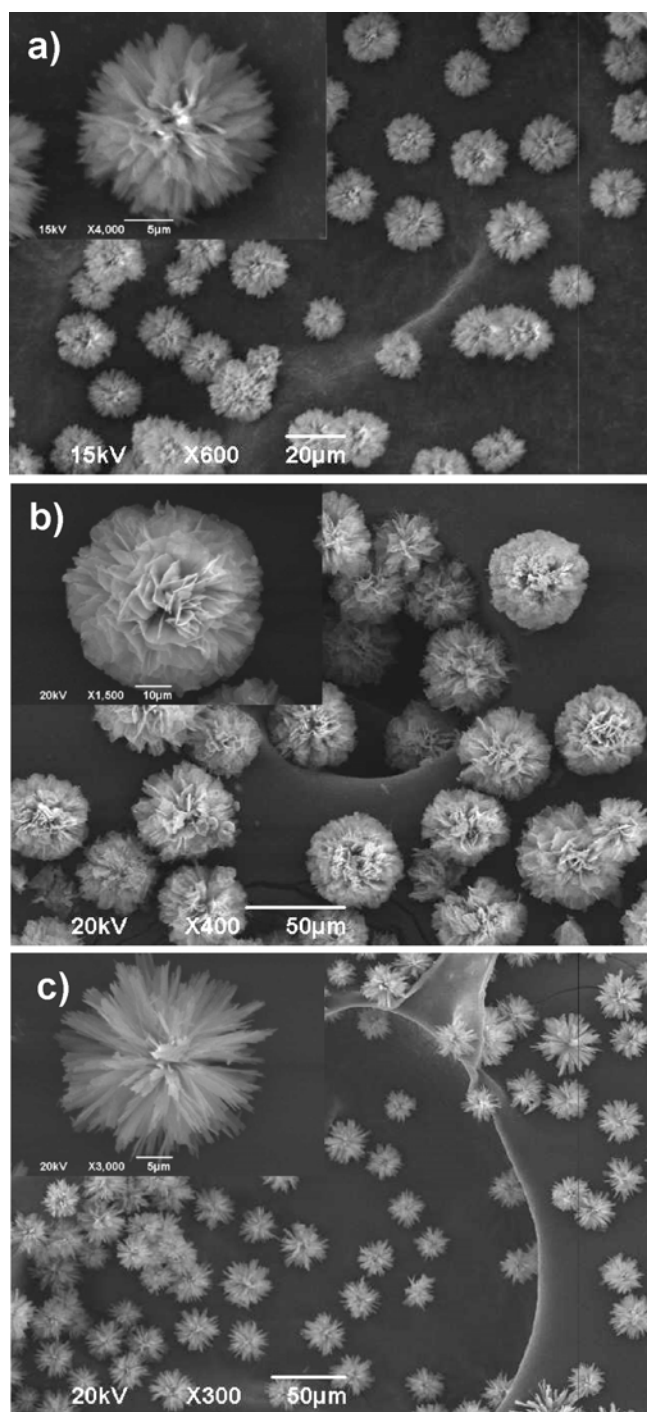


Figure 7.1 The SEM image of HA particles produced by using (a) PSS-40 g/L & urea-0.5M, at 150°C (HA-3); (b) PSS-40 g/L & urea-0.5M, at 120°C (HA-1); (c) PSS-30 g/L & urea- 0.5M, at 200°C (HA-2).

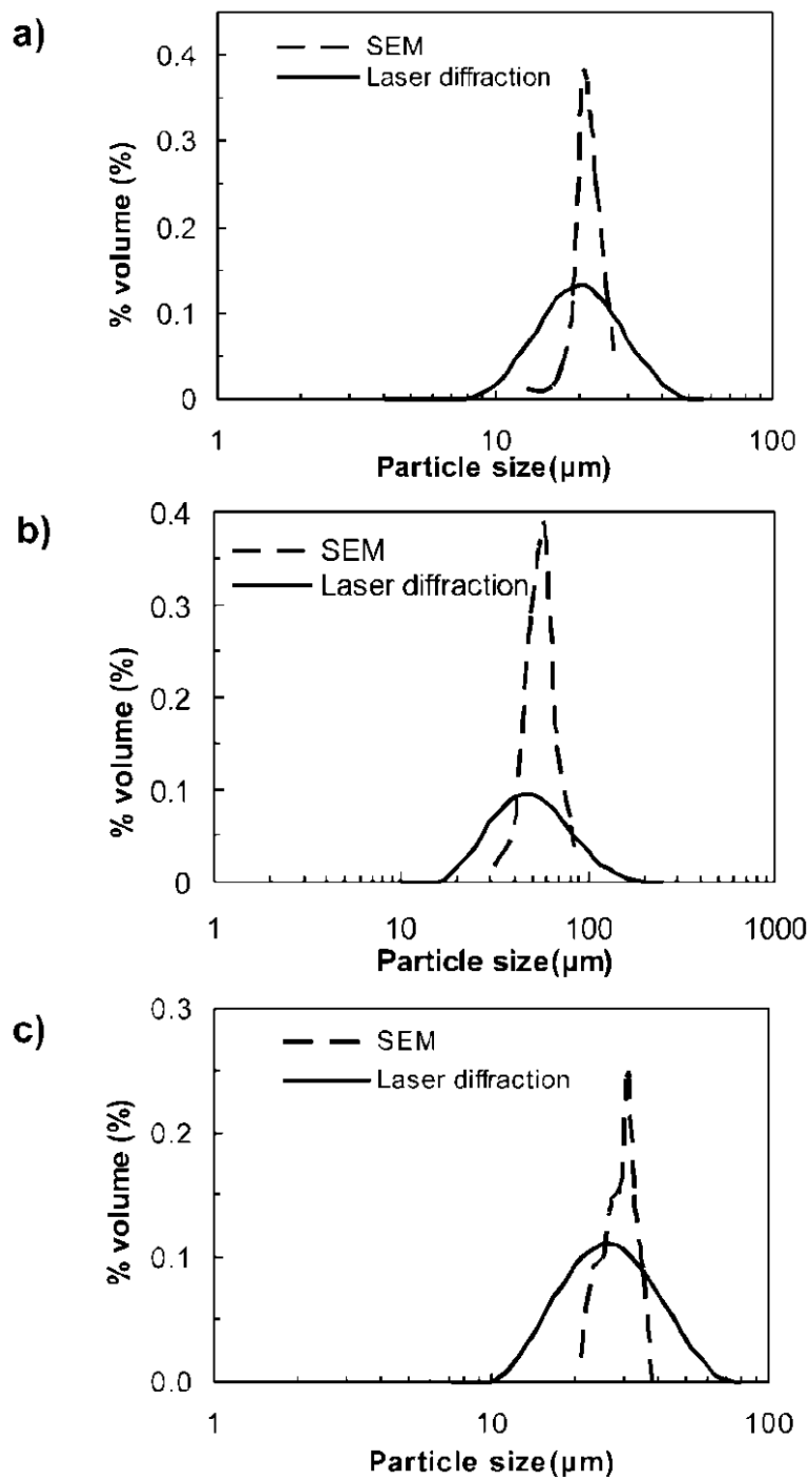


Figure 7.2 The size distribution of (a) HA-3, (b) HA-1, and (c) HA-2.

Table 7.1 Geometric and aerodynamic size of HA samples

Sample	Average dia.(SEM) (μm)	St.dev. (μm)	d(50%) (μm)	d(10%) (μm)	d(90%) (μm)	Span	d_a (calculated) (μm)
HA-3	21.1	3.30	18.6	11.1	28.9	0.96	12.89
HA-1	48.6	10.21	45.9	25.9	85.9	1.31	24.9
HA-2	27.1	4.02	24.8	15.1	41.0	1.04	12.06

The geometric and aerodynamic sizes of the particles are listed in Table 7.1. The size distributions of the HA samples are shown in Figure 7.2. The laser diffraction measurement takes into account the diffracted lights from the particles to estimate the particle size. However, for the measurement of non-spherical particles, diffracted lights depend on the orientation of the particles and the results may deviate from the actual size. Therefore particle size obtained from SEM images is also used to support and compare against the size measurement obtain from laser diffraction. It is to note that the size distributions obtained from laser diffraction measurement are volume-weighted while the size distributions from the SEM images are number-weighted. Therefore, the SEM size distributions are converted to volume-weighted distribution before comparisons are made. The conversion of number-weighted size distribution to volume-weighted size distribution requires the knowledge of particle volume. Therefore, pollen-shape particle volume is assumed to be the same as a sphere with the same diameter.

It can be seen from Table 7.1 that the SEM measured size is close to the d(50%) of the laser diffraction measured size. The minor difference is possibly due to the irregular diffraction on the particle surface. The span of all the HA particles are close to 1, which indicates a uniform size distribution.¹⁸³ The SEM measured size is used as the

equivalent diameter of the particles. The tap density is used conventionally as the particle effective density⁶⁷. The dynamic shape factor depends on the ratio of the drag force on the concerned particle and the drag force for a sphere having the same volume. The dynamic shape factor for a sphere would be 1. Since the pollen-shape would cause a higher drag than a sphere but still closely resemble a spherical shape, a dynamic shape factor of 1.1 is estimated for the pollen-shape particles. Even though the dynamic shape factor is an estimated value, the error from the estimation should have minimal effect on the calculation of the aerodynamic diameter since the aerodynamic diameter is proportional to the square root of the dynamic shape factor.

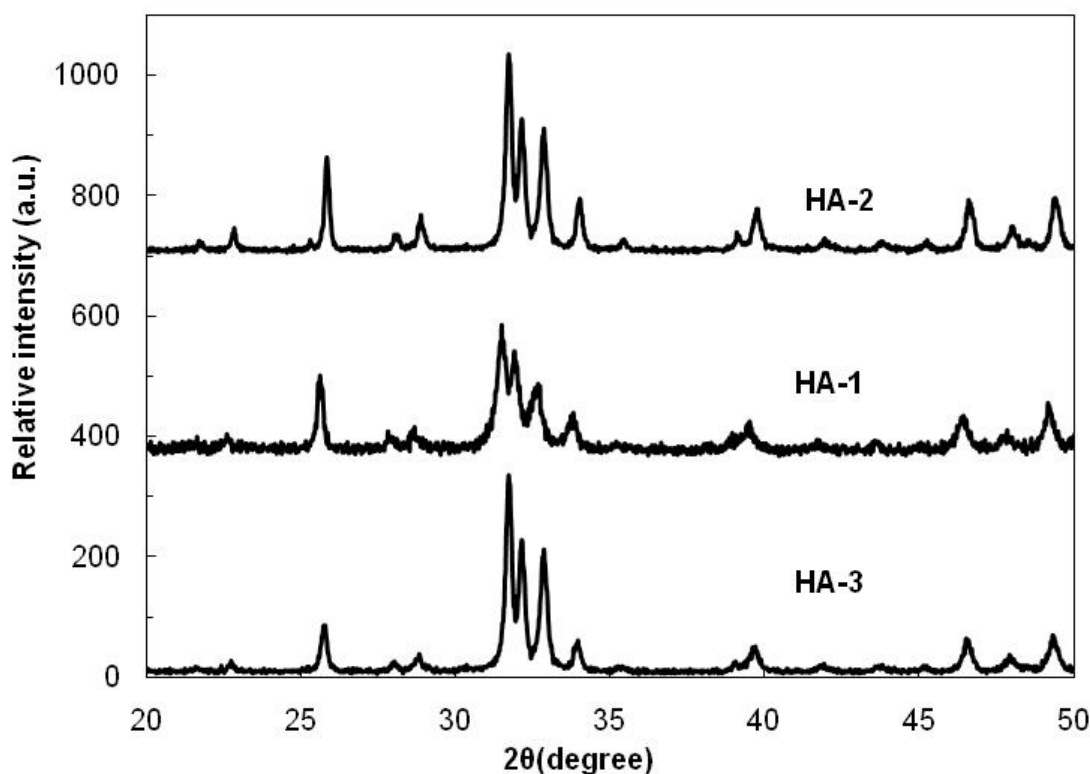


Figure 7.3 XRD patterns of the samples.

XRD pattern of the synthesized HA particles are measured to analyze their crystallographic information. The XRD analysis results of the HA particles are shown in

Figure 7.3. No preferred orientation effects were observed. The diffraction patterns are consistent with powder diffraction file (PDF) no 00-009-0432 and it indicates that all the particles exhibit hexagonal phase of HA crystalline structure. XRD results also ensure the purity of the produced samples. Crystalline form is also important for long term stability of the product.

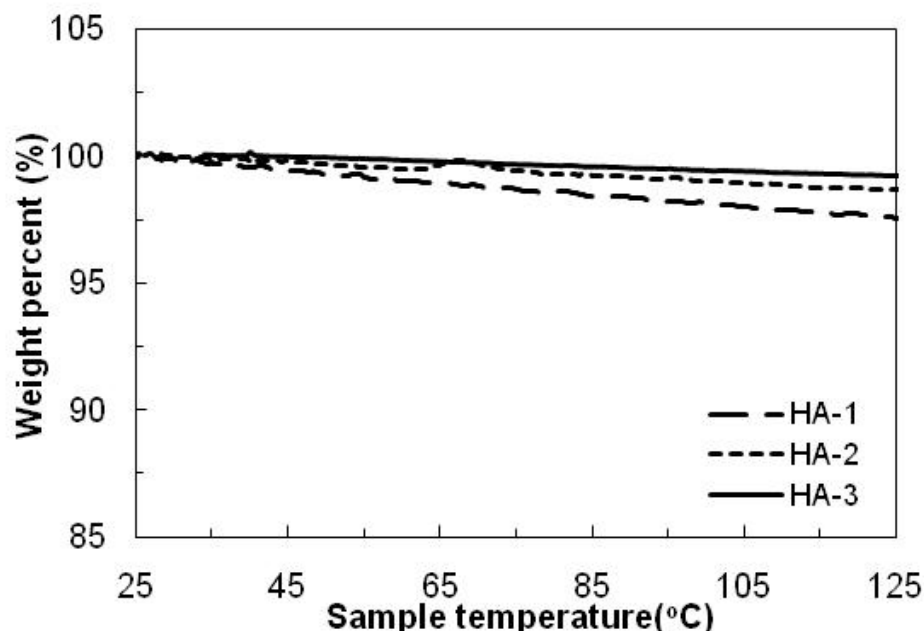


Figure 7.4 TGA spectrum of the HA samples.

Moisture content of the dry particles would hamper their dispersion and aerosolization properties. The moisture also induce agglomeration tendency of the particles. Therefore, particle surfaces are needed to be free from water molecules. The moisture content of the dry particles is analyzed by TGA. The TGA thermograms of the HA samples are shown in Figure 7.4. All the samples show negligible amount of weight loss below 100 °C, suggesting that they contain a negligible (less than 2%) amount of free water. It is reported with different sample of LA that if the free water is negligible, their inhalation properties can be compared based on their physical properties.⁹⁹

7.3.2 Particle characteristics of LA

The SEM images of the LA particles are shown in Figure 7.5. It is to note that sieving alone is not possible to completely separate all the particles. A fraction of fine particles maybe present among the larger sized ones. The bulk and tap densities of the LA samples are also listed in Table 7.2.

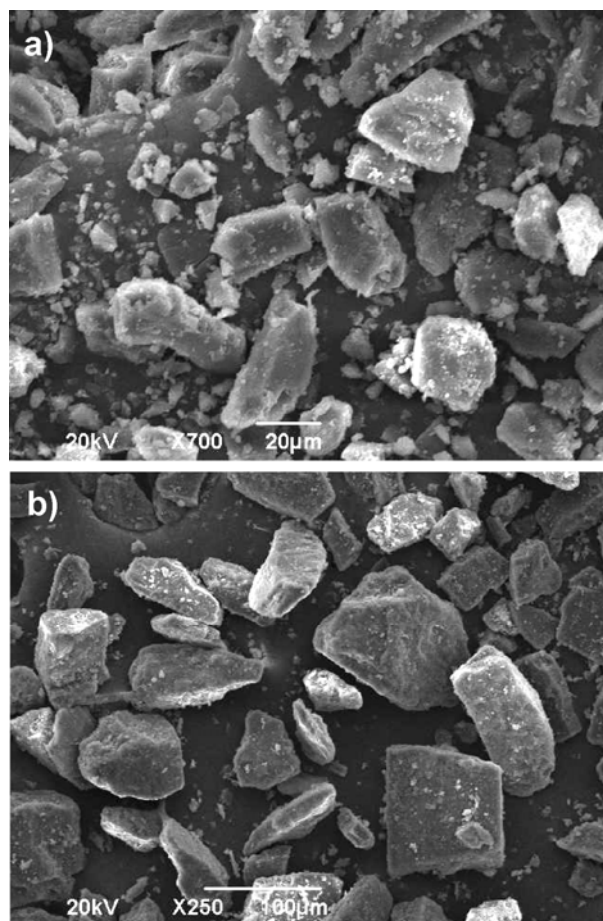


Figure 7.5 SEM images of LA with a size range of a) 20-38 μm (LA-2), and b) 38-75 μm (LA-1).

Table 7.2 Bulk and tap densities of the LA and HA samples

Sample	$\rho_{\text{bulk}}(\text{g}/\text{cm}^3)$	$\rho_{\text{tap}}(\text{g}/\text{cm}^3)$
	(n=0)	(n=2500)
HA-3	0.233	0.411
HA-1	0.215	0.289
HA-2	0.105	0.218
LA-2	0.316	0.760
LA-1	0.586	0.874

7.3.3 Powder flowability

Carr's compressibility index

Two different measurements are taken in this study to estimate the flowability of the carrier particles. Carr's compressibility index (CI) is one common method to characterize powder flow. CI value quantifies the powder flow based on the concept that compressible powders are more cohesive and therefore, experience poor flowability.⁹⁴ CI index is measured based on the difference between the bulk and tap densities of the particles. A lower CI value indicates better flowability.

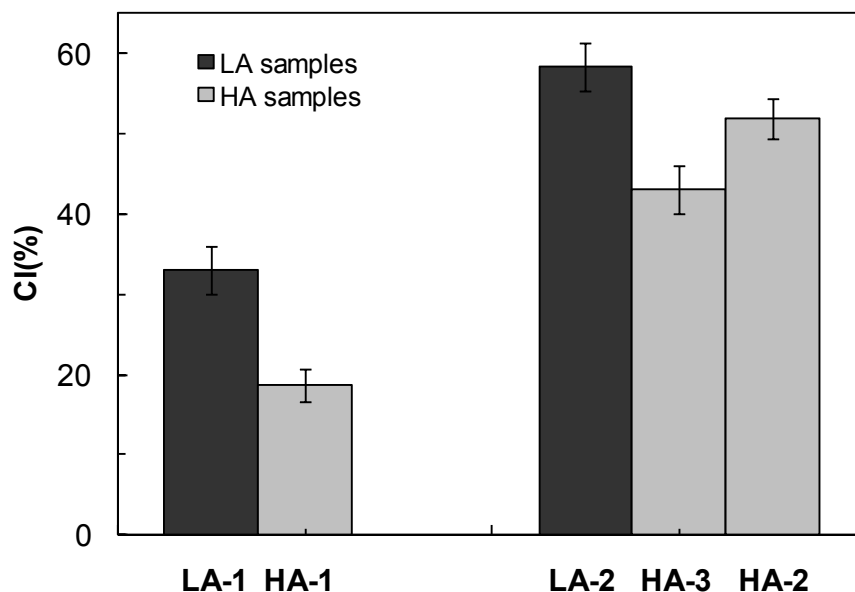


Figure 7.6 Comparison of CI values between the pollen-shape HA particles and the LA particles in similar size range.

The CI index of similar size HA and LA particles are compared in Figure 7.6. From Figure 7.6, it can be seen that particles with smaller size generally have higher CI value and lower flowability. For small size particles, aggregates are formed. Upon tapping, the aggregates may disintegrate and compact into smaller bulk volume. When the particles are large, they become less cohesive and behave as individual particles. Therefore, the particles are less compactable. HA particles for all size ranges have lower CI values than the LA particles. It is possible that the surface morphology of the HA particles increases the distance between two interacting particles and reduces the contact surface. Thus, the interparticle forces would be lower. The particles will have lower aggregation tendency and exhibit better flowability. It can also be seen in Figure 7.1 that the HA particles are well segregated. It is noted that HA-2 particles show higher CI value than HA-3 particles whereas both HA-2 and HA-3 particles have similar size. This

deviation is due to the difference in surface morphology of the HA-2 particles than the other HA samples. The HA-2 particles have needle-like surface morphology which may cause interlocking of particles. Therefore, particle rearrangement is possible for HA-2 particles and a higher CI value is observed.

Angle of slide, θ

The representation of particle flowability using CI depends on empirical understanding and sometime presents some discrepancies in characterization. Therefore, flowability of the carrier particles was also examined by the angle of slide method.¹⁰⁰ The angle of slide, θ indicates the ability of the particles to flow in the presence of shear.

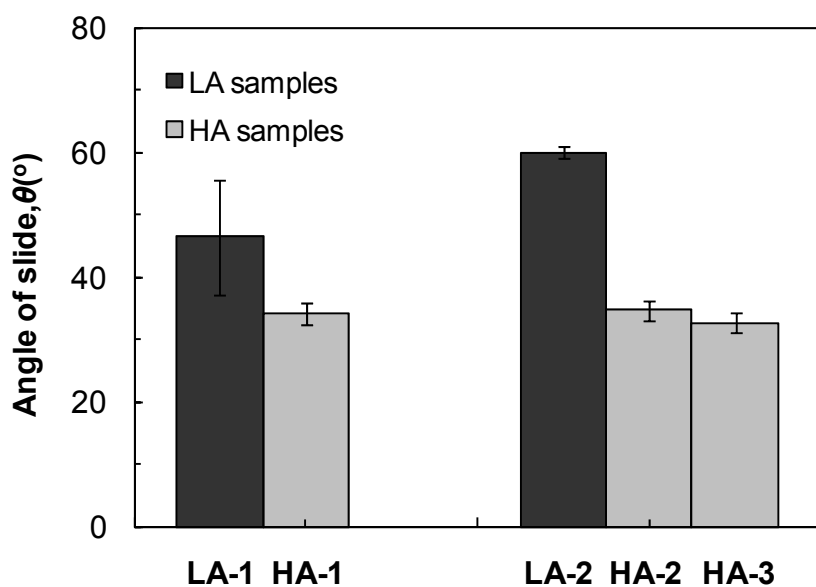


Figure 7.7 θ values of the pollen-shape HA particles with LA particles of similar size range.

In Figure 7.7 the flowability of HA particles and LA particles are compared in terms of θ . Lower θ values show better flow behavior. Therefore, it can be seen that the flowability of the HA particles is significantly higher than that of the LA particles. The fine particles present among the LA particles would aggregate with larger size particles

and reduce the flowability. On the other hand, HA particles are observed to have lower tendency of forming aggregates and separate into discrete particles easily. The surface morphology of the HA particles appear to help reduce the particle-particle and particle-surface interaction and increase the effective separation distance between them. In general, particles with higher surface asperities have lower van der Waals force⁶⁵, hence low aggregation tendency. Thus, the particles are prone to flow even under slight shear. Since the interparticle forces are weak, particles can slide on other particles resulting in a better flowability. It has been reported that irregular particles may experience mechanical interlocking which can lower their flowability. However, this phenomenon is not observed among the pollen-shape particles.

7.3.4 Drug content and content uniformity

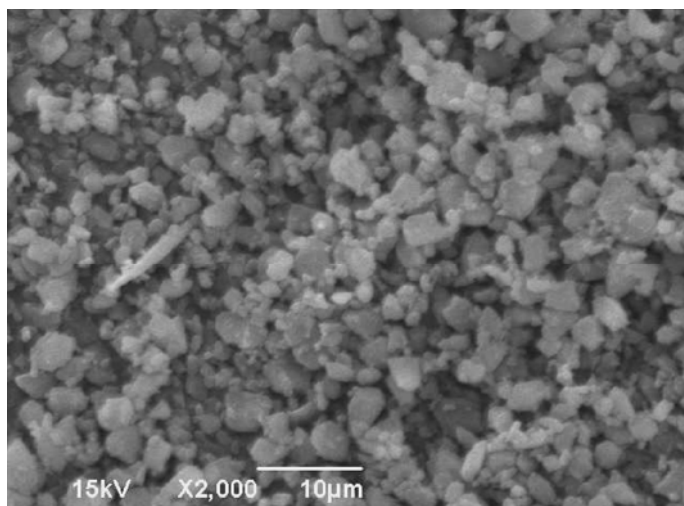


Figure 7.8 SEM image of Budesonide.

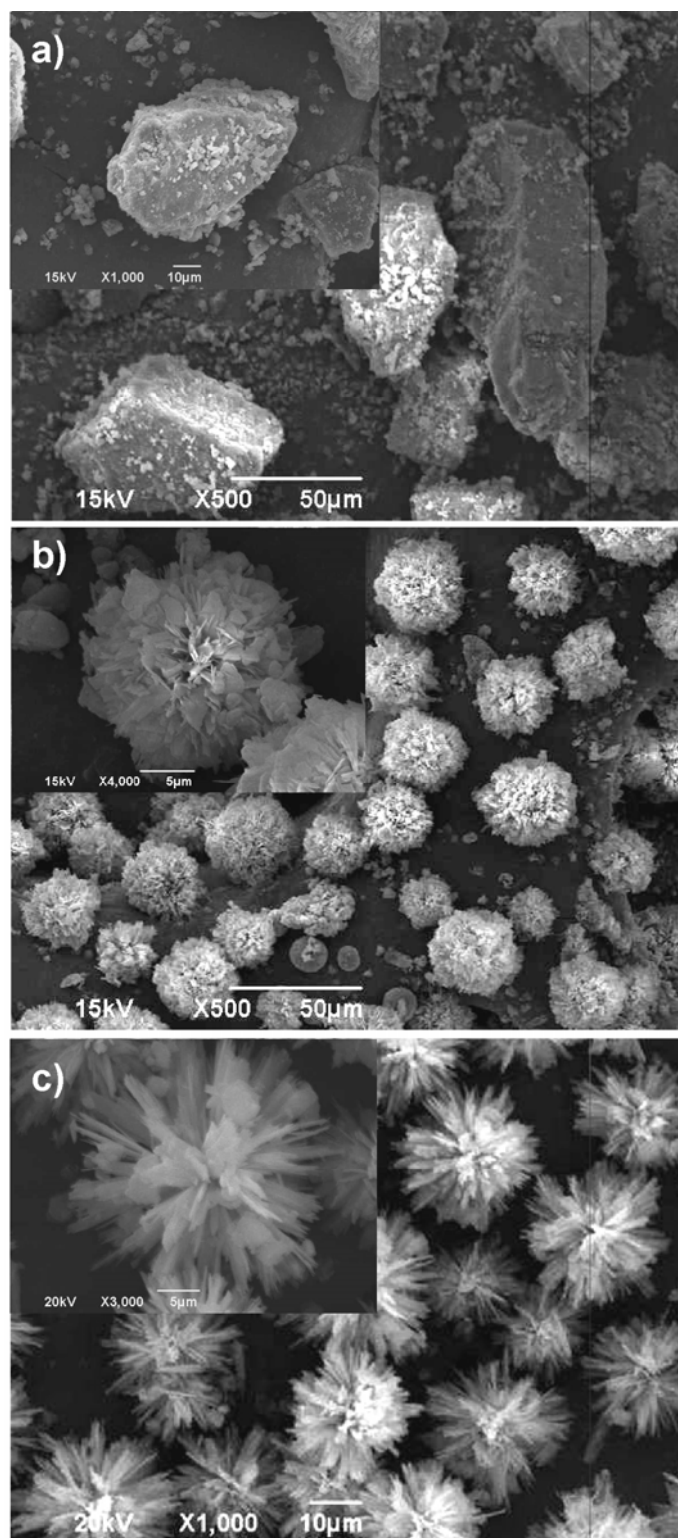


Figure 7.9 SEM image of a) blend of Budesonide with LA-1 carriers, b) blend of Budesonide with HA-1 carriers, and c) blend of Budesonide with HA-2 carriers.

The LA-2 particles contain a certain amount of fine particles which cannot be separated by sieving. The presence fine particles would influence the inhalation performance of the carrier.⁹⁶ Though LA-2 and HA-3 are of similar size range, the additional factor would make the comparison very difficult. Since HA-1 has similar surface morphology as HA-3, the comparison between HA-1 and LA-1 would be sufficient to demonstrate the feasibility of using pollen-shape drug carrier for dry powder inhalation. Unlike the HA-1 particles, HA-2 particles have needle-like surface morphology with a size range close to the LA-1 particles. Therefore, HA-2 is also used for the ACI experiments for the analysis of morphology effect.

The SEM image of the Bd is shown in Figure 7.8. The Bd used has an average size of $2.5 \pm 1.1 \mu\text{m}$. Bd is blended with LA-1, HA-1 and HA-2 carriers separately at a 10:1 carrier: drug weight ratio. The SEM images of the blended mixtures are shown in Figures 7.9(a-c). It can be seen from Figure 7.9(a) that Bd particles are attached to the surface of the LA-1 carriers but a fair amount of Bd remains unattached. As shown in Figures 7.9(b) and 7.9(c), there is significantly lower amount of unattached Bd for HA-1 and HA-2 carriers. It is anticipated that the petal-like and needle-like surface morphology of HA-1 and HA-2 carriers are the main contributors of the better drug attachment than LA carriers.

The homogeneity of the blends is represented by the coefficient of variation (CV) of the Bd content in the blends. The CV is the ratio of the standard deviation to the average of the Bd content in various random samples of the same blend. The average drug content and CV for the three blends after mixing are listed in Table 7.3. It is found that LA-1 carriers have the lower weight fraction of drug compared to HA-1 and HA-2

carriers. Though the mixing proportion of Bd and carriers are the same for all three carriers, the drug content of Bd in the blend with LA-1 carriers is lower than that in the blend with both HA carriers. Due to the low adhesion of Bd on the LA-1 surface, a certain amount of Bd is attached to the surface of the vial and lost during the mixing process. All the blends are considered uniform as the CV of Bd in the blends are found to be within 10%.

7.3.5 Drug attachment

From the SEM images in Figure 7.9, it can be seen that there are excess unattached drug particles after mixing. Though the mixing proportion of Bd with the carrier particles is same for all the blends, differences in the appearance of the drug particles in the SEM images are found. Some of the drug particles may remain unattached to the carrier particles in the blend. The unattached drug particles (or drug aggregates) may lose in the capsule during the use in the inhaler. Therefore, the adhesion characteristic is used as a rough estimation of the drug attachment and subsequent drug delivery performance of the carrier particles. In this technique, the unattached drug particles are separated from the blends. The concentration of drug in the blend before and after the separation would represent the unattached drug fraction in the blend.

To analyze what percentage of drug particles attached to the carriers, the mixtures are sieved by using a sieve which have the opening smaller than the carrier particles but larger than the drug particles. It is expected that the unattached drug particles would be separated from the mixture. However, the separation is not easy, because the drug particles are very cohesive and they attached to the sieves very easily. Moreover, by sieving some of the attached particles can also be detached. However, by collecting the

mixture after sieving and measuring their concentration, a rough estimation of the drug attachment with the carrier particles can be obtained from the reduction of the drug content in the mixture by sieving.

Table 7.3 Attachment of Budesonide drug with LA and HA carriers

Carrier	After mixing		After sieving		Reduction in drug content (wt %)
	Wt fraction of drug (wt %)	CV (%)	Wt fraction of drug (wt %)	CV (%)	
LA-1	7.85±0.41	4.46	3.73±0.16	3.76	52.5
HA-1	8.91±0.89	8.57	6.51±0.37	4.84	26.9
HA-2	9.01±0.40	3.80	7.90±0.90	9.84	12.3

The average Bd content in the blends after sieving are presented in Table 7.3. It is found that the percent reduction of Bd by sieving is more than 50% for the blend with LA-1 carriers. This result is in agreement to Figure 7.9(a) that large amount of unattached Bd is observed in the blend with LA-1 carriers. HA-3 carriers showed the best drug attachment among the three carriers used. The reduction in Bd after sieving is as low as 12.3 wt%. The HA-1 carriers have a loss of about 26.9% of Bd after the sieving process. HA-2 carriers with needle-like surface morphology provide larger surface area and binding sites for attachment of drug particles. It can reduce the loss of drug particles in the storage container and in the inhaler during inhalation.

7.3.6 *In vitro* aerosolization and deposition properties

The *in vitro* aerosolization and deposition behavior of Bd blended with different carriers without sieving are studied in an ACI at 30 L/min. The measurements are presented as emitted dose (ED), fine particle fraction (FPF) and dispersibility. As

particles are discharged from the capsule, a fraction of the particles will be dispersed and flow with the gas stream. Some of the particles in the inhaler will be picked up and entrained by the gas flow while the rest of the particles will remain in the inhaler and the capsule. Therefore ED can be considered to account for both aerosolization property and flowability of the particles. FPF is a commonly used index to represent deposition properties of aerosol particles. Particles that have less deposition at the initial regions and able to reach the lower stages of the impactor would have higher FPF. Dispersibility is defined as the amount of the particles deposited in stage 2 or lower as a percentage of ED.

The FPF of Bd in a blend with LA at a lower drug mixing ratio of 45:1 is determined at an inhalation rate of 30 L/min. A FPF value of 16% is found and it is comparable to the FPF values reported in the literature using low drug loading.¹¹⁰ Therefore, the experimental method used in this study is acceptable. The ED, FPF and dispersibility results of the three blends are listed in Table 7.4. It can be seen from the table that both HA blends have higher ED than the LA blend. Substantial amount of Bd are unattached to the LA carriers and lost in the inhaler. The two HA carriers have good flowability and allow good adhesion of Bd. Bd will be carried into the ACI together with the carriers. A comparison of the two HA carriers show that HA-1 carriers give higher ED and FPF than HA-2 carriers. Despite the fact that HA-2 particles have high drug attachment capability, their emission is slightly lower due to their lower flowability. Their drug liberation is also limited. Therefore, their FPF is lower than that of HA-1 carriers. The high ED of HA-1 carriers also contribute to a higher FPF. Among the carriers used in this study, HA-1 carriers are the most ideal for inhalation drug delivery.

Table 7.4 Deposition of Budesonide from different blends in the cascade impactor at 30 L/min

Blend	ED (%)	FPF (%)	Dispersibility (%)
LA-1	77.7±2.9	3.1±0.59	3.9±0.72
HA-1	89.6±2.4	10.9±0.57	11.5±0.28
HA-2	84.7±5.4	6.5±0.57	7.7±1.13

Though FPF indicates the amount of drug particles that would be able to reach the target area, it is a strong function of the ED. Therefore, dispersibility is sometimes used to account for the deposition property irrelevant to the ED. A high dispersibility indicates that it is possible to increase the FPF by improving the ED. The dispersibility results of Bd blend with the three carriers used again indicate that HA-1 carriers have the best performance in terms of drug detachment and deposition.

The regional deposition of Bd in the ACI can give a better understanding on the effect of different carriers on drug delivery. Figure 7.10 shows the regional deposition of Bd in the ACI. The blend with LA-1 carriers showed the highest deposition in the inhaler and throat regions. The blends with HA-1 and HA-2 carriers have relatively low drug deposition in these two regions. It indicates that HA carriers exhibit favourable flowability and dispersion even after the blending with drugs. Moreover, the high percentage of Bd unattached to the LA-1 carriers may adhere to the inhaler wall and causes the high deposition percentage in the inhaler region. When carriers are deposited in the throat region, there is insufficient flight time for the drug particles to be liberated from the carrier surface. Thus, drug particles often deposit together with carrier particles in the throat section. Previous results show that the pollen-shape HA particles have lower

deposition at the throat region than LA particles. Therefore, it is reasonable to obtain a higher deposition of Bd in the throat region for the blend with LA-1 carriers than that for the blends with HA-1 and HA-2 carriers. In most cases, carriers would be deposited largely at the preseparator region. The highest deposition percentage of Bd observed at the preseparator region indicates that some Bd are still attached to the carriers and deposited along with the carriers. A high deposition in the preseparator and the initial stages would lead to a low FPF especially under high drug mixing ratio (10:1 drug: carrier) and low inhalation rate (30 L/min)⁴⁰ used in the study.

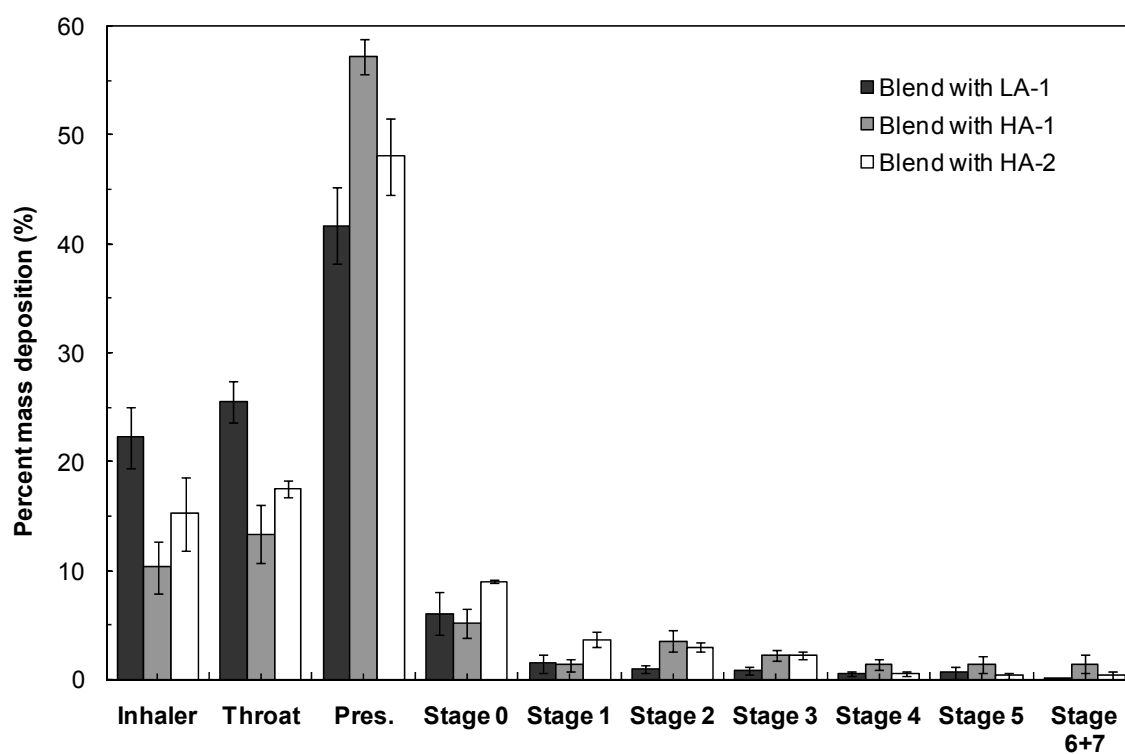


Figure 7.10 Regional deposition of Budesonide in the ACI when blended with different carriers.

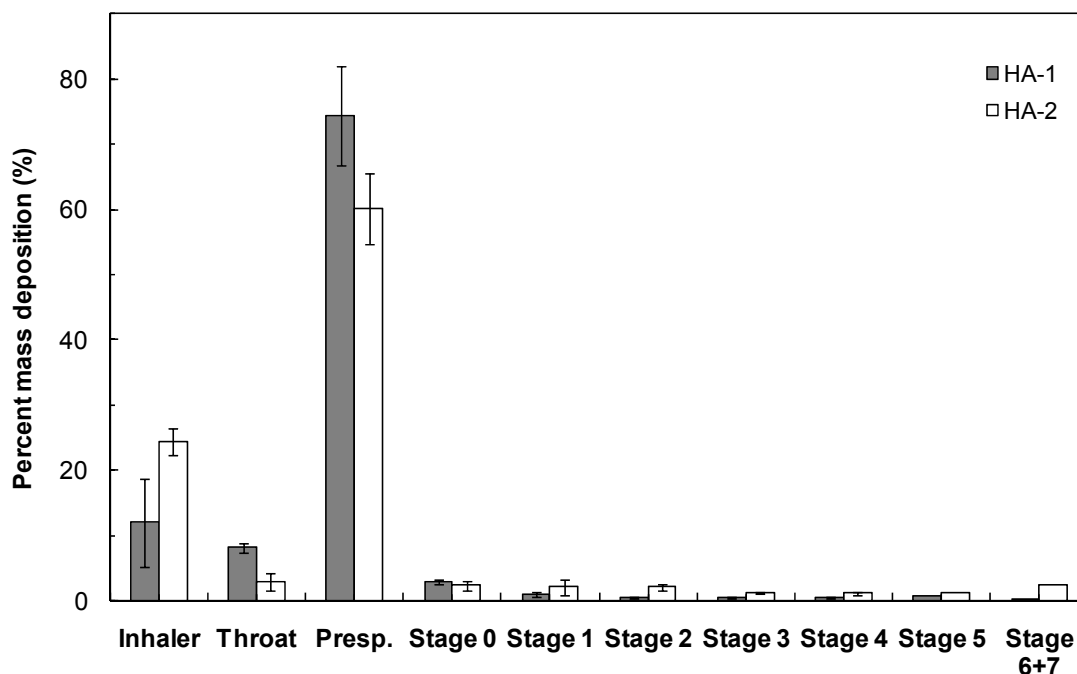


Figure 7.11 Regional deposition of HA-2 and HA-3 particles in the ACI.

Though pollen-shape HA carriers show better drug delivery than traditional LA carriers, it would be beneficial to know how the HA carriers work. The regional deposition of HA carriers as single formulation in the ACI was examined and the result is showed in Figure 7.11. It can be seen that the deposition of the HA carriers are low in the inhaler and throat regions. The deposition of HA-1 and HA-2 carriers in stage 0 and stage 1 are similar. However, Bd deposition in these two stages is higher for the HA-1 blend than the HA-2 blend as shown in Figure 7.10. This indicates that more Bd is liberated from the HA-1 carriers than from HA-2 carriers. The liberated Bd would be able to form a good dispersion and continue to travel to the lower stages (from stage 2 and lower). HA-2 carriers have smaller size than HA-1 carriers and able to travel further than HA-1 carriers. However, the limited drug liberation from HA-2 carriers would cause the Bd to deposit together with the carriers. The overall delivery of Bd in the lower stages using

HA-2 carriers are thus not as efficient as using HA-1 carriers where the Bd can travel in dispersed form. Despite the fact that only two pollen-shape carriers are used in this study, it is evidenced that a pollen-shape surface can improve the drug loading capacity, emitted dose and fine particle fraction of drug for dry powder inhalation

7.4 Concluding remarks

HA particles are synthesized into pollen-shape for drug carrier application in dry powder inhalation. The HA particles synthesized have a size range 21.1-48.6 μm and tap density 0.21-0.41 g/cm^3 . The pollen-shape HA carriers show better flowability in terms of CI and θ than conventional LA carriers of similar size. Bd is used as model drug and blend with different carriers. A higher drug attachment is found possible on the pollen-shape surface. The drug concentration in the HA blends after sieving is more than double compare to the drug concentration in the LA blends (LA=3.73%, HA=6.51-7.90%). Bd blended with pollen-shape carriers are also observed to have better ED (LA=77% and HA=84-90%) and FPF (LA=3% and HA=6.5-11%) than that blended with traditional LA carriers at a flow rate of 30 L/min. It is evidenced that pollen-shape HA carriers can be a promising drug carrier in dry powder inhalation.

Chapter 8 INHALATION PERFORMANCE OF POLLEN-SHAPE CARRIER IN DRY POWDER FORMULATION WITH DIFFERENT DRUG MIXING RATIO: COMPARISON WITH LACTOSE CARRIER

8.1 Introduction

Pollen-shape carrier particles are found to have better static and dynamic flowability, drug attachment capacity and *in vitro* aerosolization and deposition behavior than conventional lactose carriers with similar size range. In this chapter, the effect of drug mixing ratio and inhalation flow rate on the delivery performance of the pollen-shape carrier particles is studied with the comparison of lactose carrier particles.

Large carrier particles have relatively low interaction force and they can be dispersed easily. The drug particles attached to the carrier surfaces can then be carried from the inhaler to the human oral airways. As the mixture flows in the human airways, the drugs can detached from the carriers. Drug liberation occurs due to the shear forces induced in the airways. Therefore, both high shear force (induced by high inhalation rate)⁴⁰ and high TOF (long time for drug liberation)¹⁰¹ can improve drug liberation and deep lung deposition. The carriers would eventually deposit in the upper airways while the detached drug particles can travel further to the lower airways. The physical

characteristic of the carrier particles is thus a very important factor for efficient drug delivery. It is important to investigate the effect of carrier particle characteristics on the drug attachment and detachment for better control and improved drug deposition in the lower airways.

It is presented in the previous chapter that pollen-shape HA carriers are capable of high drug loading. Though pollen-shape particles show their potential as drug carriers, that result is only valid for a single drug mixing ratio at low flow rate. The drug loading and delivery performance of the pollen-shape carrier particles may vary with drug mixing ratio and inhalation flow rate. In this study, the effect of drug attachment and inhalation flow rate on the aerosolization and deposition properties of the pollen-shape HA carriers are investigated for different drug mixing ratio. HA particles are synthesized into pollen morphology with a geometric size range 48.6 μm . The *in vitro* aerosolization and deposition properties of Bd blended with pollen-shape HA carriers are compared to that blended with traditional LA carriers with three different drug mixing ratio condition at 30 and 60 L/min inhalation flow rate.

8.2 Experimental

8.2.1 Preparation of HA & LA

Pollen-shape HA-1 with a PSS concentration 40 g/L & at 120°C is used in this study. α -Lactose monohydrate is used as the control carrier for comparison with the performance of pollen-shape HA carriers. The lactose particles with a size range 38-75 (LA-1) μm are used.

8.2.2 Particle characterization

Synthesized particles are characterized with their size, morphology, powder density, XRD diffraction, TGA isotherm and BET surface area.

8.2.3 Drug content, content uniformity and drug attachment

Bd and different carriers are mixed at carrier to drug weight ratio of 2:1, 10:1, and 45:1. Blends are characterized with SEM images and physical appearance of the mixture vials. The content uniformity of each blend is examined by analyzing the quantity of Bd in the blend. Drug attachment capacity of each carrier is evaluated using sieving method that is discussed in the previous chapter (Section 7.2.3).

8.2.4 *In vitro* aerosolization and deposition properties

Pre-sieved blends are used in the ACI experiments.^{40,98,99,104,107,110,113} Experiments are carried out with a pre-separator at air flow rates of 30 L/min and 60 L/min. 10 ± 0.3 mg of each blend is loaded into the capsule for a run. An actuation time of 4 sec and 8 sec are used for flow rates of 60 and 30 L/min, respectively, for each capsule to completely disperse all the particles. Experimental runs are conducted in triplicate.

8.3 Results and discussion

8.3.1 Particle characteristics of HA

The SEM image and physical properties of the HA-1 particles are shown in Figure 8.1 and Table 8.1 respectively. It can be seen that the particles have a petal-like surface morphology. Particle tip to tip distance is measured from the SEM images and the distance is taken as the particle size. It is to note that the size distributions measured from laser diffraction measurement are volume-weighted while the size distributions measured from the SEM images are number-weighted. Therefore, the SEM size distributions are

converted to volume-weighted distribution before comparisons are made. The conversion of number-weighted size distribution to volume-weighted size distribution requires the knowledge of particle volume. The volume of pollen-shape particle is assumed to be the same as a sphere having the same diameter. Hence, it can be seen from Table 8.1 that the SEM measured sizes are slightly higher than $d(50\%)$ of the laser diffraction measured size. The span of the HA-1 particles is close to 1, which indicates a uniform size distribution¹⁸³. The particle aerodynamic diameter, d_a is also shown in the table.

Table 8.1 Physical characteristics of the HA sample

Sample	HA-1
Average dia.(SEM) (μm)	48.6
St.dev. (μm)	10.21
$d(50\%)$ (μm)	45.9
$d(10\%)$ (μm)	25.9
$d(90\%)$ (μm)	85.9
Span	1.31
ρ_{bulk} (g/cc) (n=0)	0.215
ρ_{tap} (g/cc) (n=2500)	0.289
d_a (μm)	24.9
Specific Surface area (m^2/g)	17.1

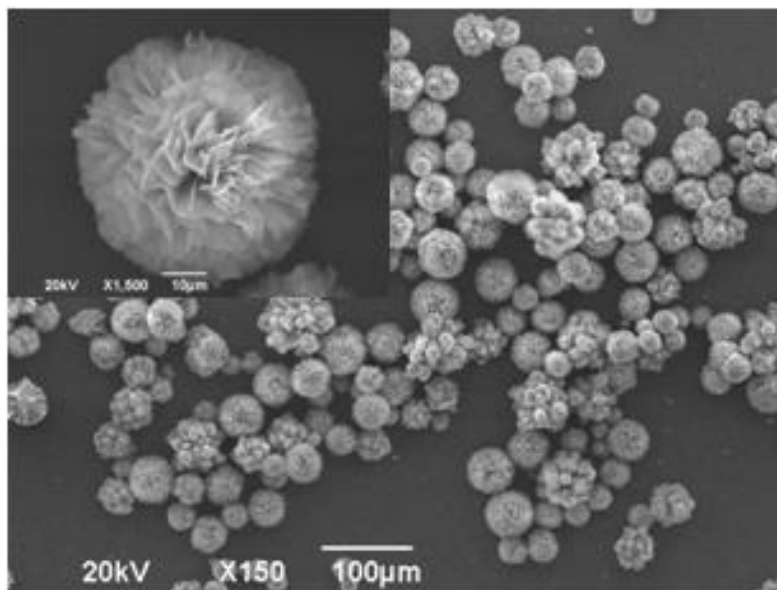


Figure 8.1 The SEM image of HA-1

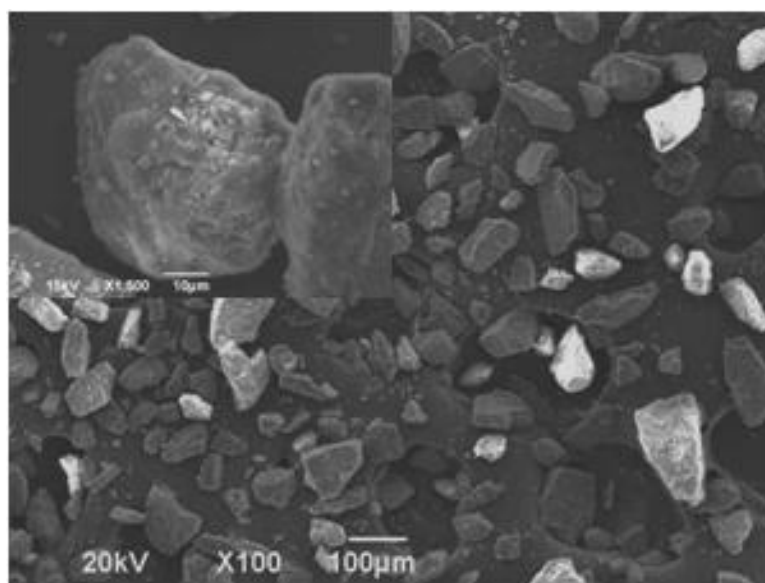
The XRD and TGA results of the HA-1 particles are presented in Chapter 7. Diffraction result shows a high purity of HA-1 sample. TGA result shows that the sample contain negligible amount of free water and the hygroscopic effect on the inhalation behavior of the sample can be considered as negligible.⁹⁹.

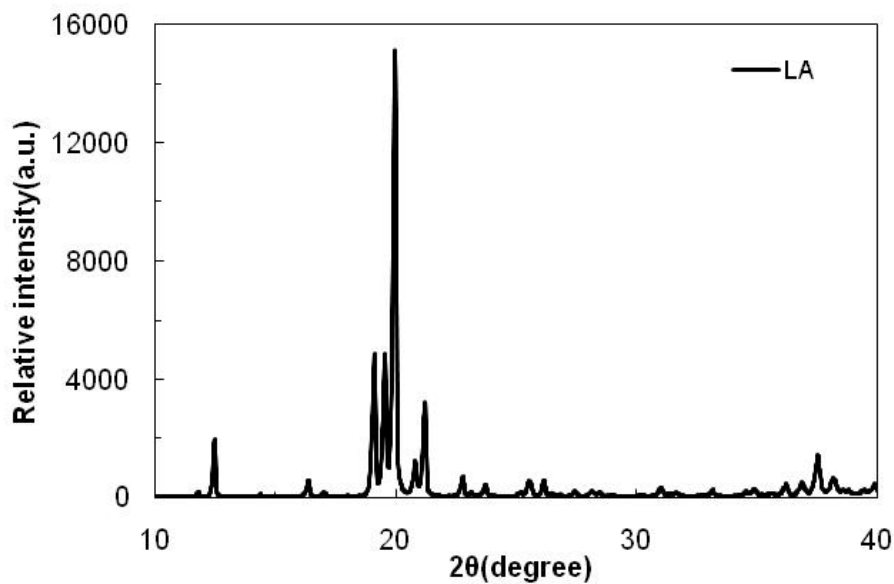
8.3.2 Particle characteristics of LA

It can be seen from the SEM image of the sieved LA-1 sample in Figure 8.2 that there is negligible amount of fine particles present. The physical properties of the sample are listed in Table 8.2. The LA-1 particles have higher tap density than both HA-1 particles, which would also translate to a higher aerodynamic diameter than the HA particles.

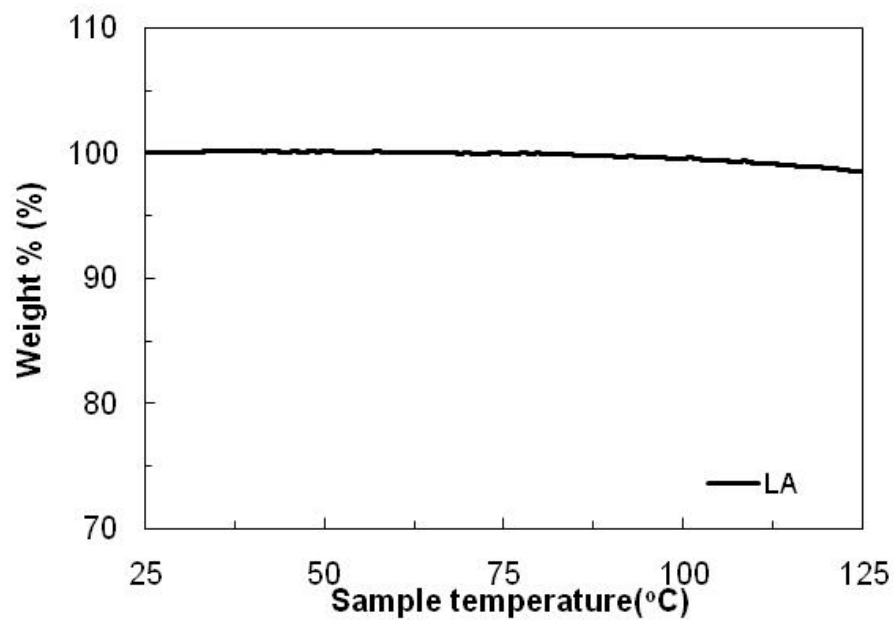
Table 8.2 Physical properties the LA particles

Sample	LA-1
Size range (μm)	38-75
ρ_{bulk} (g/cm^3) (n=0)	0.586
ρ_{tap} (g/cm^3) (n=2500)	0.874
Specific Surface area (m^2/g)	9.45

**Figure 8.2** SEM image of LA monohydrate with a size range of 38-75 μm .



(a)



(b)

Figure 8.3 (a) XRD pattern, (b) TGA spectrum of LA-1.

XRD and TGA analysis are also performed for the LA-1 sample and the result is shown in Figure. 8.3. The XRD pattern shows a high purity and crystallinity of the LA-1

sample (consistent with PDF no 00-030-1716 monoclinic). TGA spectrum also shows that the weight loss of the sample due to moisture content is negligible within 100 °C.

8.3.3. Drug blending

Bd is used as the model drug in this study. These particles have an average size of $2.5 \pm 1.1 \mu\text{m}$. Bd is blended with carriers at a drug mixing ratio of 2:1, 10:1 and 45:1 (carrier: drug) in small transparent vials. The appearance of the blended Bd is compared with that of pure carrier particles. The main reason observed for the cloud of the vial is the attachment of the drug particles. It is a qualitative representation of the drug attachment with the carrier. When the drug particles easily attach to the carrier, a lesser amount is left to attach to the vial wall and clear vial is found. The qualitative representation shows the attachment capability of the carrier particles depending on the accessible surface area by the drug particles. The vials with the carrier only and carrier with drug formulations are shown in Figure 8.4. It can be seen from Figure 8.4(a) that a substantial amount of particles are attached to the wall of the vials for all the blends with LA-1 carriers. Since there is no obvious attachment of particles to the vial surface, it is reasonable to consider all the attached particles to be the Bd. As shown in Figure 8.4(b), obvious Bd attachment on the vial surface is only observed at high drug mixing ratio of 2:1 for HA-1 carriers.

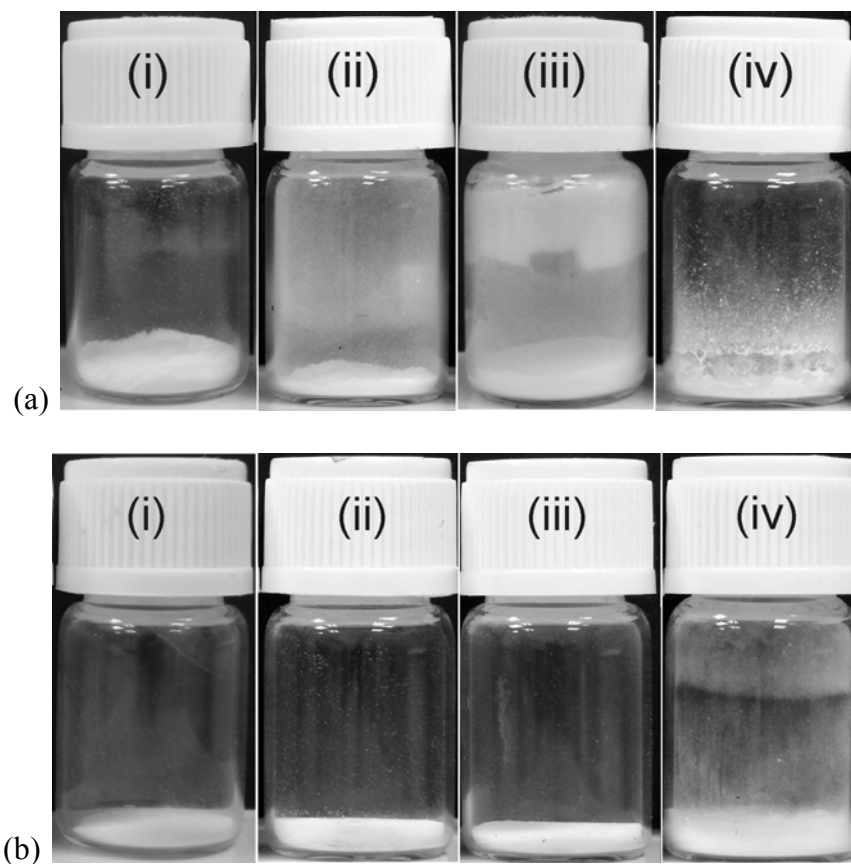
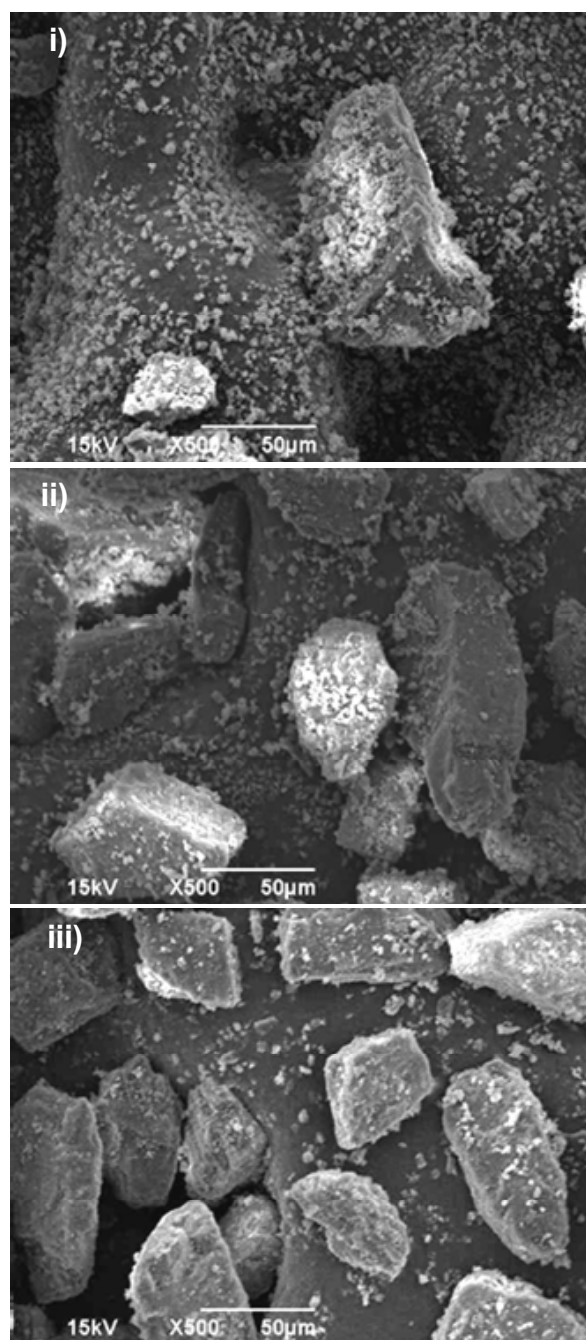
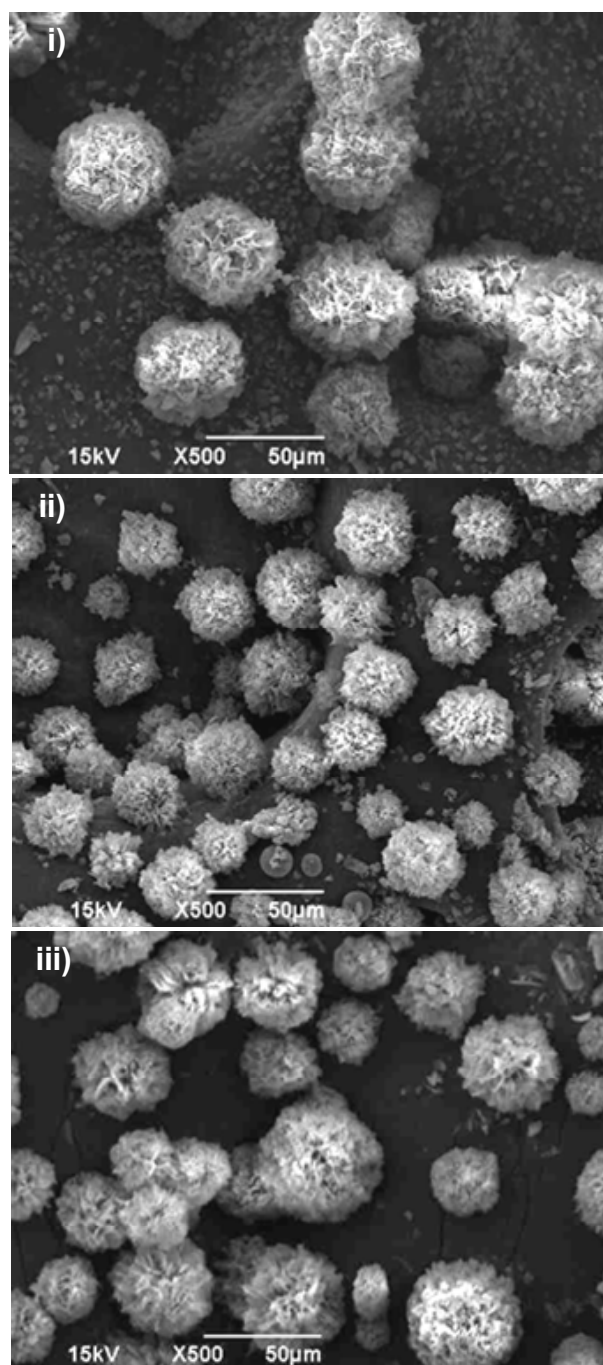


Figure 8.4 Comparisons of the blends in the mixture vials, with different carrier to drug mixing ratio (w/w). a) LA-1 carrier (i) without drug; with wt ratio (ii) 45:1, (iii) 10:1, (iv) 2:1, b) HA-1 carrier (i) without drug; with wt ratio (ii) 45:1, (iii) 10:1, (iv) 2:1.



(a)

Figure 8.5 Comparisons of the blends of the three carriers with Bd with different carrier to drug mixing ratios (w/w). a) LA-1 carrier with ratio (i) 45:1, (ii) 10:1, (iii) 2:1, b) HA-1 carrier with ratio ((i) 45:1, (ii) 10:1, (iii) 2:1.



(b)

Figure 8.6 Cont.

The blends are also characterized by SEM. The SEM images for the blends with different drug mixing ratio are shown in Figure 8.5. A comparison of the SEM images of the carriers (Figure 8.1 & 8.2) and Bd (Figure 7.8) indicates that the small particles in

Figure 8.5 would be Bd and the large particles would be the carriers. The SEM images show consistent results compared with the observation of drug attachment on vial surfaces. As shown in Figure 8.5(a), a substantial amount of unattached Bd can be seen in the blend with LA-1 carriers for all drug mixing ratios. For HA-1 carriers, significant amount of unattached Bd can only be seen at high drug mixing ratio of 2:1 indicated in Figure 8.5(b). It is believed that the pollen morphology of HA-1 carriers is the main contributor of the better drug attachment than LA-1 carriers. The petal-like surfaces allow the attachment of larger amount of Bd compared to the relatively flat LA-1 surfaces.

8.3.4 Blending homogeneity

The homogeneity of the blends can be represented by the coefficient of variation (CV) of the Bd content in the blends. The CV is the ratio of the standard deviation to the average of the Bd content in various random samples of the same blend. The average drug content and CV for the blends after mixing are listed in Table 8.3. It is found that the blend homogeneity increases with an increase in drug mixing ratio for HA-1 carriers while the opposite is observed for LA-1 carriers. The opposite effect of drug mixing ratio on the blend homogeneity of the carriers is anticipated to be due to the number of sites available for drug attachment. For LA-1 carriers, there is limited number of sites and they will be occupied by the drug particles easily at low drug mixing ratios. An increase in drug mixing ratio would thus reduce the blend homogeneity. On the other hand, HA-1 carriers have large number of sites. During the blending step, drug particles will be attached to the first available site and at low drug mixing ratio, the attachment may not be uniform. As the drug mixing ratio is increased, the sites would be occupied more uniformly and the blend homogeneity is increased. Though the blend homogeneity of the

samples exhibits some variations, all the CV values are lower than 8%. Therefore, the blends are considered as homogenous.^{100,104,113}

Table 8.3 Average Bd content and homogeneity of the blends

Wt ratio (Carrier: drug)	Carrier	Wt fraction of drug (wt %)	CV (%)
2:1	LA-1	23.0	7.87
	HA-1	22.5	0.26
10:1	LA-1	7.9	2.99
	HA-1	8.9	2.14
45:1	LA-1	1.73	0.95
	HA-1	1.81	6.26

8.3.5. Drug attachment

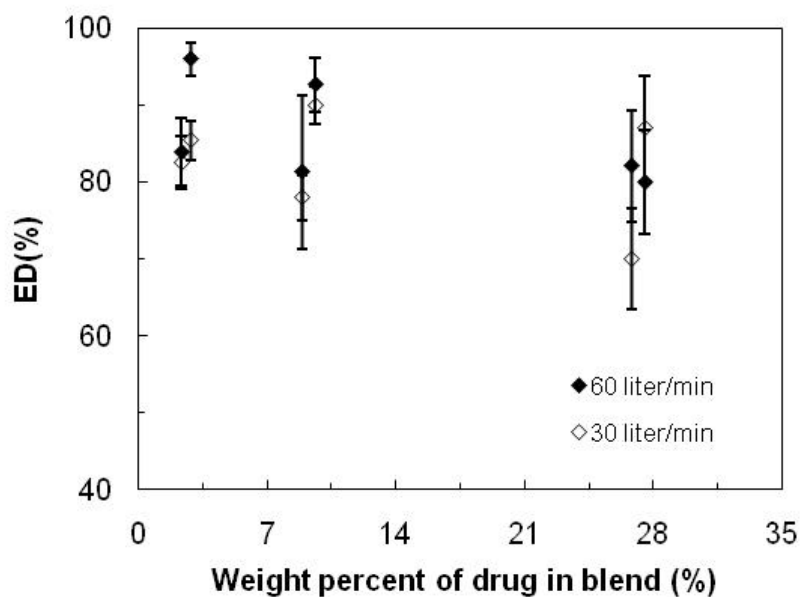
From the SEM images of Figure 8.5, it can be seen that a certain amount of drugs are not attached onto the carrier surfaces. The unattached drugs may attach to the inhaler surfaces and cause unnecessary loss of expensive drugs. Therefore, the drug attachment characteristic on carriers is very important for effective usage of the formulation. Sieving tests are performed for the different blends and the drug contents before and after sieving are compared. The average drug content in the samples and the % CV of the samples after sieving are listed in Table 8.4. It can be seen that the average drug content is significantly lower for LA-1 particles than the HA-1 particles after sieving with an exception of high drug mixing ratio of 2:1. For drug mixing ratio of 45:1 and 10:1, the LA-1 blends show a substantial reduction in average drug content after sieving. On the other hand, HA-1 particles show a fairly low reduction. At a high drug mixing ratio of 2:1, the surfaces of the carrier particles are fully covered with drug particles. The excess

unattached drug particles may form aggregates and limit the physical separation. Therefore, reductions in the drug content from these two blends with ratio 2:1 are not significantly different. It is also noticed that the homogeneity of the blends reduces after sieving. A minor variation in drug content may be observed because the drug content after sieving depends on different factors like the storage time and condition, sieving time and condition, the degree of mixing etc. It is believed that drug attachment ability of the carrier particles is proportional to carrier surface area. The BET surface areas of the carriers are shown in Table 8.1 & 8.2. The average opening diameter among the petals are found to be around 5 μm while the average drug particles size is around 3 μm . It would be reasonable to assume that the drug particles would have access to most of the surface area of the petal like morphology of the HA-1 carriers.

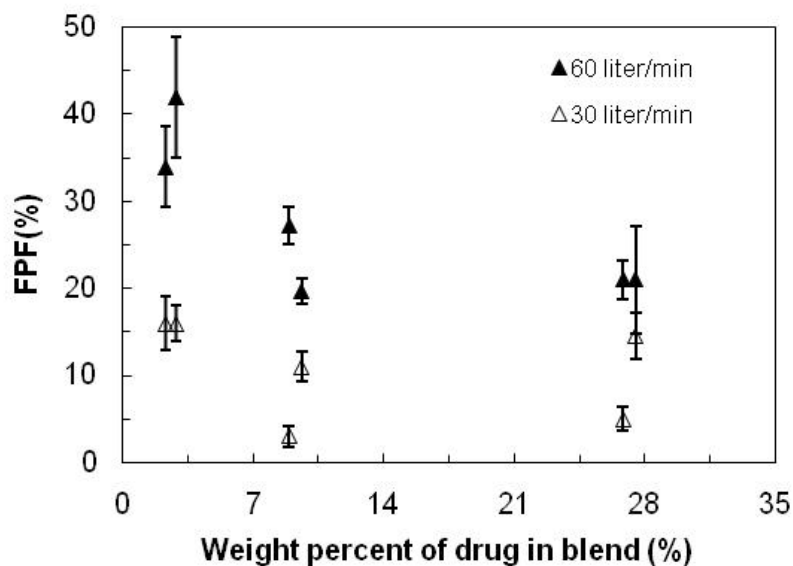
Table 8.4 Attachment of Bd drug with LA and HA carriers.

Wt ratio (Carrier: drug)	Carrier	After sieving		Reduction in drug content (wt %)
		Wt fraction of drug (wt %)	CV (%)	
2:1	LA-1	12.9	13.5	43.8
	HA-1	13.9	3.84	38.3
10:1	LA -1	4.35	3.76	52.5
	HA-1	7.55	4.84	27.0
45:1	LA-1	0.62	7.73	62.2
	HA-1	1.72	11.8	20.7

8.3.6 *In vitro* aerosolization and deposition behavior with blending formulation with different drug mixing ratio



(a)



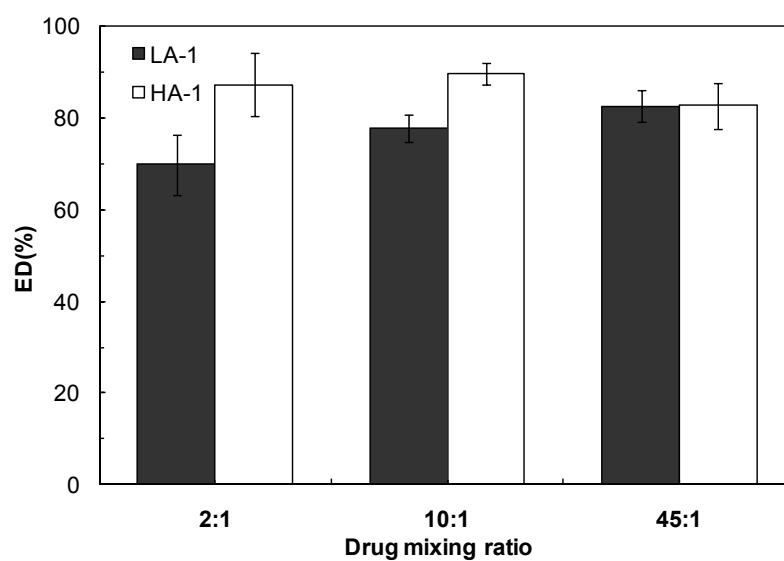
(b)

Figure 8.7 a) Emitted dose and b) fine particle fraction of the blends with different drug weight percentage obtained from homogeneity test for 60 and 30 L/min.

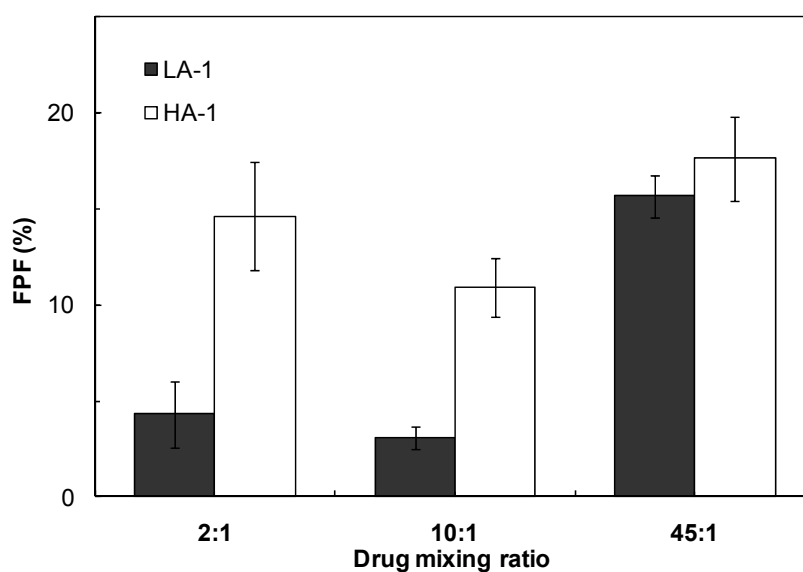
The ED and FPF results obtained from the ACI experiments are plotted as a function of the weight percent of drug in the blend and shown in Figure 8.6. The weight percent of drug in the blend is obtained from the blend homogeneity test (Table 8.3). In Figure 8.6(a), the ED results do not seem to be a strong function of weight percent of drug. However, blends with a higher drug percentage show a slightly lower average ED. A flow rate of 60 L/min is found to provide slightly higher ED than a flow rate of 30 L/min. A higher flow rate generates higher shear force, which improves the aerosolization of the drug particles⁴⁰. It can be seen in Figure 8.6(b) that FPF decreases with an increase in drug mixing ratio. Gas flow rate has a more significant effect on the FPF than the ED. FPF at 60 L/min is substantially higher than that at 30 L/min. The increased shear force at higher gas flow rate does not only improve the aerosolization of the blend but improve the liberation of the drug particles as well, thus allow more drug particles to travel to the lower stages.

A certain variation in ED and FPF can be observed at similar drug weight percentage since different carriers are used. It would be beneficial to analyze the emission and deposition properties of the each carrier separately. The ED and FPF results at a flow rate of 30 L/min for the different blends are compared separately in Figure 8.7. The LA-1 blend is found to give lower ED than the HA-1 blends at a flow rate of 30 L/min especially at high drug mixing ratios of 2:1 and 10:1. It is believed to be due to the high amount of unattached drug particles in the LA-1 blends. LA-1 particles can attach lower number of drug particles due to their lower surface area. On the other hand, the morphology of HA-1 carriers allows the attachment of more drug particles than the LA-1

carriers. Lower amount of unattached drug would be lost in the inhaler and therefore a higher ED is observed.



(a)



(b)

Figure 8.8 a) Emitted dose and b) fine particle fraction of blends with carrier to drug mixing ratio 2:1, 10:1 and 45:1 at 30 L/min.

Similar to the ED result, the FPF of the LA-1 blend is also lower than that of the HA-1 blends as shown in Figure 8.7(b). The low FPF of the LA-1 blends is a result of low emission from the inhaler. While the aerodynamic diameter of both LA-1 and HA-1 particles are high enough not to penetrate into the lower stages, the FPF would only be contributed from the liberated drug particles.

The flow and deposition behavior of these blends can be comprehended based on the regional deposition results of the blends at 30 L/min. The detailed deposition results of the carrier blends with different drug mixing ratios are shown in Figure 8.8 and 8.9. From the detailed deposition, it can be seen that the depositions are mostly happen in the initial three regions. The deposition in the inhaler is due to the attachment of drug particles on the inhaler surface. The bending at the throat region causes some of the blend to deposit due to inertial impaction. The preseparator is designed in such a way that most particles having an aerodynamic diameter larger than 10 μm will be retained. High drug deposition would be observed if carriers are deposited without sufficient drug liberation.

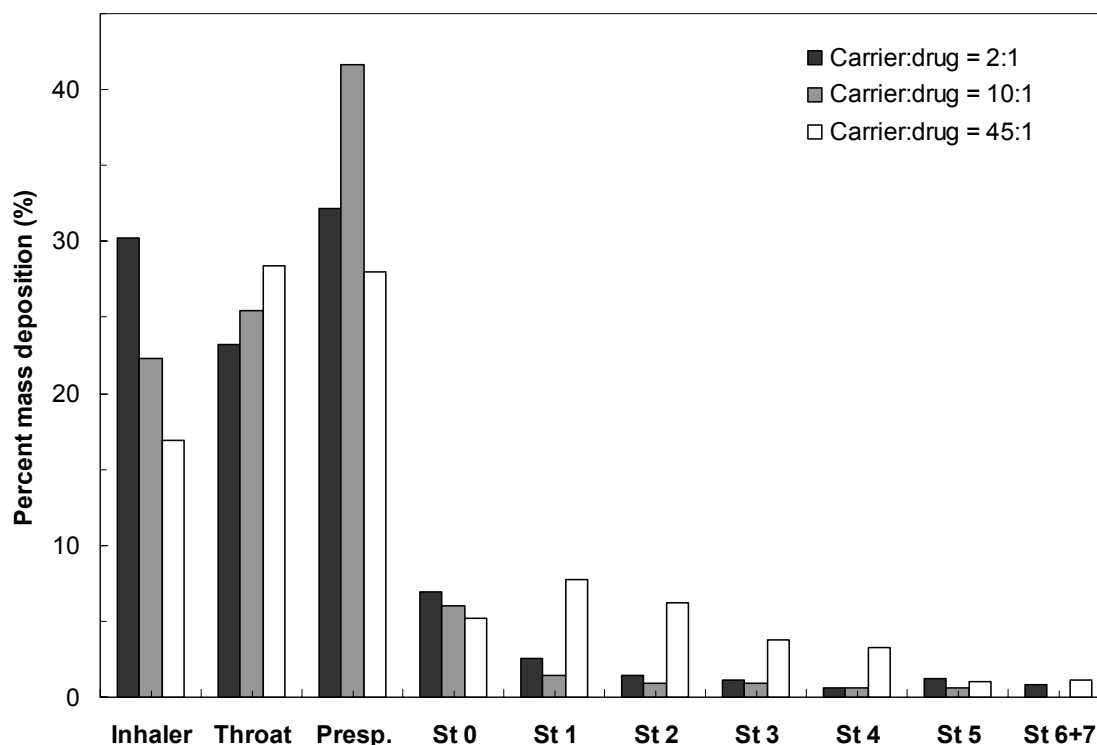


Figure 8.9 Detailed deposition of the drug from the LA-1 blends with different carrier to drug mixing ratio at 30 L/min.

The regional deposition result in Figure 8.8 indicates that for the LA-1 blends a substantial amount of drug particles is deposited in the inhaler. It is reasonable considering the large amount of unattached drug particles observed in the LA-1 blends. Additional drug deposition in the throat and preseparator would be due to the deposition of LA-1 carriers together with drug particles not being liberated. After these three initial stages, only a small amount of drug particles are left to travel to the lower stages of the impactor and exhibit low FPFs. An increase in drug mixing ratio indicates a higher deposition at the inhaler region, which is in agreement with the observations of higher amount of unattached drug particles at higher drug mixing ratios. While more drug particles are deposited in the inhaler region at higher drug mixing ratios, less drug

particles would be able to go into the system and subsequently travel to the lower stages.

Thus, the ED and FPF also decrease with an increase in drug mixing ratio.

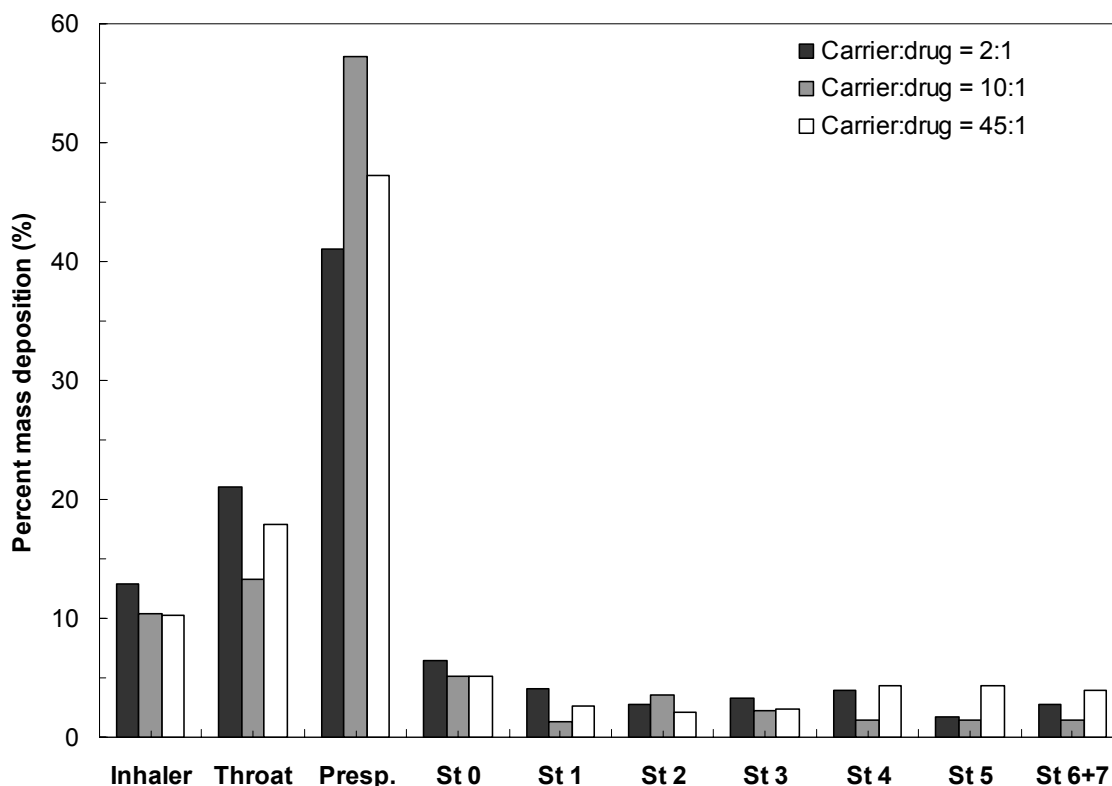
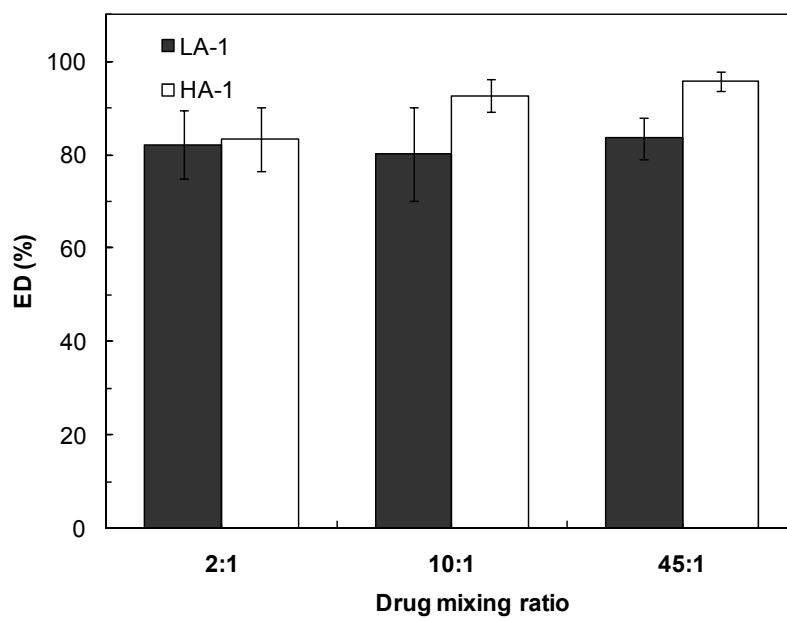
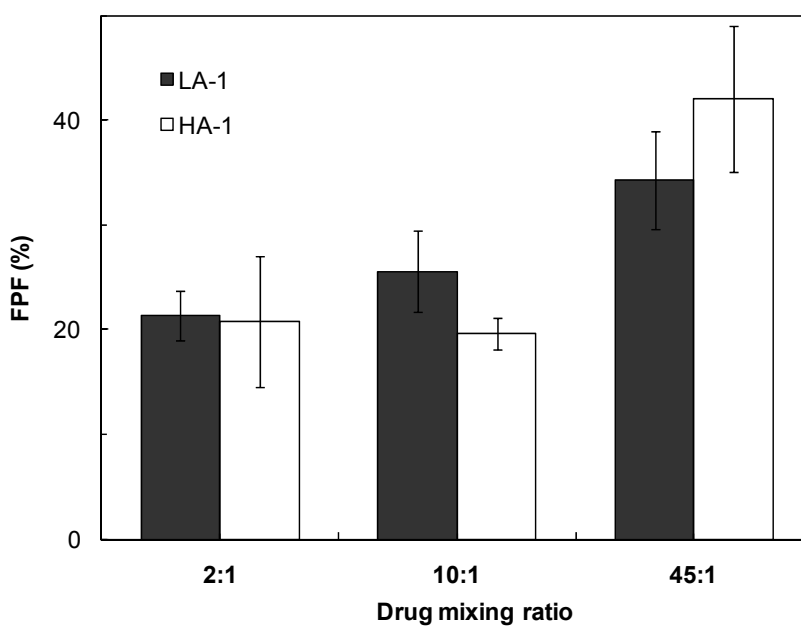


Figure 8.10 Regional deposition of the drug from HA-1 blends with different carrier to drug mixing ratio at 30 L/min.

The detailed deposition results of HA-1 blends are showed in Figure 8.9 As expected, the deposition in the inhaler region for HA-1 blends is lower than that for the LA-1 blend due to the better drug attachment. The comparable amount of deposition in the inhaler region at drug mixing ratios of 10:1 and 45:1 for HA-1 blends indicate that the drug attachment limit of the HA-1 carriers has not been reached even at a relatively high drug mixing ratio of 10:1.



(a)



(b)

Figure 8.11 a) Emitted dose and b) fine particle fraction of blends with carrier to drug mixing ratio 2:1, 10:1 and 45:1 at 60 L/min.

A flow rate of 60 L/min is also used to approximate the maximum inhalation rate of a normal adult in the dry powder inhalation process. The ED and FPF results for the blends with different carriers at a flow rate of 60 L/min are shown in Figure 8.10. A flow rate of 60 L/min is found to have higher ED than 30 L/min for all formulations. An increase in the air flow rate increases the turbulence and shear forces, which in turn to improve the aerosolization of the drug particles. It can be seen in Figure 8.10(a) that HA-1 blends again show higher ED than the LA-1 blends. However, the relationship between ED and drug mixing ratios at a flow rate of 60 L/min is not as obvious as that at a flow rate of 30 L/min owing to the higher shear forces at higher flow rate. Figure 8.10(b) shows that the FPF of the blends increases with the decrease of drug mixing ratio. When Figure 8.10(b) is compared with Figure 8.7(b), it can be seen that the LA-1 blend has the higher increase in FPF as the gas flow rate is increased, especially at high drug mixing ratios. This is because the large shear force generated at high gas flow rate would be able to aerosolize some of the unattached drug particles. Once these unattached drug particles are aerosolized, they would be able to travel directly to the lower stages without the need of liberation. The HA-1 blends also shows an improvement in FPF with an increase in gas flow rate. However, the improvement is not as significant as the LA-1 blends because the HA-1 blends does not have as much unattached drug particles. The improvement in FPF would be mainly due to the improved drug liberation at the higher gas flow rate. It is also found that the drug liberation is more effective at low drug mixing ratios than at high drug mixing ratios.

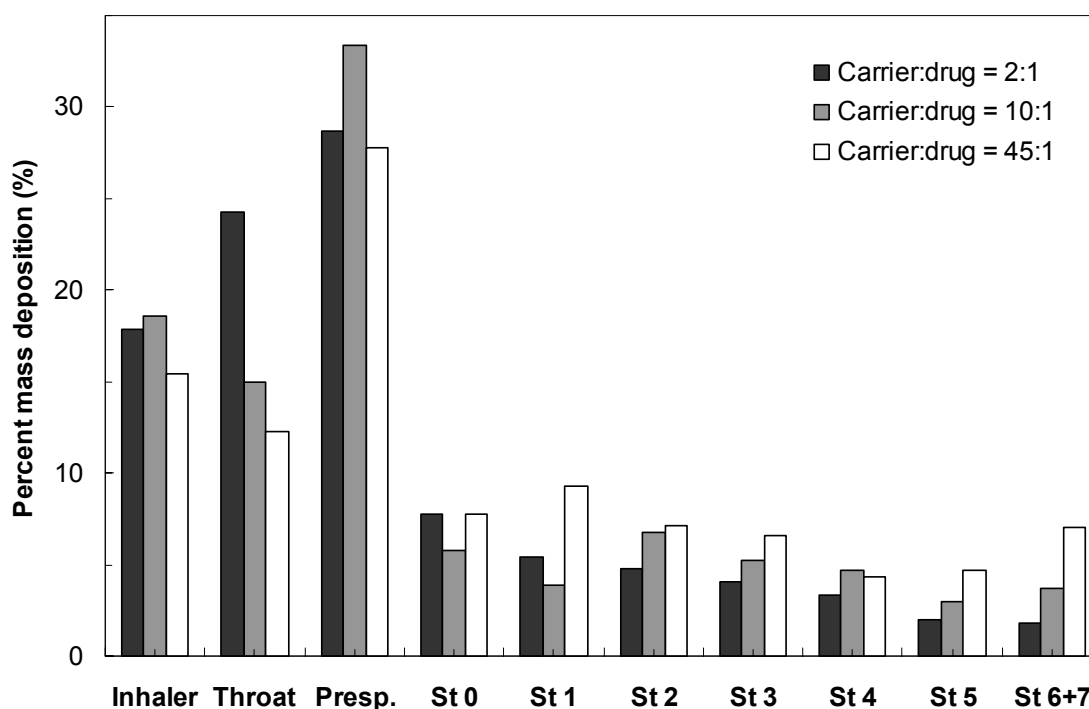


Figure 8.12 Regional deposition of the drug from LA-1 blends with different carrier to drug mixing ratio at 60 L/min.

The regional deposition of the blends at 60 L/min is also showed in Figure 8.11 and 8.12. A comparison of Figure 8.8 and 8.9 shows that an increase in gas flow rate from 30 L/min to 60 L/min essentially reduces the amount of drug particles deposited in the earlier regions. As mentioned earlier, the high gas flow rate helps improve the drug liberation from the carriers and reduces early depositions.

A summary of all the results reported in this study suggests that pollen-shape particles can be a viable choice as drug carriers in inhalation drug delivery. The pollen-shape morphology allows high drug mixing ratio without sacrificing the delivery efficiency compared to traditional LA carriers especially at lower inhalation flow rate. A much wider application of the pollen-shape carriers can be possible if the drug liberation can be improved.

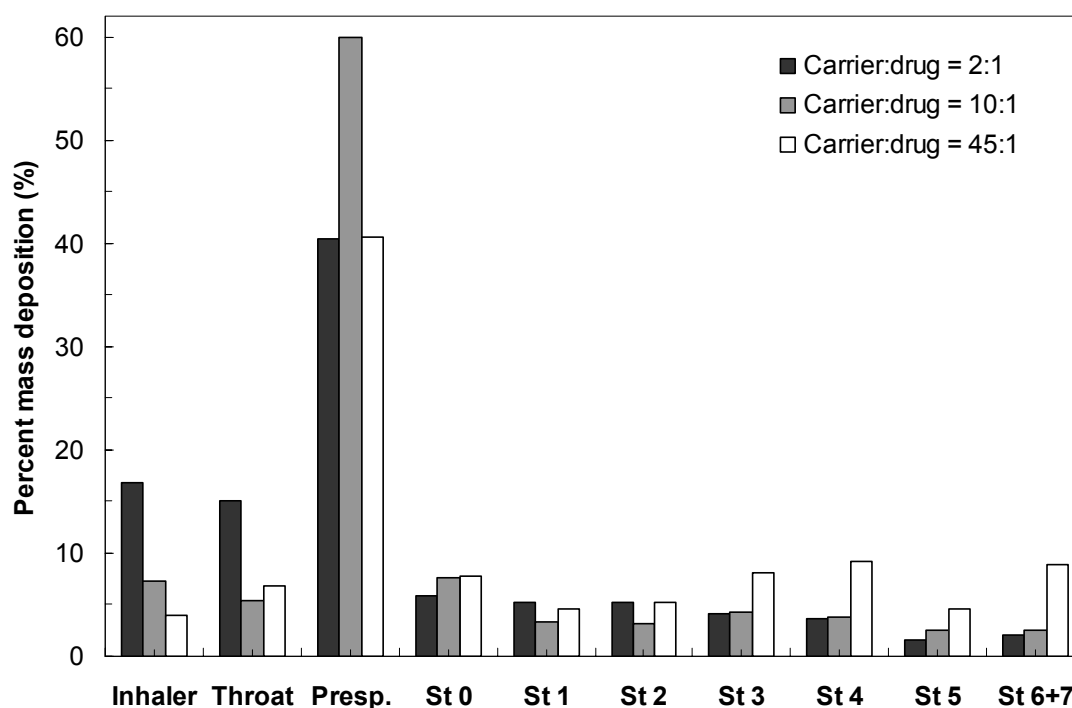


Figure 8.13 Regional deposition of the drug from HA-1 blends with different carrier to drug mixing ratio at 60 L/min.

8.4 Conclusion

The inhalation performance of pollen-shape HA carriers with a geometric diameter of 48.6 μm is compared with LA carriers with a size range 38–75 μm with different drug mixing ratios. The drug attachment ability of the HA carriers is higher than that of the LA carriers. A significant amount of the drug particles is found to remain unattached to the LA carriers. These unattached drug particles maybe lost in the inhaler and reduce the aerosolization and deposition performance of the blends. The HA blends show high ED of 82–90% at 30 L/min while the LA blends are observed to have ED of 69–82% at the same conditions. The high emission of the HA blends also allows high FPF of 10–18% while the FPF of the LA blends are 3–15%. At a gas flow rate of 60

L/min, all the HA and LA blends show compatible ED (83–95% for HA blends and 82–84% for LA blends) and FPF (19–41% for HA blends and 21–34% for LA blends). This study shows the suitability of pollen-shape carriers especially at a low flow rates and high drug mixing ratios. The liberation of drug particles from the pollen-shape carriers need to be improved for a more diverse application of pollen-shape carriers.

Chapter 9 INHALATION PERFORMANCE OF POLLEN-SHAPE CARRIER IN DRY POWDER FORMULATION WITH DIFFERENT DRUG MIXING RATIO: COMPARISON WITH DIFFERENT POLLEN-SHAPE CARRIERS

9.1 Introduction

Pollen-shape drug carrier particles are found to have improved inhalation performance. Carrier morphology plays the key role in their performance. Therefore, the study of the effect of size and surface morphology of pollen-shape carrier particles on delivery efficiency is essential.

Carrier particles are normally too large to reach the lower generations of the lung. While the carrier particles are swallowed, drug particles attached on the carrier surfaces will need to be detached and resuspended in the air stream for deep lung delivery. The detachment of the drug particles is found to be a function of carrier particle size and the interaction between drug and carrier particles. Corn¹⁸⁶ found that once the drug particles are attached to the carriers, they will need to slide and roll along the carrier surface before being suspended in the air stream again. The travel distance along the carrier surface and hence the aerosolization properties of the drug particles are determined to be a function of carrier particle size¹⁸⁷⁻¹⁸⁹. Surface morphology can also improve the drug liberation by

increasing the time of flight.¹⁰¹. Hence, the physical properties of the carrier particles are important for efficient drug delivery.

It is found that particles having pollen-shape surface morphology can be good candidates as drug carriers for dry powder inhalation. Better flow and aerosolization behavior and better drug attachment are observed in pollen-shape carriers compared to traditional lactose carriers. However, the morphology and size of the pollen shape carrier particles may have high influence on the aerosolization and deposition behavior for different drug loading and inhalation flow rate. In this study, the drug attachment behavior and *in vitro* aerosolization and deposition properties of Bd blended with two different pollen-shape HA carriers are investigated. One pollen-shape carrier has a petal-like surface morphology while the other has a needle-like surface morphology. Comparisons are made with three different drug mixing ratios and at inhalation flow rate of 30 and 60 L/min.

9.2 Experimental

9.2.1 Preparation of HA

The preparation of HA particles has been described in the Experimental chapter (Section 3.2.1). HA-1 and HA-2 with distinct surface morphology and size are used in this study.

9.2.2 Particle characterization

Synthesized particles are characterized with their size, morphology, powder density, XRD diffraction, TGA isotherm and BET surface area.

9.2.3 Drug content, content uniformity, and drug attachment

Blends with carrier to drug weight ratios of 2:1, 10:1, and 45:1 are used. The blends are characterized first with SEM and mixture vial appearance. Average drug content and content uniformity of all the blends are measured by analyzing the quantity of Bd. An estimation of drug attachment capacity of the HA particles is carried out using sieving method discussed in chapter 7 (Section 7.2.3).

9.2.4 *In vitro* aerosolization and deposition properties

10±0.3 mg of each blend is loaded in the capsule before introducing into the Rotahaler[®]. Two air flow rates of 30 L/min and 60 L/min are used. In order to maintain the same inhalation volume, an actuation time of 4 sec and 8 sec are used for 60 and 30 L/min, respectively.

3. Results and discussion

9.3.1 Particle characteristics

Two types of pollen-shape HA particles are synthesized. Physical properties of the particles are presented in Table 9.1. The SEM image of HA-1 particles, reported in the first part of the study, showed that HA-1 particles have a petal-like surface morphology. On the other hand, a needle-like surface morphology can be seen in the SEM image of smaller sized HA-2 particles in Figure 9.1.

Particle tip to tip distance measured from the SEM images is taken as the particle diameter. Since the size distributions measured from the SEM images are number-weighted while the size distributions measured from laser diffraction measurement are volume-weighted, the SEM size distributions need to be converted to volume-weighted distribution for comparison. For the conversion of number-weighted size distribution to

volume-weighted size distribution, the pollen-shape particle volume is estimated as the sphere volume having the same diameter. Therefore, the SEM measured sizes are slightly higher than $d(50\%)$ of the laser diffraction measured size. Nonetheless, the size distribution can be considered uniform since the span of all the HA particles are close to 1¹⁸³.

A large difference between ρ_{bulk} and ρ_{tap} can be found for HA-2. It is possibly due to the presence of the relatively sparse needle-like surface that allows the particles to pack loosely. Upon tapping, the needle-like surface would allow the particles to pack tightly into each other. The particle size is commonly expressed as the particle aerodynamic diameter.

Table 9.1 Physical characteristics of the HA samples

Sample	HA-1	HA-2
Average dia.(SEM) (μm)	48.6	27.1
St.dev. (μm)	10.21	4.02
$d(50\%)$ (μm)	45.9	24.8
$d(10\%)$ (μm)	25.9	15.1
$d(90\%)$ (μm)	85.9	41.0
Span	1.31	1.04
ρ_{bulk} (g/cm^3) (n=0)	0.215	0.105
ρ_{tap} (g/cm^3) (n=2500)	0.289	0.218
d_a (μm)	24.9	12.06
Specific Surface area (m^2/g)	17.1	20.83

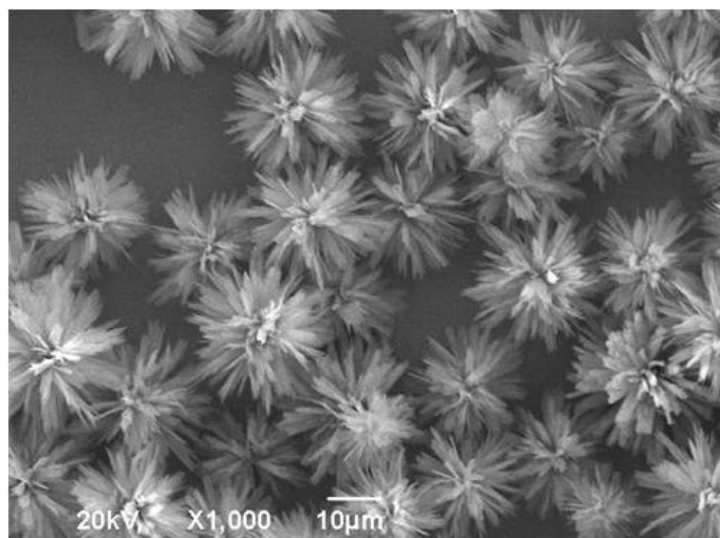


Figure 9.1 The SEM image of HA particles produced by using PSS-30 g/L & urea-0.5M, at 200⁰C (HA-2).

Both HA particles shows similar diffraction patterns in the XRD results presented in Chapter 7. It indicates that both HA particles exhibit the same hexagonal phase crystalline structure according to the powder diffraction file (PDF) no 00-009-0432. XRD results also confirms the purity of the samples. TGA thermograms of both HA particles shown in presented in Chapter 7 indicates that there is negligible weight loss (<2%) below 100 °C. The effect of moisture on particle-particle interactions can be neglected for both HA particles⁹⁹.

9.3.2 Drug blending

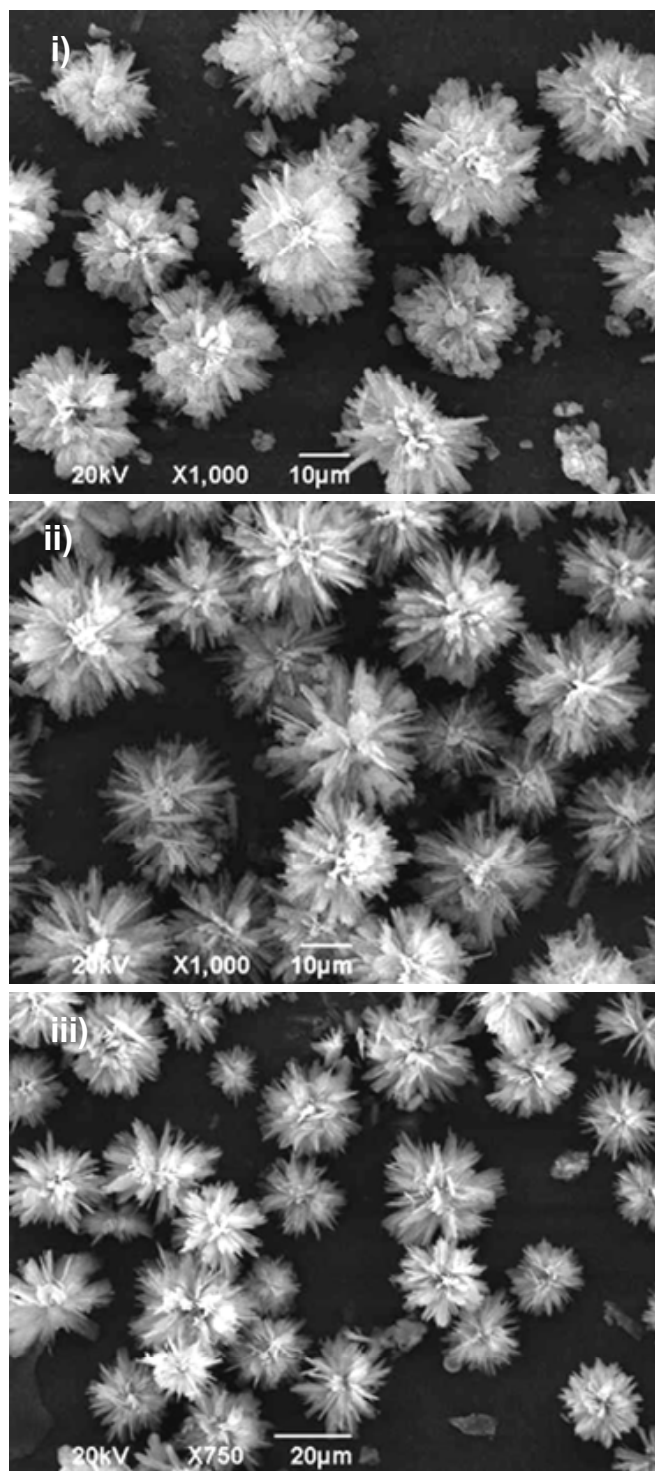
The model drug, Bd has an average size of $2.5 \pm 1.1 \mu\text{m}$. Three drug mixing ratios of 2:1, 10:1 and 45:1 (carrier: drug) are used. The physical appearances of the blends are observed in small transparent vials and by SEM and the results are shown in Figure 9.3. In chapter 8, it is shown that HA-1 blends only show a minor amount of unattached drug particles at high drug mixing ratio of 2:1. As shown in Figure 9.2(a), no obvious drug attachment on the vial surface is physically observed for HA-2 blends at all drug mixing

ratios. The SEM images in Figure 9.2(b) indicate that most of the drug particles are attached to the needle-like surface morphology.



(a)

Figure 9.2 Comparison of the blends, (a) in the mixture vials, with different carrier to drug mixing ratio for HA-2 particles (i) without drug; with wt ratio (ii) 45:1, (iii) 10:1, (iv) 2:1, (b) SEM images with drug mixing ratios, (i) 45:1, (ii) 10:1, (iii) 2:1.



(b)

Figure 9.3 Cont.

9.3.3 Blending homogeneity

Blend homogeneity is represented by the coefficient of variance (CV) of the Bd content in the blends. The average drug content and CV for the blends after mixing are listed in Table 9.2. Though the blend homogeneity of HA-1 carriers increases with an increase in drug mixing ratio, no particular trend is observed for HA-2 carriers. Moreover, HA-2 carriers consistently yield higher drug content than HA-1 carriers. It indicates that needle-like surface of HA-2 has better drug attachment capability than the petal-like surface of HA-1 carriers. Nonetheless, all the blends can be considered as uniform since the CV of the blends are all less than 10%.

Table 9.2 Average Bd content and homogeneity of the blends

Wt ratio (Carrier: drug)	Carrier	Wt fraction of drug (wt %)	CV (%)
2:1	HA-1	22.5	0.26
	HA-2	23.7	5.74
10:1	HA-1	8.9	2.14
	HA-2	9.0	0.72
45:1	HA-1	1.81	6.26
	HA-2	2.16	5.44

9.3.4 Drug attachment

At high drug mixing ratio, all the drug particles may not be attached onto the carrier surfaces. The unattached drugs may attach to the inhaler wall and cause unnecessary loss of expensive drugs. Loose attachments of the drug particles also cause loss of drug during handing. Therefore, the drug attachment characteristic on carriers is very important for effective usage of the formulation. Sieving tests are performed for the

characterization of drug attachment in different blends and the drug contents before and after sieving are compared. The average drug content in the samples, CV and the reduction of drug content in the samples after sieving are listed in Table 9.3. HA-2 blends show the lower reduction than HA-1 blends after sieving for 10:1 and 45:1 drug mixing ratio. At a high drug mixing ratio of 2:1, the surfaces of the particles are fully covered with drug particles. The excess unattached drug particles may form aggregates and limit the separation by sieving. Therefore, a similar reduction in the drug content is found in the blends with 2:1 ratio.

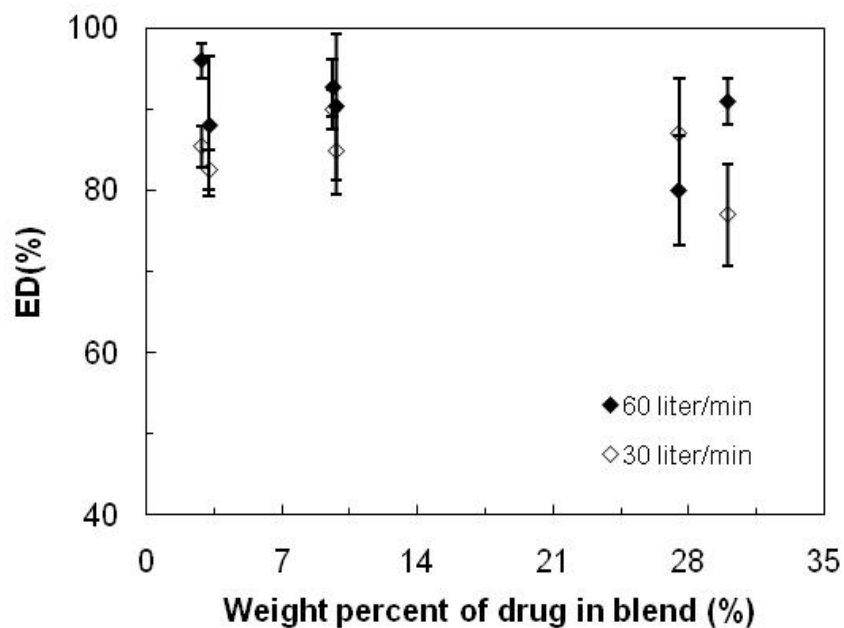
It is in agreement with the blend homogeneity results that the needle-like surface morphology of the HA-2 carriers can provide higher accessibility and more binding sites than the petal-like morphology of HA-1 carriers. Therefore, drug attachment with the HA-2 particles is higher than that from the HA-1. As drug particles attach to the surface of the carrier, specific surface area of the pollen-shape particles has a significant effect on drug attachment. HA-2 particles possess higher specific surface area than HA-1, presented in Table 9.1. Hence, in general, drug attachment ability of the particles is proportional to their specific surface area.

Table 9.3 Attachment of Bd drug with LA and HA carriers.

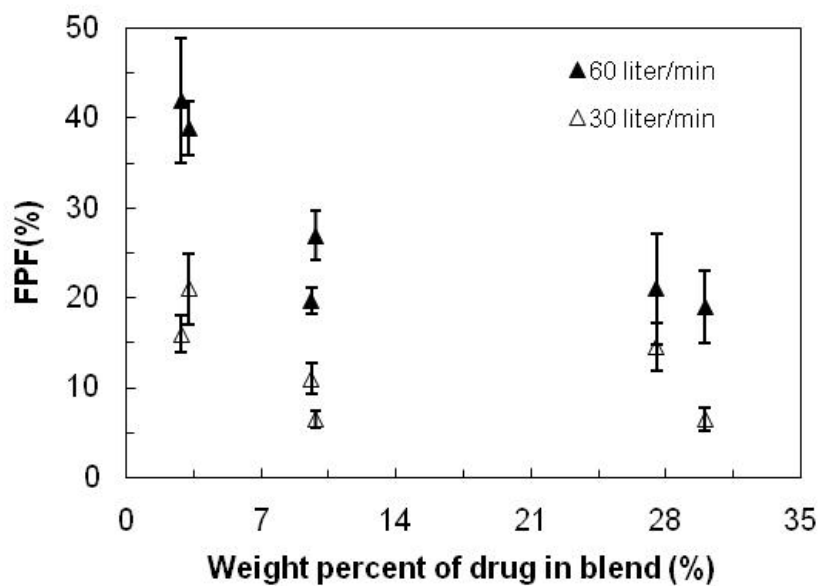
Wt ratio (Carrier: drug)	Carrier	After sieving		Reduction in drug content (wt %)
		Wt fraction of drug (%)	CV (%)	
2:1	HA-1	13.9	3.84	38.3
	HA-2	14.6	7.03	38.4
10:1	HA-1	7.55	4.84	27.0
	HA-2	9.05	9.84	12.3
45:1	HA-1	1.72	11.8	20.7
	HA-2	2.14	5.03	6.82

9.3.5 *In vitro* aerosolization deposition behavior with blending formulation with different drug mixing ratio

The ED and FPF results for the blends with HA-1 and HA-2 carriers as a function of drug weight percent (wt%) in the blend are presented in Figure 9.3 (a) and 9.3 (b), respectively. The wt % of drug in the blends is obtained from the drug content test. It can be seen in Figure 9.3(a) that a flow rate of 60 L/min provides higher ED than a flow rate of 30 L/min. It is in agreement with the finding that higher flow rate improves aerosolization capacity of powder formulation⁴⁰.



(a)



(b)

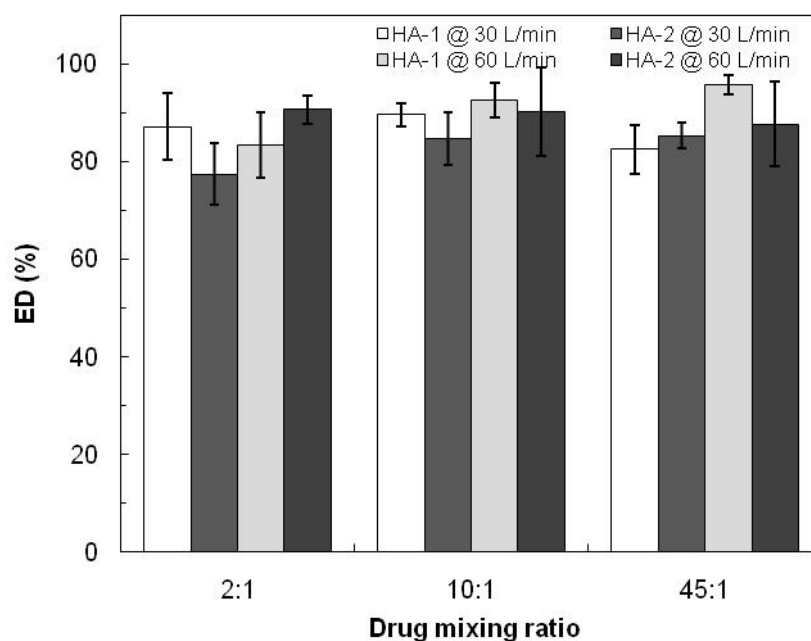
Figure 9.4 a) ED and b) FPF of the blends as a function of drug wt% at 60 and 30 L/min.

Nonetheless, all the blends exhibit ED value over 80% which is an indication of the good aerosolization properties of the pollen-shape HA particles. A more distinct difference in the FPF results with a change in flow rate can be seen in Figure 9.3(b). An increase in flow rate increases the FPF of the blends. The degree of FPF improvement is more significant at low drug wt% than that at high drug wt%. FPF mainly depends on the drug liberation from the carriers. As the shear force increases with gas flow rate, the drug liberation is also increased. However, the drug particles tend to form coagulates at high drug wt%. The formation of coagulates limits the aerosolization and inhibits the liberation of the drug particles. Hence, a lower FPF is observed.

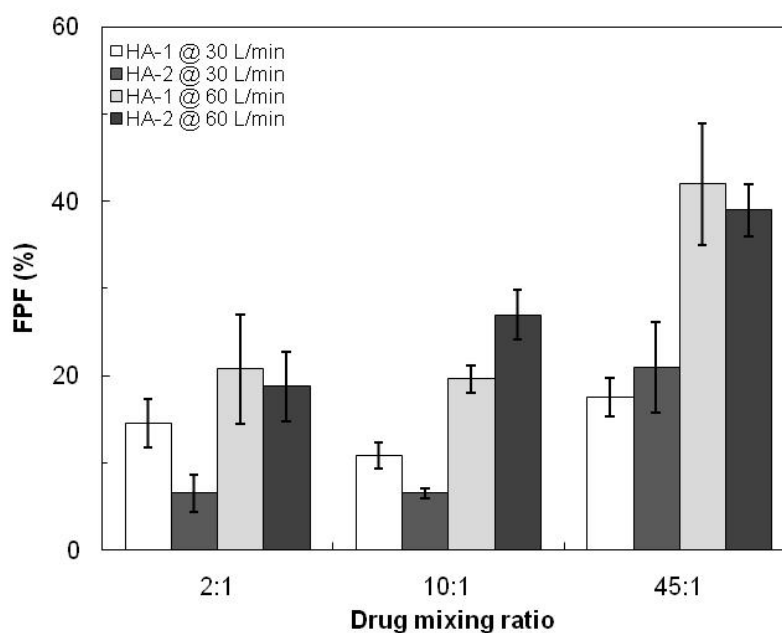
Variations in ED and FPF results for both HA-1 and HA-2 blends can be observed due to the different physical properties of HA-1 and HA-2 carriers. The ED and FPF results for both HA blends will need to be compared separately. A comparison of the ED of both HA blends are shown in Figure 9.4 (a). It can be seen that an increase in drug mixing ratio generally reduces the ED of both HA blends, especially at 60 L/min for HA-1 blend and at 30 L/min for HA-2 blend. However, the effect of drug mixing ratio on the ED is not significant. On the other hand, an increase in gas flow rate for 30 L/min to 60 L/min generally increases the ED of both HA blends. The effect of gas flow rate is more distinct for HA-1 blend at low drug mixing ratio and for HA-2 blend at high drug mixing ratio. It is anticipated that the variation in ED among the HA blends is a result of the difference in carrier size and morphology. As indicated in the drug attachment section, HA-2 carriers are capable of higher drug attachment than HA-1 carriers. A previous study showed that HA-1 particles have better flowability than HA-2 particles.¹⁹⁰ The better the powder flowability, the better the dispersion and aerosolization properties¹³¹. Moreover,

the attachment of large amount of drug particles may also reduce the aerosolization of the HA-2 blend and causes a lower ED at low gas flow rate of 30 L/min than the HA-1 blend. However, as the gas flow rate is increased to 60 L/min, the high shear force generated may cause some of the drug particles to be liberated from the carrier surface and aerosolized together with the carriers in the inhaler. In summary, the effect of carrier characteristics and gas flow rate on ED is stronger than the effect of drug mixing ratio.

As shown in Figure 9.4 (b), the effect of drug mixing ratio, gas flow rate and carrier characteristics on FPF is significantly higher than that on ED. An increase in drug mixing ratio decreases the FPF of both HA blends. An increase in gas flow rate from 30 L/min to 60 L/min substantially increases the FPF of both HA blends especially for the HA-2 blend at high drug mixing ratio. A comparison of the FPF of both the HA-1 and the HA-2 blends indicates that the difference in the FPF at low drug mixing ratio is less significant at both gas flow rates. However, the HA-1 blends tend to have higher FPF than the HA-2 blends at 30 L/min whereas the HA-2 blends tend to have higher or equivalent FPF than the HA-1 blends at 60 L/min. High drug attachment ability would typically be corresponding to low drug liberation and should give rise to a low FPF. The FPF result of the HA-2 blends at 60 L/min is contradictory to this principle and a more detailed analysis on the regional deposition results will be needed. Nonetheless, both HA-1 and HA-2 carriers are capable of high drug mixing ratios and deliver reasonably high FPF.



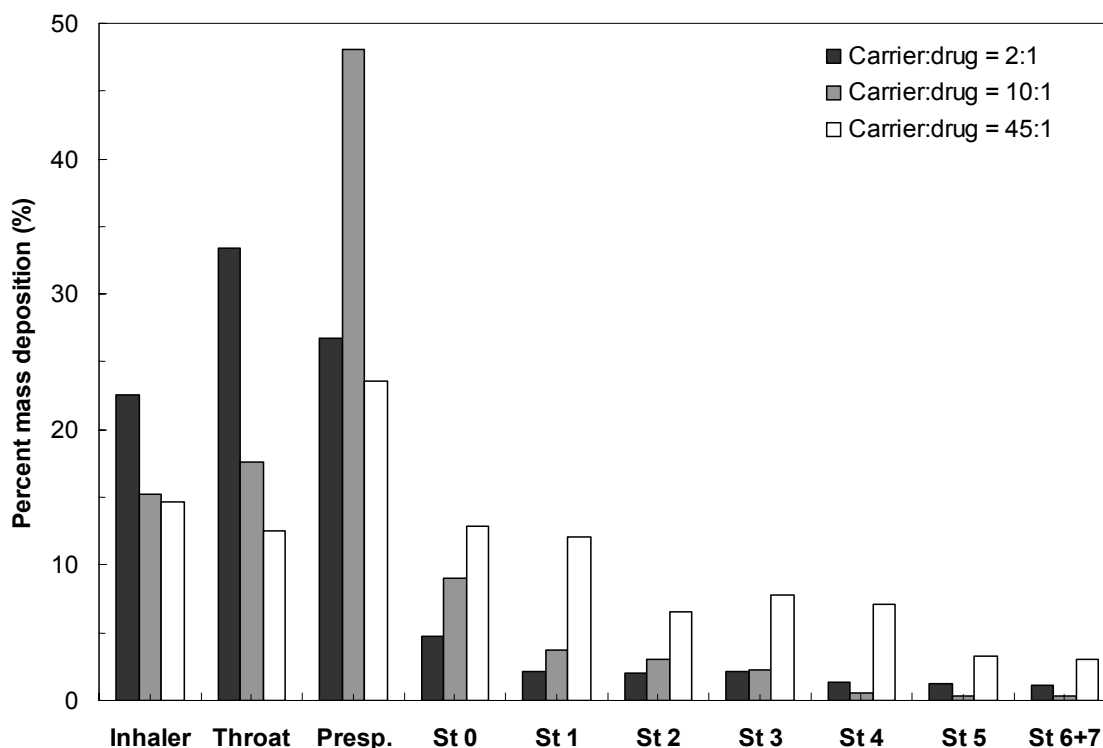
(a)



(b)

Figure 9.5 a) Emitted dose and b) fine particle fraction of blends with carrier to drug mixing ratio 2:1, 10:1 and 45:1 at 30 and 60 L/min.

Regional deposition results are important for a complete understanding of the flow and deposition behavior of the dry formulations. Regional deposition results of the HA-1 blends with the same drug mixing ratio and different flow rate are presented in the chapter 8. It was found that even at a low gas flow rate of 30 L/min, the HA-1 blends show low deposition in the inhaler and throat regions due to better flowability. However, a high deposition is found in the preseparator region, which indicates that there is drug liberation problem for the HA-1 carriers. The detailed deposition result of the HA-2 blends at 30 L/min and 60 L/min are shown in Figure 9.5 (a) and (b), respectively. It is shown in Figure 9.5 (a) that heavy depositions are mostly seen in the inhaler, throat and preseparator regions at 30 L/min. The high drug attachment ability of the HA-2 carriers reduces the flowability of the HA-2 blends at high drug mixing ratios. Thus a high deposition in the inhaler can be observed for a drug mixing ratio of 2:1. As the drug mixing ratio is reduced to 10:1, the aerosolization is improved. However, a high deposition is still observed in the throat region due to the limited drug liberation from the HA-2 carriers. On the other hand, at low drug mixing ratio of 45:1, the HA-2 blend shows lower deposition in the initial three regions and higher FPF result is observed compared to drug mixing ratios of 2:1 and 10:1.

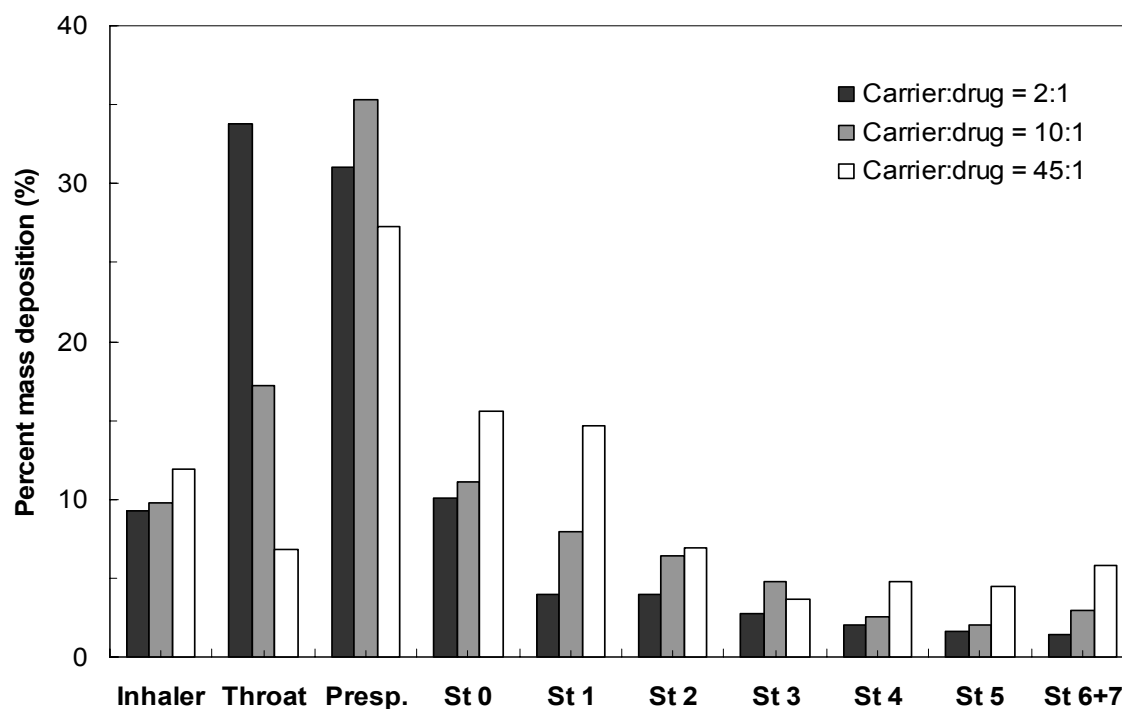


(a)

Figure 9.6 Regional deposition of the drug from the HA-2 blends with different carrier to drug mixing ratio at, (a) 30 L/min and (b) 60 L/min.

In Figure 9.5(b), the regional deposition result of the HA-2 blends at 60 L/min shows that an increase in gas flow rate from 30 L/min to 60 L/min essentially reduces the amount of drug particles deposited in the earlier regions especially in the inhaler. A high deposition is still observed in the throat region at high drug mixing ratio of 2:1 because of inertial impaction. For the HA-1 blends, an increase in the gas flow rate reduces the deposition in the inhaler and throat regions. However, similar to the HA-2 blends, a relative high deposition is still observed at high drug mixing ratios. A comparison of the regional deposition results between the HA-1 and HA-2 blends indicates that the HA-1 blends has lower deposition in the inhaler and throat regions but significantly higher

deposition in the preseparator region than the HA-2 blends. It is possibly because the HA-2 carriers has low aerodynamic diameter and can be dispersed as discrete particles at 60 L/min. It allows the carriers to travel to the lower stages of the impactor together with the drug particles and improves the FPF.



(b)

Figure 9.7 Cont.

The overall result of this experimental study shows that small pollen-shape HA-2 carriers with needle-like surface morphology and higher specific surface area can exhibit high drug attachment capacity. However, large amount of attached drug particles would lower the aerosolization properties. On the other hand, large pollen-shape particles with petal like surface morphology show better performance as drug carriers for low flow rate and high drug mixing ratio. Therefore, size and surface morphology of pollen-shape

carrier particles has important effect on drug loading and inhalation performance and the behaviors can be improved by optimizing the properties. Both HA-1 and HA-2 carriers have limited drug liberation, an improvement in this aspect would further make the inhalation performance of the pollen-shape carriers more efficient.

9.4 Conclusion

The inhalation performance of pollen-shape model HA carriers with a geometric diameter range of 27-48 μm and pollen-like surface morphology are compared with different drug mixing ratios with Budesonide. The drug attachment ability of the small HA-2 carriers with needle-like surface morphology is higher than that of the large HA-1 carriers with petal-like surface morphology. It is found that the effect of carrier surface morphology, size and gas flow rate has stronger effect on ED than the effect of drug mixing ratio. The blends are observed to have an average ED value of around 90%. The HA-1 carriers provide higher FPF than the HA-2 carriers at low gas flow rate while the opposite happens at high gas flow rate. The reason is that the small HA-1 carriers are capable of passing to the lower stages at high gas flow rate together with the drug particles without liberation. This study shows the suitability of pollen-shape particles as drug carriers in dry powder inhalation with different physical properties. Additional improvement in the drug liberation from these carriers would facilitate more efficient pulmonary drug delivery.

Chapter 10 CONCLUSIONS AND RECOMMENDATIONS

10.1 Conclusions

Pulmonary route possesses a high potential in delivering aerosol medication in human body. In this technique, drug formulations are delivered to the lung epithelium to treat different lung or systemic diseases. This route offers several advantages in comparison to other means of drug administration. Improvements in delivery efficiency through this route would deem great potential to future drug delivery trends. Optimization of the physical properties of drug formulation is the most useful way for improved delivery. In this research work, the insights of the dry particle shape and morphology effect on drug delivery efficiency is studied. Particle shape effect in macro-particle system is studied at the beginning. Then, similar sized micro particles with different shapes are synthesized for inhalation studies. The essential properties for inhalation, like flowability, aerosolization and deposition behavior of the particles are determined experimentally. A pollen shape is found to exhibit preferable inhalation properties. After a wide particle size range is being formulated, the suitability of the large-sized pollen-shape particles as drug carrier is assessed. The behavior of these carrier particles is checked with different drug loading at various inhalation flow rate. The size and morphology effect of this type of carrier particles is also compared to traditional carriers.

In the preliminary study, effect of particle shape on gravitational sedimentation is assessed. The settling of different shape micro-particle is investigated in a confined medium under the inertial flow regime. It is observed that the presence of wall has strong impact on the settling velocities. Most of the available wall effect models developed with spherical particles can roughly predict the wall effects of bicone and cube shape particles. However, these models fail to predict the wall effect for cylindrical particles with high aspect ratios in the experimental Reynolds number range. A transition from a decreasing to an increasing trend of $u_t/u_{t\infty}$ is observed with an increase of equivalent particle diameter to column diameter ratio, λ . It is found that the particle change its settling orientation in the response of the wall restriction which attributes the change in their velocity. The result indicates that the significant effect of the wall on the particle settling is highly influenced by particle shape. Favourable settling or flowing of particles can be obtained by changing the shape of particles.

The study with micro particle indicates that flow and deposition properties can be altered by utilizing irregular shapes. The behaviors of different shape particles with d_a range of 1-5 μm are compared. Comparisons are performed in terms of both d_a and characteristic size of the particles. It is found that, the flowability and *in vitro* aerosolization and deposition properties do not follow a regular trend with aerodynamic diameter and characteristic size. It is also found that particle shape has a strong effect on this behavior and shape factor alone is not enough to account for the difference. Pollen-shape particles are found to have improved flowability, aerosolization and deposition performance compared to other shape particles. Pollen-shape micro-particles are found to have ED values over 80% where other shape particles show in the range 50-75%. The

FPF result of the pollen shape particles is over 15% where for other shape particles it varies in the range 2-10%. It is anticipated that surface asperities of the pollen-shape particles reduce the inter-particle interactions and adhesion properties. Hence, the better flow properties and dispersibility as discrete particles allow the pollen-shape particles to be aerosolized easily and achieve a high FPF.

As the pollen-shape particles have good flowability and aerosolization behavior, the feasibility of its application as drug carriers in dry powder formulation is studied. Large carrier-sized pollen-shape HA particles are produced. The flow behavior, velocity field and turbulence occurrence induced by this particle laden gas flow are examined physically in an idealized inhalation path model using PIV technique. The pollen-shape particles exhibit a flowability index CI which is almost twofold better than that of traditional LA carrier particles (CI is 18-35% for HA and 32-60% for LA). PIV results show that the pollen-shape HA particles have better flow behavior. Due to the low surface density, the pollen-shape particles show lower inertia and able to follow the geometry of the inhalation path model. Smaller separation regions are generated compared with the LA particles flowing through the path model. Deposition study with gravimetric measurement shows that most of the deposition happened in the critical bending section (resemble to the throat of human lung) and inertial impaction & gravitational deposition are responsible mechanism for that. It is found that particles with low d_a exhibit reduced deposition in the bending section. Therefore, pollen-shape particles have higher potential to travel further way in the lung airways and they can be a good candidate as drug carriers in dry powder inhalation.

The positive findings on the flow properties of large pollen-shape HA particles urge for a feasibility study of using the HA particles as model carriers binary mixture with a model drug. *In vitro* aerosolization and deposition properties of the model carriers in binary mixtures with a model drug Bd are studied at a constant mixing ratio. The drug loading capability of the pollen-shape HA particles are found more than twice compare to the LA carriers. The drug concentration in the HA blends (mixed in 10:1 w/w carrier to drug ratio) after sieving is 6.51-7.90% where the drug concentration in the LA blends is 3.73%. Bd blended with pollen-shape carriers are also observed to have better ED (LA=77% and HA=84-90%) and FPF (LA=3% and HA=6.5-11%) than that blended with traditional LA carriers at 30 L/min. It is evidenced that the pollen-shape HA carriers can be a promising drug carrier in dry powder inhalation.

As pollen-shape carriers have high delivery potential, influence of drug loading and inhalation flow rate on their delivery performance is studied. Delivery performance of pollen-shape carrier with petal-like surface morphology is compared with conventional rock-like LA carrier of a similar size range with different drug mixing ratios at 30 and 60 L/min. HA carriers show higher drug attachment ability than LA carriers. Significant amount of the drug particles is found to remain unattached in the blends with LA carriers. The excess drug particles mostly lost in the inhaler and reduce the aerosolization and deposition performance of the blends. The results of the *in vitro* experiments of the HA blends show improved ED and FPF than the LA blends, especially at high drug mixing ratios and low gas flow rate of 30 L/min. The HA blends show high ED of 82–90% at 30 L/min while the LA blends are observed to have ED of 69–82% at the same conditions. The high emission of the HA blends also allows high FPF of 10–18% while the FPF of

the LA blends are 3–15%. At a gas flow rate of 60 L/min, all the HA blends show better ED (83–95% for HA and 82–84% for LA) and compatible FPF (19–41% for HA and 21–34% for LA) against the LA blends. This study ensures the preferable outcome of pollen-shape carrier particles in dry powder formulation.

It is found that morphology of the pollen-shape carriers is the main contributor in their good delivery performance. Therefore, the impact of size and surface morphology of pollen-shape particles on their delivery performance is investigated. HA carrier with a geometric diameter size of 27 μm with needle-like surface morphology and larger specific surface is compared with another HA carrier of 48 μm with petal-like surface morphology. Their drug attachment, *in vitro* aerosolization and deposition behavior are examined with different drug mixing ratios and flow rate. Smaller HA carriers exhibit higher drug attachment ability than the larger HA carriers. Size, surface morphology of the carrier, and gas flow rate are observed to have stronger effect on ED than the drug mixing ratio for pollen-shape carriers. ED value of around 90% is found for the blends. The smaller-sized HA carriers exhibit lower aerosolization than the larger-sized HA. Hence, at low gas flow rate, blends with smaller HA carriers show earlier deposition. Eventually, they have lower FPF than the blends with larger HA carriers. However, at high flow rate smaller HA carriers are dispersed well as discrete particles. Therefore, at low drug mixing ratio, they can travel to the lower stages together with the drug particles. This study proves that pollen-shape particles with different size and morphology are suitable as drug carriers in dry powder inhalation.

On the whole, it is found that particle shape has significant effect on their aerodynamic behavior. Pollen-shape particles are found to have better flow properties

compare to other shape particles in similar size range. The flow behavior of the pollen-shape particles is also proved suitable to flow through inhalation path using PIV technique. Drug blends with large pollen-shape particles showed higher ED and FPF than traditional LA carriers in *in vitro* aerosolization and deposition experimentations. Drug attachment capacity of the pollen shape particles is significantly higher than LA carriers. They showed significant improvement in delivery performance especially with high drug mixing ratio and low flow rate. Moreover, pollen-shape particles with different size and surface morphology showed excellent drug loading in dry formulation, aerosolization and flowability to improve the overall delivery efficiency of the blends. A diverse application would be possible with this novel formulation comprising the particles with pollen-like morphology if additional improvement in the drug liberation is achieved.

10.2 Recommendations

To apply the benefits of pollen shape surface morphology to drug and carrier particles additional studies need to be conducted. The future recommended studies are listed here:

Effect of fine particles and surface modification for the pollen-shape carrier

Pollen-shape carrier particles show higher possibility in drug loading and inhalation performance for dry powder inhalation. However, they have limitations in drug liberation. The respirable fraction of drug particles blended with carriers can be increased more by improving the liberation. Two most common techniques for the improvement are using of fine particles in the blend preparation and surface modification of carrier particles. Fine particles in the binary mixture of drug and carrier can improve the delivery efficiencies. However, the effect of these fine particles for carriers with pollen-like surface morphology is not revealed yet. Generally, fine particles fill up the active sites on the surface of the carrier and let the drug particles to attach to less active sites. This will help the liberation of the drug particles from the carrier. Due to their morphology, pollen-shape carriers have larger specific surface area than traditional LA carriers. Hence, the effect of concentration and physical properties of the fine particles to improve the inhalation performance of the blends with pollen-shape carrier particles need to be studied.

Surface modification of the carrier particles has significant influence on their delivery performance. A small amount of surface roughness can improve the overall performance of the carriers. However, the effectiveness of surface treatment for the carriers with pollen-shape surface morphology is yet to known. A standard technique of

surface modification e.g. thin surface coating can be applied to the pollen-shape carriers before the mixing with drug particles. Surface modification can improve the drug liberation for higher delivery.

Effect of inhaler design

Drug liberation from carrier particles is mainly driven by shear forces and turbulences generated by the airflow in the inhaler and the initial airways.^{21,40,41} The liberated drug particles are carried into the deep lung by the air flow to exhibit proper clinical outcome. Inhaler design critically influence the inhalation flow rate and turbulence occurrence inside the inhaler. Hence, the flow field and circulation of air inside the inhaler to produce higher turbulence and shear force need to be investigated extensively to correlate with overall inhalation performance. An optimized design of the inhaler with sufficient amount of turbulence need to be developed to deliver the drug properly into the lung. An ideal inhaler can help to achieve further improvement in delivery performance of the formulation with pollen-shape carrier particles.

Synthesis of pollen-shape particles with high porosity

Pollen-shape can attribute high surface porosity and low surface density to the particles. It is found that pollen-shape particles have excellent flowability and dispersion quality. With this property, it is possible to produce some particles with large geometric diameter and low aerodynamic diameter which would be suitable for better deep lung deposition. Moreover, due to their large geometric size, they would be aerosolized easily without the help of large carrier particles. Drug particles would be loaded by adsorption technique to these particles and *in vitro* techniques would be followed to assess their feasibility in drug delivery.

Reference

1. Kehlenbeck R, Di Felice R 1999. Empirical relationships for the terminal settling velocity of spheres in cylindrical columns. *Chemical Engineering & Technology* 22(4):303-308.
2. Di Felice R 1996. A relationship for the wall effect on the settling velocity of a sphere at any flow regime. *International Journal of Multiphase Flow* 22(3):527-533.
3. Thompson Philip J 1998. Drug delivery to the small airways. *American Journal of Respiratory and Critical Care Medicine* 157(5):199S-202.
4. www.nextsafety.com.
5. O'Callaghan C, Nerbrink O, Vidgren MT. 2001. The history of inhaled drug therapy. In Bisgaard H, O'Callaghan C, Smaldone GC, editors. *Drug delivery to the lung*, ed., New York: Marcel Dekker, Inc.
6. Patton JS, Byron PR 2007. Inhaling medicines: delivering drugs to the body through the lungs. *Nature Reviews Drug Discovery* 6(1):67-74.
7. Patton JS 1996. Mechanisms of macromolecule absorption by the lungs. *Advanced Drug Delivery Reviews* 19(1):3-36.
8. Weibel ER. 1963. *Morphometry of human lung*. 1st ed., New York: Academic Press.
9. Owens DR, Zinman B, Bolli G 2003. Alternative routes of insulin delivery. *Diabetic Medicine* 20(11):886-898.
10. Gonda I. 2004. Targeting by deposition. In Hickey AJ, editor *Pharmaceutical inhalation aerosol technology*, 2nd ed., New York: Marcel Dekker, Inc. p 65-88.
11. Labiris NR, Dolovich MB 2003. Pulmonary drug delivery. Part I: Physiological factors affecting therapeutic effectiveness of aerosolized medications. *British Journal of Clinical Pharmacology* 56(6):588-599.
12. Otulana T, Thippawong J. 2002. Systemic delivery of small molecules: Product development issues focused on pain therapeutics. *Respiratory Drug Delivery VIII*, 1st ed., Arizona. p 97-104

13. Rabinowitz JD, Wensley M, Lloyd P, Myers D, Shen W, Lu A, Hodges C, Hale R, Mufson D, Zaffaroni A 2004. Fast onset medications through thermally generated aerosols. *Journal of Pharmacology and Experimental Therapeutics* 309(2):769-775.
14. RK W 1998. Safety of inhaled proteins for therapeutic use. *Journal of Aerosol Medicine* 11:197-219.
15. Byron PR, Patton JS 1994. Drug delivery via the respiratory tract. *Journal of Aerosol Medicine* 7:49-75.
16. Ann T, Bo N, Hans M, Anna-Karin W, Hans L, Ursula Hultkvist B 2003. Pulmonary absorption rate and bioavailability of drugs *in vivo* in rats: Structure-absorption relationships and physicochemical profiling of inhaled drugs. *Journal of Pharmaceutical Sciences* 92(6):1216-1233.
17. O'Connor BJ 2004. The ideal inhaler: Design and characteristics to improve outcomes. *Respiratory Medicine* 98(Supplement 1):S10-S16.
18. Bisgaard H 1997 Delivery of inhaled medication to children *Journal of Asthma* 34(6):443-467.
19. Partridge MR 1994. Metered-dose inhalers and CFCs: what respiratory physicians need to know. *Respiratory Medicine* 88(9):645-647.
20. Shaheen MZ, Ayres JG, Benincasa C 1994. Incidence of acute decreases in peak expiratory flow following the use of metered dose inhalers in asthmatic patients. *European respiratory journal* 7(12):2160-2164.
21. Newman SP, Busse WW 2002. Evolution of dry powder inhaler design, formulation, and performance. *Respiratory Medicine* 96(5):293-304.
22. Thorsson L, Edsbäcker S, Conradson T-B 1994. Lung deposition of budesonide from turbuhaler® is twice that from a pressurized metered-dose inhaler pMDI. *European Respiratory Journal* 7:1839-1844.
23. Agertoft L, Pedersen S 1994. Budesonide administered via Turbuhaler® and a nebulator. *Ugeskr Laeger* 156(28):4134-4137.
24. Chapman KR, Love L, Brubaker H 1993. A comparison of breath-actuated and conventional metered-dose inhaler inhalation techniques in elderly subjects. *Chest* 104:1332-1337.

25. Taylor A, Gustafsson P 2005. Do all dry powder inhalers show the same pharmaceutical performance? *International Journal of Clinical Practice* 59(s149):7-12.
26. Hickey AJ, Dunbar CA 1997. A new millennium for inhaler technology. *Pharmacy Technology* 21:116-125.
27. Prime D, Atkins PJ, Slater A, Sumby B 1997. Review of dry powder inhalers. *Advanced Drug Delivery Reviews* 26:51-58.
28. Crowder TM, Louey MD, Sethuraman VV, Smyth HDC, Hickey. AJ 2001. 2001: An Odyssey in inhaler formulations and design. *Pharmacy Technology* 25:99-113.
29. Crowder TM, Rosati JA, Schroeter JD, Hickey AJ, Martonen TB 2002. Fundamental effects of particle morphology on lung delivery: Predictions of Stokes' law and the particular relevance to dry powder inhaler formulation and development. *Pharmaceutical Research* 19(3):239-245.
30. Gonda I 1990. Aerosols for delivery of therapeutic and diagnostic agents to the respiratory tract. *Critical reviews in therapeutic drug carrier systems* 6(4):273-313.
31. Newman SP, Agnew JE, Pavia D, Clarke SW 1982. Inhaled aerosols: Lung deposition and clinical applications. *Clinical Physics and Physiological Measurement* 3(1):1-20.
32. Yeh HC, Phalen RF, Raabe OG 1976. Factors influencing the deposition of inhaled particles. *Environmental Health Perspectives* 15:147-156.
33. Phipps PR, Gonda I, Anderson SD, Bailey D, Bautovich G 1994. Regional deposition of saline aerosols of different tonicities in normal and asthmatic subjects. *European Respiratory Journal* 7(8):1474-1482.
34. Ferron GA, Hornik S, Kreyling WG, Haider B 1985. Comparison of experimental and calculated data for the total and regional deposition in the human lung. *Journal of Aerosol Science* 16(2):133-143.
35. Ferron GA, Oberdörster G, Henneberg R 1989. Estimation of the deposition of aerosolized drugs in the human respiratory tract due to hygroscopic growth. *Journal of Aerosol Medicine* 2(3):261-270.
36. Smith S, Cheng Y-S, Yeh HC 2001. Deposition of ultrafine particles in human tracheobronchial airways of adults and children. *Aerosol Science and Technology* 35(3):697 - 709.

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37. Martonen TB, Katz IM 1993. Deposition patterns of aerosolized drugs within human lungs: Effects of ventilatory parameters. *Pharmaceutical Research* 10(6):871-878.
 38. Farr S, Gonda I, Licko V. 1998. Physicochemical and physiological factors influencing the effectiveness of inhaled insulin. In Dalby R, Byron P, Farr S, editors. *Respiratory drug delivery* 6th ed., Buffalo Grove, IL: Interpharm Press Inc. p 25–33.
 39. Heinemann L, Traut T, Heise T 1997. Time-action profile of inhaled insulin. *Diabetic Medicine* 14(1):63-72.
 40. French DL, Edwards DA, Niven RW 1996. The influence of formulation on emission, deaggregation and deposition of dry powders for inhalation. *Journal of Aerosol Science* 27(5):769-783.
 41. Voss A, Finlay WH 2002. Deagglomeration of dry powder pharmaceutical aerosols. *International Journal of Pharmaceutics* 248(1-2):39-50.
 42. Folkesson HG, Matthay MA, Westrom BR, Kim KJ, Karlsson BW, Hastings RH 1996. Alveolar epithelial clearance of protein. *Journal of Applied Physiology* 80(5):1431-1445.
 43. Dolovich M, Newhouse M. 1993. Aerosols. Generation, methods of administration, and therapeutic applications in asthma. In Middleton EJ, Reed C, Ellis E, Adkinson NJ, Yunginger J, Busse W, editors. *Allergy Principles and practice*, 1st ed., St Louis: Mosby Year Book, Inc., p 712–739.
 44. Lourenco R, Cotromanes E 1982. Clinical aerosols I. Characterization of aerosols and their diagnostic uses. *Archives of Internal Medicine* 142:2163–2172.
 45. Brain J, Blanchard J. 1993. Mechanisms of aerosol deposition and clearance. In Moren F, Newhouse M, Dolovich M, editors. *Aerosols in medicine principles, diagnosis and therapy*, 2nd ed., New York: Elsevier Science Publishers (Biomedical Division). p 117–156.
 46. Heyder J 1982. Particle transport onto human airway surfaces. *European Journal of Respiratory Diseases* 63(Suppl 119):29–50.
 47. Gerrity T. 1990. Pathophysiological and disease constraints on aerosol deposition. In Byron P, editor *Respiratory drug delivery*, 1st ed., Boca Raton, FL: CRC Press, Inc. p 1-38.
-

-
48. Coates M, Chan H-K, Fletcher D, Chiou H 2007. Influence of mouthpiece geometry on the aerosol delivery performance of a dry powder inhaler. *Pharmaceutical Research* 24(8):1450-1456.
 49. Matthew SC, David FF, Hak-Kim C, Judy AR 2004. Effect of design on the performance of a dry powder inhaler using computational fluid dynamics. Part 1: Grid structure and mouthpiece length. *Journal of Pharmaceutical Sciences* 93(11):2863-2876.
 50. Coates MS, Chan H-K, Fletcher DF, Raper JA 2006. Effect of design on the performance of a dry powder inhaler using computational fluid dynamics. Part 2: Air inlet size. *Journal of Pharmaceutical Sciences* 95(6):1382-1392.
 51. Podczek F 1999. The influence of particle size distribution and surface roughness of carrier particles on the *in vitro* properties of dry powder inhalations. *Aerosol Science and Technology* 31(4):301 - 321.
 52. Hinds WC. 1999. *Aerosol technology: Properties, behavior, and measurement of airborne particles*. 2nd ed., New York: John Wiley & Sons, Inc.
 53. Heyder J 2004. Deposition of inhaled particles in the human respiratory tract and consequences for regional targeting in respiratory drug delivery. *Proceedings of the American Thoracic Society* 1(4):315-320.
 54. Vincent JH. 1989. *Aerosol Sampling*. 1st ed., Chichester: John Wiley and Sons.
 55. Esmen NA 1996. Adhesion and aerodynamic resuspension of fibrous particles. *Journal of Environmental Engineering* 122:379-383.
 56. Warnke G, König D, Bauer K-H, Brandl M 1994. Aerosol-fractionation performance of a multistage liquid impinger: Evaluation by means of light microscopy and a light extinction particle counter. *Pharmaceutical and Pharmacological Letters* 4:5-7.
 57. Hindle M, Byron PR 1995. Size distribution control of raw materials for dry powder inhalers using the aerosizer with the aero-disperser. *Pharmacy Technology* 19:64-78.
 58. Edwards DA 2002. Delivery of biological agents by aerosols. *AIChE Journal* 48(1):2-6.
 59. Harris RL, Fraser DA 1976. A model for deposition of fibers in the human respiratory system. *American Industrial Hygiene Association Journal* 37(2):73 - 89.
-

-
60. Gradon L, Podgorski A 1992. Experimental study on fibrous particle deposition in the human nasal cast *Journal of Aerosol Science* 23(Suppl. 1):S469-S472.
 61. Martonen TB, Johnson DL 1990. Aerodynamic size classification of fibers with aerosol centrifuges. *Particulate Science and Technology* 8:37-53.
 62. Chan H-K, Gonda I 1989. Aerodynamic properties of elongated particles of cromoglycic acid. *Journal of Aerosol Science* 20(2):157-168.
 63. Vaghi A, Berg E, Liljedahl S, Svensson JO 2005. *In vitro* comparison of nebulised budesonide (Pulmicort Respules®) and beclomethasone dipropionate (Clenil® per Aerosol). *Pulmonary Pharmacology & Therapeutics* 18(2):151-153.
 64. Balásházy I, Martonen TB, Hofmann W 1990. Fiber deposition in airway bifurcations. *Journal of Aerosol Medicine* 3:243-260.
 65. Visser J 1989. Van der Waals and other cohesive forces affecting powder fluidization. *Powder Technology* 58(1):1-10.
 66. Watanabe M, Ozeki T, Shibata T, Murakoshi H, Takashima Y, Yuasa H, Okada H 2003. Effect of shape of sodium salicylate particles on physical property and *in vitro* aerosol performance of granules prepared by pressure swing granulation method. *AAPS PharmSciTech* 4(4).
 67. Edwards DA, Hanes J, Caponetti G, Hrkach J, Ben-Jebria A, Eskew ML, Mintzes J, Deaver D, Lotan N, Langer R 1997. Large porous particles for pulmonary drug delivery. *Science* 276(5320):1868.
 68. Edwards DA, Ben-Jebria A, Langer R 1998. Recent advances in pulmonary drug delivery using large, porous inhaled particles. *Journal of Applied Physiology* 85(2):379-385.
 69. Musante CJ, Schroeter JD, Rosati JA, Crowder TM, Hickey AJ, Martonen TB 2002. Factors affecting the deposition of inhaled porous drug particles. *Journal of Pharmaceutical Sciences* 91(7):1580-1590.
 70. Tong HHY, Yu Shekunov B, York P, Chow AHL 2001. Characterization of two polymorphs of salmeterol xinafoate crystallized from supercritical fluids. *Pharmaceutical Research* 18(6):852-858.
 71. Rita V, Abdellaziz B-J, Jeffrey DM, Robert L, David AE 1999. Sustained release of insulin from insoluble inhaled particles. *Drug Development Research* 48(4):178-185.
-

-
72. Bot AI, Tarara TE, Smith DJ, Bot SR, Woods CM, Weers JG 2000. Novel lipid-based hollow-porous microparticles as a platform for immunoglobulin delivery to the respiratory tract. *Pharmaceutical Research* 17(3):275-283.
 73. Bot A, Smith D, Bot S, Dellamary L, Tarara T, Harders S, Phillips W, Weers J, Woods C 2001. Receptor-mediated targeting of spray-dried lipid particles coformulated with immunoglobulin and loaded with a prototype vaccine. *Pharmaceutical Research* 18(7):971-979.
 74. Dellamary LA, Tarara TE, Smith DJ, Woelk CH, Adrastas A, Costello ML, Gill H, Weers JG 2000. Hollow porous particles in metered dose inhalers. *Pharmaceutical Research* 17(2):168-174.
 75. Duddu SP, Sisk SA, Walter YH, Tarara TE, Trimble KR, Clark AR, Eldon MA, Elton RC, Pickford M, Hirst PH, Newman SP, Weers JG 2002. Improved lung delivery from a passive dry powder inhaler using an engineered PulmoSphere® powder. *Pharmaceutical Research* 19(5):689-695.
 76. Hirst PH, Pitcairn GR, Weers JG, Tarara TE, Clark AR, Dellamary LA, Hall G, Shorr J, Newman SP 2002. *In vivo* lung deposition of hollow porous particles from a pressurized metered dose inhaler. *Pharmaceutical Research* 19(3):258-264.
 77. Hadinoto K, Phanapavudhikul P, Kewu Z, Tan RBH 2006. Novel formulation of large hollow nanoparticles aggregates as potential carriers in inhaled delivery of nanoparticulate drugs. *Industrial & Engineering Chemistry Research* 45(10):3697-3706.
 78. Hadinoto K, Phanapavudhikul P, Kewu Z, Tan RBH 2007. Dry powder aerosol delivery of large hollow nanoparticulate aggregates as prospective carriers of nanoparticulate drugs: Effects of phospholipids. *International Journal of Pharmaceutics* 333(1-2):187-198.
 79. Hadinoto K, Zhu K, Tan RBH 2007. Drug release study of large hollow nanoparticulate aggregates carrier particles for pulmonary delivery. *International Journal of Pharmaceutics* 341(1-2):195-206.
 80. Tsapis N, Bennett D, Jackson B, Weitz DA, Edwards DA 2002. Trojan particles: Large porous carriers of nanoparticles for drug delivery. *Proceedings of the National Academy of Sciences of the United States of America* 99(19):12001-12005.
-

-
81. Sung JC, Pulliam BL, Edwards DA 2007. Nanoparticles for drug delivery to the lungs. *Trends in Biotechnology* 25(12):563-570.
 82. Sung J, Padilla D, Garcia-Contreras L, VerBerkmoes J, Durbin D, Peloquin C, Elbert K, Hickey A, Edwards D 2009. Formulation and pharmacokinetics of self-assembled rifampicin nanoparticle systems for pulmonary delivery. *Pharmaceutical Research* 26(8):1847-1855.
 83. Grenha A, Seijo B, Remuñán-López C 2005. Microencapsulated chitosan nanoparticles for lung protein delivery. *European Journal of Pharmaceutical Sciences* 25(4-5):427-437.
 84. Grenha A, Grainger CI, Dailey LA, Seijo B, Martin GP, Remuñán-López C, Forbes B 2007. Chitosan nanoparticles are compatible with respiratory epithelial cells *in vitro*. *European Journal of Pharmaceutical Sciences* 31(2):73-84.
 85. Bailey MM, Gorman EM, Munson EJ, Berkland C 2008. Pure insulin nanoparticle agglomerates for pulmonary delivery. *Langmuir* 24(23):13614-13620.
 86. Peek LJ, Roberts L, Berkland C 2008. Poly(D,L-lactide-co-glycolide) nanoparticle agglomerates as carriers in dry powder aerosol formulation of proteins. *Langmuir* 24(17):9775-9783.
 87. Azarmi S, Tao X, Chen H, Wang Z, Finlay WH, Löbenberg R, Roa WH 2006. Formulation and cytotoxicity of doxorubicin nanoparticles carried by dry powder aerosol particles. *International Journal of Pharmaceutics* 319(1-2):155-161.
 88. Shi L, Plumley CJ, Berkland C 2007. Biodegradable nanoparticle flocculates for dry powder aerosol formulation. *Langmuir* 23(22):10897-10901.
 89. Morrow PE 1986. Factors determining hygroscopic aerosol deposition in airways. *Physiological Reviews* 66:330-376.
 90. Martonen TB 1982. Analytical model of hygroscopic particle behavior in human airways. *Bulletin of Mathematical Biology* 44:425-442.
 91. Hickey AJ, Gonda I, Irwin WJ, Fildes FJT 1990. The effect of hydrophobic coating upon the behavior of a hygroscopic aerosol powder in an environment of controlled temperature and relative humidity. *Journal of Pharmaceutical Sciences* 79:1009-1014.
-

-
92. Hickey AJ, Martonen TB 1993. The effect of hydrophobic coating upon the behavior of a hygroscopic aerosol powder in an environment of controlled temperature and relative humidity. *Journal of Pharmaceutical Sciences* 10:1-7.
 93. Broday DM, Georgopoulos PG 2001. Growth and deposition of hygroscopic particulate matter in the human lungs. *Aerosol Science and Technology* 34(1):144 - 159.
 94. Louey M, Van Oort M, Hickey A 2004. Aerosol dispersion of respirable particles in narrow size distributions produced by jet-milling and spray-drying techniques. *Pharmaceutical Research* 21(7):1200-1206.
 95. Islam N, Stewart P, Larson I, Hartley P 2004. Effect of carrier size on the dispersion of salmeterol xinafoate from interactive mixtures. *Journal of Pharmaceutical Sciences* 93(4):1030-1038.
 96. Steckel H, Müller BW 1997. *In vitro* evaluation of dry powder inhalers II: Influence of carrier particle size and concentration on *in vitro* deposition. *International Journal of Pharmaceutics* 154(1):31-37.
 97. Zimon AD. 1982. Adhesion of dust and powder. 1st ed., New York: Consultants Bureau.
 98. Kawashima Y, Serigano T, Hino T, Yamamoto H, Takeuchi H 1998. Effect of surface morphology of carrier lactose on dry powder inhalation property of pranlukast hydrate. *International Journal of Pharmaceutics* 172(1-2):179-188.
 99. Larhrib H, Zeng XM, Martin GP, Marriott C, Pritchard J 1999. The use of different grades of lactose as a carrier for aerosolised salbutamol sulphate. *International Journal of Pharmaceutics* 191(1):1-14.
 100. Larhrib H, Martin GP, Marriott C, Prime D 2003. The influence of carrier and drug morphology on drug delivery from dry powder formulations. *International Journal of Pharmaceutics* 257(1-2):283-296.
 101. Larhrib H, Cespi M, Dyas MA, Roberts M, Ford JL. 2006. Engineered carrier with a long time of flight (TOF) to improve drug delivery from dry powder inhalation aerosols. *Drug Delivery to the Lung* 17, Edinburgh.
 102. Larhrib H, Martin GP, Prime D, Marriott C 2003. Characterisation and deposition studies of engineered lactose crystals with potential for use as a carrier for aerosolised
-

salbutamol sulfate from dry powder inhalers. *European Journal of Pharmaceutical Sciences* 19(4):211-221.

103. Guchardi R, Frei M, John E, Kaerger JS 2008. Influence of fine lactose and magnesium stearate on low dose dry powder inhaler formulations. *International Journal of Pharmaceutics* 348(1-2):10-17.

104. Flament M-P, Leterme P, Gayot A 2004. The influence of carrier roughness on adhesion, content uniformity and the *in vitro* deposition of terbutaline sulphate from dry powder inhalers. *International Journal of Pharmaceutics* 275(1-2):201-209.

105. Zeng X-M, Martin GP, Marriott C, Pritchard J 2001. Lactose as a carrier in dry powder formulations: The influence of surface characteristics on drug delivery. *Journal of Pharmaceutical Sciences* 90(9):1424-1434.

106. Iida K, Hayakawa Y, Okamoto H, Danjo K, Leuenberger H 2001. Evaluation of flow properties of dry powder inhalation of salbutamol sulfate with lactose carrier. *Chemical & Pharmaceutical Bulletin* 49 (10):1326.

107. Zeng XM, Martin GP, Marriott C, Pritchard J 2000. The influence of carrier morphology on drug delivery by dry powder inhalers. *International Journal of Pharmaceutics* 200(1):93-106.

108. Heng PWS, Chan LW, Lim LT 2000. Quantification of the surface morphologies of lactose carriers and their effect on the *in vitro* deposition of salbutamol sulphate. *Chemical & Pharmaceutical Bulletin* 48(3):393-398.

109. Zeng XM, Martin GP, Tee S-K, Ghoush AA, Marriott C 1999. Effects of particle size and adding sequence of fine lactose on the deposition of salbutamol sulphate from a dry powder formulation. *International Journal of Pharmaceutics* 182(2):133-144.

110. Srichana T, Martin GP, Marriott C 1998. On the relationship between drug and carrier deposition from dry powder inhalers *in vitro*. *International Journal of Pharmaceutics* 167(1-2):13-23.

111. Bell JH, Hartley PS, Cox JSG 1971. Dry powder aerosols I: A new powder inhalation device. *Journal of Pharmaceutical Sciences* 60:1559-1564.

112. Young PM, Edge S, Traini D, Jones MD, Price R, El-Sabawi D, Urry C, Smith C 2005. The influence of dose on the performance of dry powder inhalation systems. *International Journal of Pharmaceutics* 296(1-2):26-33.

-
113. Louey M, Van Oort M, Hickey A 2004. Aerosol dispersion of respirable particles in narrow size distributions using drug-alone and lactose-blend formulations. *Pharmaceutical Research* 21(7):1207-1213.
 114. Begat P, Morton DAV, Staniforth JN, Price R 2004. The cohesive-adhesive balances in dry powder inhaler formulations II: Influence on fine particle delivery characteristics. *Pharmaceutical Research* 21(10):1826-1833.
 115. Louey MD, Razia S, Stewart PJ 2003. Influence of physico-chemical carrier properties on the *in vitro* aerosol deposition from interactive mixtures. *International Journal of Pharmaceutics* 252(1-2):87-98.
 116. Kassem NM, Ganderton D 1990. The influence of carrier surface on the characteristics of inspirable powder aerosols. *Journal of Pharmacy and Pharmacology* 42:11P.
 117. Braun MA, Oschmann R, Schmidt PC 1996. Influence of excipients and storage humidity on the deposition of disodium cromoglycate (DSCG) in the twin impinger. *International Journal of Pharmaceutics* 135(1-2):53-62.
 118. Ganderton D 1992. The generation of respirable cloud from coarse powder aggregates. *Journal of Biopharmaceutical Sciences* 3:101-105.
 119. Adi H, Larson I, J. PS 2009. Influence of the polydispersity of the added fine lactose on the dispersion of salmeterol xinafoate from mixtures for inhalation. *European Journal of Pharmaceutical Sciences* 36(2-3):265-274.
 120. Lai Wah C, Liang Theng L, Paul WSH 2003. Immobilization of fine particles on lactose carrier by precision coating and its effect on the performance of dry powder formulations. *Journal of Pharmaceutical Sciences* 92(5):975-984.
 121. Louey MD, Stewart PJ 2002. Particle interactions involved in aerosol dispersion of ternary interactive mixtures. *Pharmaceutical Research* 19(10):1524-1531.
 122. Zeng XM, Martin GP, Tee S-K, Marriott C 1998. The role of fine particle lactose on the dispersion and deaggregation of salbutamol sulphate in an air stream *in vitro*. *International Journal of Pharmaceutics* 176(1):99-110.
 123. Lucas P, Anderson K, Staniforth JN 1998. Protein deposition from dry powder inhalers: Fine particle multiplets as performance modifiers. *Pharmaceutical Research* 15(4):562-569.
-

-
124. Zeng XM, Pandhal KH, Martin GP 2000. The influence of lactose carrier on the content homogeneity and dispersibility of beclomethasone dipropionate from dry powder aerosols. *International Journal of Pharmaceutics* 197(1-2):41-52.
125. Wang Y, Hassan MS, Gunawan P, Lau R, Wang X, Xu R 2009. Polyelectrolyte mediated formation of hydroxyapatite microspheres of controlled size and hierarchical structure. *Journal of Colloid and Interface Science* 339(1):69-77.
126. Wang C, He C, Tong Z, Liu X, Ren B, Zeng F 2006. Combination of adsorption by porous CaCO₃ microparticles and encapsulation by polyelectrolyte multilayer films for sustained drug delivery. *International Journal of Pharmaceutics* 308(1-2):160-167.
127. Yu J, Tang H, Cheng B, Zhao X 2004. Morphological control of calcium oxalate particles in the presence of poly-(styrene-alt-maleic acid). *Journal of Solid State Chemistry* 177(10):3368-3374.
128. Yu J, Lei M, Cheng B, Zhao X 2004. Facile preparation of calcium carbonate particles with unusual morphologies by precipitation reaction. *Journal of Crystal Growth* 261(4):566-570.
129. Lucas A, Gaude' J, Carel C, Michel J-F, Cathelineau G 2001. A synthetic aragonite-based ceramic as a bone graft substitute and substrate for antibiotics. *International Journal of Inorganic Materials* 3:87-94.
130. Hickey AJ, Concessio NM 1994. Flow properties of selected pharmaceutical powders from a vibrating spatula. *Particle & Particle Systems Characterization* 11(6):457-462.
131. Anthony JH, Heidi MM, Martin JT, Zhen X, Hugh DCS, Tako M, Richard M, John L, Dimitris P 2007. Physical characterization of component particles included in dry powder inhalers. II. Dynamic characteristics. *Journal of Pharmaceutical Sciences* 96(5):1302-1319.
132. Chhabra RP 1996. Wall effects on terminal velocity of non-spherical particles in non-Newtonian polymer solutions. *Powder Technology* 88(1):39-44.
133. Madhav GV, Chhabra RP 1995. Drag on non-spherical particles in viscous fluids. *International Journal of Mineral Processing* 43(1-2):15-29.
134. Chhabra RP 1995. Wall effects on free-settling velocity of non-spherical particles in viscous media in cylindrical tubes. *Powder Technology* 85(1):83-90.
-

135. Madhav GV, Chhabra RP 1994. Settling velocities of non-spherical particles in non-Newtonian polymer solutions. *Powder Technology* 78(1):77-83.
136. Newton I. 1687. *Principia*. Lib. II, Prop. XXXIX, Theor. XXXI. 1st ed., Cambridge: Cambridge University Press.
137. Francis AW 1933. Wall effect in falling ball method for viscosity. *Physics* 4:403.
138. Fidleris V, Whitmore RL 1961. Experimental determination of the wall effect for spheres falling axially in cylindrical vessels. *British Journal of Applied Physics* 12:490-494.
139. Di Felice R, Kehlenbeck R 2000. Sedimentation velocity of solids in finite size vessels. *Chemical Engineering & Technology* 23(12):1123-1126.
140. Di Felice R, Parodi E 1996. Wall effects on the sedimentation velocity of suspensions in viscous flow. *AIChE Journal* 42(4):927-931.
141. Munroe HS 1888. The english versus the continental system of jigging-is close sizing advantageous? *Transactions of the American Institute of Mining and Metallurgical Engineers* 17:637-659.
142. Bougas A, Stamatoudis M 1993. Wall factor for acceleration and terminal velocity of falling spheres at high Reynolds numbers. *Chemical Engineering & Technology* 16:314-317.
143. Brown PP, Lawler DF 2003. Sphere Drag and settling velocity revisited. *Journal of Environmental Engineering* 129(3):222.
144. Christiansen EB, Barker DH 1965. The effect of shape and density on the free settling of particles at high Reynolds numbers. *AIChE Journal* 11(1):145-151.
145. Reynolds PA, Jones TER 1989. An experimental study of the settling velocities of single particles in non-Newtonian fluids. *International Journal of Mineral Processing* 25(1-2):47-77.
146. Singh AN, Roychowdhury KC 1969. Study of the effects of orientation and shape on the settling velocity of non-isometric particles. *Chemical Engineering Science* 24(7):1185-1186.
147. Pettyjohn ES, Christiansen EB 1948. Effect of particle shape on free-settling rates of isometric particles. *Chemical Engineering Progress* 44(2).

-
148. Unnikrishnana A, Chhabra RP 1991. An experimental study of motion of cylinders in newtonian fluids: Wall effects and drag coefficient. *Canadian Journal of Chemical Engineering* 69:729.
 149. Kasper G, Niida T, Yang M 1985. Measurements of viscous drag on cylinders and chains of spheres with aspect ratios between 2 and 50. *Journal of Aerosol Science* 16:535-556.
 150. Yin C, Rosendahl L, Knudsen Kaer S, Sorensen H 2003. Modelling the motion of cylindrical particles in a nonuniform flow. *Chemical Engineering Science* 58(15):3489-3498.
 151. Baron PA, Willeke K. 2001. *Aerosol measurement: Principles, techniques and applications*. 2nd ed., New York: Willy-Interscience.
 152. Tang P, Chan HK, Raper JA 2004. Prediction of aerodynamic diameter of particles with rough surfaces. *Powder Technology* 147(1-3):64-78.
 153. Fults KA, Miller IF, Hickey AJ 1997. Effect of particle morphology on emitted dose of fatty acid-treated disodium cromoglycate powder aerosols. *Pharmaceutical Development and Technology* 2(1):67 -79.
 154. Otsuka A, Iida K, Danjo K, Sunda H 1988. Measurement of the adhesive forces between particles of powdered materials and a glass substrate by means of the impact separation method. II. Effects of particle shape and surface asperity. *Chemical & Pharmaceutical Bulletin* 36:741–749.
 155. Zimon AD. 1982. *Adhesion of dust and powder*. 2nd ed., London: Consultant Bureau. p 46–47.
 156. Mullins ME, Michaels LP, Menon V, Locke B, Ranade MB 1992. Effect of geometry on particle adhesion. *Aerosol Science and Technology* 17(2):105 - 118.
 157. Bailey AG 1984. Electrostatic phenomena during powder handling. *Powder Technology* 37:71–85.
 158. Karra VK, Fuerstenau DW 1977. The effect of humidity on the trace mixing kinetics in fine powders. *Powder Technology* 16:97–105.
 159. Hassan MS, Lau R. 2009. Pollen shape particles for pulmonary drug delivery: *In vitro* study of flow and deposition properties. 13th International Conference on Biomedical Engineering, Singapore, pp 1434-1437.
-

-
160. Hassan M, Lau R 2009. Effect of particle shape on dry particle inhalation: Study of flowability, aerosolization, and deposition properties. *AAPS PharmSciTech* 10(4):1252-1262.
161. Davies CN 1979. Particle-fluid interaction. *Journal of Aerosol Science* 10(5):477-513.
162. Kanig JL, Lachman L, Lieberman HA. 1986. The theory and practice of industrial pharmacy. 3rd ed., Philadelphia: Lea & Febiger.
163. Cheng YS, Yazzie D, Gao J, Muggli D, Etter J, Rosenthal GJ 2003. Particle characteristics and lung deposition patterns in a human airway replica of a dry powder formulation of polylactic acid produced using supercritical fluid technology. *Journal of Aerosol Medicine* 16(1):65-73.
164. Svartengren K, Lindestad P, Svartengren M, Bylin G, Philipson K, Camner P 1994. Deposition of inhaled particles in the mouth and throat of asthmatic subjects *European Respiratory Journal* 7:1467-1473.
165. Stahlhofen W, Gebhart J, Heyder J 1980. Experimental determination of the regional deposition of aerosol particles in the human respiratory tract. *American Industrial Hygiene Association Journal* 41(6):385 - 398.
166. Chan TL, Lippmann M 1980. Experimental measurements and empirical modelling of the regional deposition of inhaled particles in humans. *American Industrial Hygiene Association Journal* 41(6):399 - 409.
167. Foord N, Black A, Walsh M 1978. Regional deposition of 2.5-7.5 μm diameter inhaled particles in healthy male non-smokers. *Journal of Aerosol Science* 9(4):343-357.
168. Emmett PC, Aitken RJ, Hannan WJ 1982. Measurements of the total and regional deposition of inhaled particles in the human respiratory tract. *Journal of Aerosol Science* 13(6):549-560.
169. Cass LMR, Brown J, Pickford M, Fayinka S, Newman SP, Johansson CJ, Bye A 1999. Pharmacoscintigraphic evaluation of lung deposition of inhaled zanamivir in healthy volunteers. *Clinical Pharmacokinetics* 36(6):21-31.
170. Zhou Y, Cheng Y-S 2005. Particle deposition in a cast of human tracheobronchial airways. *Aerosol Science and Technology* 39(6):492 - 500.
-

-
171. Cheng YS, Zhou Y, Chen BT 1999. Particle deposition in a cast of human oral airways. *Aerosol Science and Technology* 31(4):286 - 300.
172. Cheng K-H, Swift DL, Cheng Y-S, Yeh H-C, Swift DL 1997. An experimental method for measuring aerosol deposition efficiency in the human oral airway. *American Industrial Hygiene Association Journal* 58(3):207 - 213.
173. Swift DL 1992. Apparatus and method for measuring regional distribution of therapeutic aerosols and comparing delivery systems. *Journal of Aerosol Science* 23(Supplement 1):495-498.
174. Heenan A, Matida E, Pollard A, Finlay W 2003. Experimental measurements and computational modeling of the flow field in an idealized human oropharynx. *Experiments in Fluids* 35(1):70-84.
175. Grgic B, Finlay WH, Heenan AF 2004. Regional aerosol deposition and flow measurements in an idealized mouth and throat. *Journal of Aerosol Science* 35(1):21-32.
176. Grgic B, Finlay WH, Burnell PKP, Heenan AF 2004. *In vitro* intersubject and intrasubject deposition measurements in realistic mouth-throat geometries. *Journal of Aerosol Science* 35(8):1025-1040.
177. Martonen TB, Schroeter JD, Hwang D, Fleming JS, Conway JH 2000. Human lung morphology models for particle deposition studies. *Inhalation Toxicology* 12(11 supp 4):109 - 121.
178. Renotte C, Bouffieux V, Wilquem F 2000. Numerical 3D analysis of oscillatory flow in the time-varying laryngeal channel. *Journal of Biomechanics* 33(12):1637-1644.
179. Corcoran T, Chigier N 2000. Characterization of the laryngeal jet using Phase Doppler Interferometry. *Journal of Aerosol Medicine* 13:125-137.
180. Olson DE, Sudlow MF, Horsfield K, Filley GF 1973. Convective patterns of flow during inspiration. *Archives of Internal Medicine* 131:51-57.
181. Marchioli C, Soldati A, Kuerten JGM, Arcen B, Tanière A, Goldensoph G, Squires KD, Cargnelutti MF, Portela LM 2008. Statistics of particle dispersion in direct numerical simulations of wall-bounded turbulence: Results of an international collaborative benchmark test. *International Journal of Multiphase Flow* 34(9):879-893.
182. Jaeger HM, Nagel SR, Behringer RP 1996. The physics of granular matter. *Physics Today* 49:32-38.
-

183. Li Q, Rudolph V, Weigl B, Earl A 2004. Interparticle van der Waals force in powder flowability and compactibility. *International Journal of Pharmaceutics* 280(1-2):77-93.
184. Ikegami K, Kawashima Y, Takeuchi H, Yamamoto H, Isshiki N, Momose D-i, Ouchi K 2002. Improved inhalation behavior of steroid KSR-592 *in vitro* with Jethaler® by polymorphic transformation to needle-like crystals (β -form). *Pharmaceutical Research* 19(10):1439-1445.
185. Vanbever R, Mintzes JD, Wang J, Nice J, Chen D, Batycky R, Langer R, Edwards DA 1999. Formulation and physical characterization of large porous particles for inhalation. *Pharmaceutical Research* 16(11):1735-1742.
186. Corn M. 1966. Adhesion of particles in aerosol science. 1st ed., New York: Academic Press.
187. Podczek F 1997. The relationship between particulate properties of carrier materials and the adhesion force of drug particles in interactive powder mixtures. *Journal of Adhesion Science and Technology* 11:1089-1104.
188. Podczek F 1997. The development of a cascade impactor simulator based on adhesion force measurements to aid the development of dry powder inhalations. *Chemical & Pharmaceutical Bulletin* 45:911-917.
189. Podczek F 1998. Adhesion forces in interactive powder mixtures of a micronized drug and carrier particles of various particle size distributions. *Journal of Adhesion Science and Technology* 12:1323-1339.
190. Hassan MS, Lau R 2010. Feasibility study of pollen-shape drug carriers in dry powder inhalation. *Journal of Pharmaceutical Sciences* 99(3):1309-1321.