

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

CORRECTED VERSION

(19) World Intellectual Property Organization
International Bureau



(43) International Publication Date
4 February 2016 (04.02.2016)

(10) International Publication Number
WO 2016/019253 A9

(51) International Patent Classification:
A61K 9/14 (2006.01)

HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(21) International Application Number:
PCT/US2015/043128

(22) International Filing Date:
31 July 2015 (31.07.2015)

(25) Filing Language:
English

(26) Publication Language:
English

(30) Priority Data:
62/031,811 31 July 2014 (31.07.2014) US

(71) Applicant: OTITOPIC INC. [US/US]; 10435 Santa Monica Blvd, 1st Floor, Los Angeles, CA 90025 (US).

(72) Inventor: YADIDI, Kambiz; 717 Lakefield Road, #D, Westlake Village, CA 91361 (US).

(74) Agents: DAVITZ, Michael, A. et al.; Ascenda Law Group PC, 333 W. San Carlos St., Suite 200, San Jose, CA 95110 (US).

(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT,

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Published:

- with international search report (Art. 21(3))
- with information concerning authorization of rectification of an obvious mistake under Rule 91.3 (b) (Rule 48.2(i))

(48) Date of publication of this corrected version:

12 May 2016

(15) Information about Correction:
see Notice of 12 May 2016



WO 2016/019253 A9

(54) Title: DRY POWDER FORMULATIONS FOR INHALATION

(57) Abstract: A respirable dry powder including acetylsalicylic acid in particles having a mass median aerodynamic diameter (MMAD) within a range of about 0.5 μm to about 10 μm . The respirable dry powder may contain a pharmaceutically acceptable excipient, such as a phospholipid, in an amount ranging from about 0.1% (w/w) to about 10% (w/w) of the particles.

DRY POWDER FORMULATIONS FOR INHALATION

Cross Reference to Related Applications

This application claims priority to U.S. Provisional Application No. 62/031,811 (filed on July 31, 2014), which is incorporated herein by reference in its entirety.

FIELD

The subject technology relates generally to pulmonary delivery of dry powder formulations of nonsteroidal anti-inflammatories (NSAIDs), such as aspirin or acetylsalicylic acid. The subject technology also relates generally to apparatuses and methods for delivery of substances, e.g., medication, to the lungs by inhalation for treating disease, such as ischemic or thromboembolic events, including cardiovascular disease.

BACKGROUND

Pulmonary delivery of therapeutic agents offers several advantages over other modes of delivery. These advantages include rapid onset, the convenience of patient self-administration, the potential for reduced drug side-effects, ease of delivery by inhalation, the elimination of needles, and the like. Inhalation therapy is capable of providing a drug delivery system that is easy to use in an inpatient or outpatient setting, results in very rapid onset of drug action, and produces minimal side effects.

Dry powder inhalation offers the possibility of delivering accurate and reproducible doses of a drug to the pulmonary vasculature. Telko et al. Dry Powder Inhaler Formulation, Respiratory Care 50(9):1209 (2005). One problem that dry powder inhaler formulations encounter is that they have historically relied on lactose blending to allow for the dosing of particles that are small enough to be inhaled, but are not sufficiently dispersible on their own. Not only is this process inefficient, but also it does not work for some drugs. Several groups have tried to improve on these shortcomings by developing dry powder inhaler (DPI) formulations that are respirable and dispersible and thus do not require lactose blending. Dry powder formulations for inhalation therapy are described in U.S. Pat. No. 5,993,805 to Sutton et al.; U.S. Pat. No. 6,9216527 to Platz et al.; WO 0000176 to Robinson et al.; WO 9916419 to Tarara et al.;

WO 0000215 to Bot et al; U.S. Pat. No. 5,855,913 to Hanes et al.; and U.S. Pat. Nos. 6,136,295 and 5,874,064 to Edwards et al.

Broad clinical application of dry powder inhalation delivery has been limited by difficulties in generating dry powders of appropriate particle size, particle density, and dispersibility, keeping the dry powder stored in a dry state and developing a convenient, hand-held device that effectively disperses the respirable dry particles to be inhaled in air. In addition, the particle size of dry powders for inhalation delivery is inherently limited by the fact that smaller respirable dry particles are harder to disperse in air. Dry powder formulations, while offering advantages over cumbersome liquid dosage forms and propellant-driven formulations, 10 are prone to aggregation and low flowability, which considerably diminish dispersibility and the efficiency of dry powder-based inhalation therapies. For example, interparticular Van der Waals interactions and capillary condensation effects are known to contribute to aggregation of dry particles. Hickey, A. *et al.*, Factors Influencing the Dispersion of Dry Powders as Aerosols, Pharmaceutical Technology, August, 1994.

To overcome such interparticle adhesive forces, Batycky *et al.* in U.S. Patent No. 7,182,961 teach production of so called "aerodynamically light respirable particles," which have a volume median geometric diameter (VMGD) of greater than 5 microns (μm) as measured using a laser diffraction instrument such as HELOS (manufactured by Sympatec, Princeton, N.J.). See Batycky *et al.*, column 7, lines 42-65. Another approach to improve dispersibility of 20 respirable particles of average particle size of less than 10 μm , involves the addition of a water soluble polypeptide or addition of suitable excipients (including amino acid excipients such as leucine) in an amount of 50% to 99.9% by weight of the total composition. Eljamal *et al.*, U.S. Patent No. 6,582,729. However, this approach reduces the amount of active agent that can be delivered using a fixed amount of powder. Therefore, an increased amount of dry powder is required to achieve the intended therapeutic results, for example, multiple inhalations and/or frequent administration may be required. Still other approaches involve the use of devices that apply mechanical forces, such as pressure from compressed gasses, to the small particles to disrupt interparticular adhesion during or just prior to administration. See, *e.g.*, U.S. Pat. Nos. 7,601,336 to Lewis *et al.*, 6,737,044 to Dickinson *et al.*, 6,546,928 to Ashurst *et al.*, or U.S. Pat. 30 Applications 20090208582 to Johnston *et al.*

Despite the advances in dry powder formulation, clinically there has been no effective dry powder formulation of acetylsalicylic acid (aspirin). Given the known effectiveness of acetylsalicylic acid for treating cardiovascular disease and more generally thromboembolic (ischemic) diseases such as stroke, there remains a need to develop dry powder formulations of acetylsalicylic acid, acetylsalicylic acid which can effectively and rapidly deliver doses to the patient in distress. Acetylsalicylic acid is an antiplatelet drug, which inhibits cyclooxygenase activity of platelet prostaglandin H synthase-1 and almost completely suppresses platelet capacity to generate the prothrombotic and proatherogenic thromboxane A2. De Caterina.

Clinical use of acetylsalicylic acid in ischemic heart disease: past, present and future. Curr. Phar,

10 Des. 18(33):5215-23 (2012). It has clearly been shown that antiplatelet therapy with acetylsalicylic acid reduces the risk of serious vascular events. Id. Acetylsalicylic acid has also been shown to be useful in the treatment of acute ischemic stroke as well as transient ischemic events. ZhengMing et al. Indications for Early Acetylsalicylic acid Use in Acute Ischemic Stroke. Stroke 31:1240-1249 (2000); Warlow Controversies in Stroke: Acetylsalicylic acid Should Be First-Line Antiplatelet Therapy in the Secondary Prevention of Stroke. Stroke 33:2137-2138 (2000). However, given the rapid first-pass metabolism of acetylsalicylic acid, there remains a need for providing novel formulations of non-steroidal anti-inflammatory drugs ("NSAIDs"), such as acetylsalicylic acid, that are suitable for pulmonary delivery and may therefore bypass first-pass metabolism and avoid gastrointestinal side effects.

SUMMARY

The compositions of the present invention comprise dry powders that comprise an NSAID, such as acetylsalicylic acid or a pharmaceutically acceptable salt thereof, as the active ingredient. The dry powders comprise dry particles. The dry particles may be respirable. The dry particles may vary in size, *e.g.*, a geometric diameter (VMGD) between 0.5 μm and 30 μm . Alternatively, the dry powders can have a mass median aerodynamic diameter (MMAD) of about 20 μm or less. Optionally, the MMAD of the particles may be between 0.5 and 10 μm or between 1 and 10 μm .

In one embodiment, the particles may have a size distribution where 90% of the 10 formulation comprises particles with an MMAD of about 6 μm or less, 50% of the formulation comprises particles having an MMAD of about 3 μm or less, and 10% of the formulation comprises particles having an MMAD of about 1 μm or less.

The dry powders may comprise a mixture of particles having different sizes, *e.g.*, 20-30 μm (larger) and particles having a size of 5 μm or less (smaller). When administered, the smaller particles could reach the lower respiratory tract, while the larger particles would be captured in the upper respiratory tract.

The dry powder compositions can include one or more pharmaceutically acceptable 20 excipients, such as leucine, sodium citrate, maltodextrin, mannitol, sodium lauryl sulfate (SLS), lactose, starch, and/or cellulose. The one or more excipient may be present in an amount of about 5% to about 90% or by weight. The inclusion of an excipient is optional.

In one embodiment, the NSAID, such as acetylsalicylic acid or a pharmaceutically acceptable salt thereof, is provided in a dry powder formulation comprising a mixture of particles of various sizes, for example, a mixture of (i) particles having a mean geometric diameter (VMGD) and/or mass median aerodynamic diameter (MMAD) of about 5 μm or less, and (ii) particles having a mean geometric diameter (VMGD) and/or mass median aerodynamic diameter (MMAD) of 15 μm or greater. In one embodiment, the composition may include a pharmaceutically acceptable excipient. In another embodiment, the composition is free or substantially free of excipient. In certain embodiments, the composition is free or substantially free of anti-aggregation excipient.

30 In another embodiment, the NSAID, such as acetylsalicylic acid or a pharmaceutically acceptable salt thereof, comprises dry particles having a mass median aerodynamic diameter

(MMAD) within a range of about 0.5 μm to about 10 μm , wherein the dry powder further comprises one or more phospholipids in an amount ranging from about 0.1% (w/w) to about 10% (w/w) of the dry particles. The particles may have an MMAD size distribution where the particles exhibit: (i) a DV90 less than about 20 μm , a DV50 less than about 7 μm , and a DV10 less than about 2 μm ; (ii) a DV90 less than about 10 μm , a DV50 less than about 4 μm , and a DV10 less than about 1 μm ; (iii) a DV90 less than about 6 μm , a DV50 less than about 3 μm , and a DV10 less than about 1 μm ; (iv) a DV50 less than about 5 μm , and a DV10 less than about 2 μm ; or (v) a DV50 ranging from about 2.5 μm to about 4 μm , and a DV10 ranging from about 0.8 μm to about 1.5 μm .

10 The present composition may comprise acetylsalicylic acid or a pharmaceutically acceptable salt thereof, in an amount greater than 20% (w/w), greater than 30% (w/w), greater than 40% (w/w), greater than 50% (w/w), greater than 60% (w/w), greater than 70% (w/w), greater than 80% (w/w), or greater than 90% (w/w), of the composition.

The NSAID, such as acetylsalicylic acid, or a pharmaceutically acceptable salt thereof, of the present composition may be crystalline or amorphous.

20 The subject technology also relates to a dry powder or dry particle for use in therapy (e.g., treatment, prophylaxis, or diagnosis). The dry particle or dry powder may be used for treatment (including prophylactic treatment, such as prevention or reducing the risk) of a cardiovascular disease (such as caused by thrombosis) as described herein, and in the manufacture of a medicament for the treatment, prophylaxis or diagnosis of a cardiovascular or thromboembolic disease (such as thrombosis). The thromboembolic event may be treated within about 5, 10 or 15 minutes of onset of an ischemic event.

30 The subject technology also provides a drug delivery system for treating (including prophylactic treatment or reducing the risk of) a cardiovascular disease (such as thrombosis). The system may comprise: a therapeutically effective dose of an NSAID (such as acetylsalicylic acid or a pharmaceutically acceptable salt thereof) in dry powder form; a dry powder inhaler, the dry powder inhaler comprising a mouthpiece, a reservoir for receiving the dose of the NSAID (such as acetylsalicylic acid or a pharmaceutically acceptable salt thereof), and an actuation member for making available the dose of the acetylsalicylic acid for inhalation by the patient through the mouthpiece. In one embodiment, a single inhaled dose of the NSAID (such as acetylsalicylic acid or a pharmaceutically acceptable salt thereof) is about 40 mg or less. In

another embodiment, a single inhaled dose of the NSAID (such as acetylsalicylic acid or a pharmaceutically acceptable salt thereof) is about 30 mg or less. In particular, it is expected that a dosing regimen using inhalable acetylsalicylic acid will be able to deliver pharmaceutically equivalent levels of acetylsalicylic acid to the patient more quickly than is possible by oral dosing.

The dry powder may also comprise particles coated with a pharmaceutically acceptable excipient. The pharmaceutically acceptable excipient can be a phospholipid having surfactant properties, such as dipalmitoyl phosphatidylcholine (DPPC), distearoyl phosphatidylcholine (DSPC) or soy lecithin, in an amount ranging from about 0.1% to about 10% w/w or in an amount ranging from about 1% to about 5% w/w. In one embodiment, the weight percentage of the DSPC of the dry particles is 5% w/w. In another embodiment, the weight percentage of soy lecithin of the dry particles is 0.1% w/w.

The dose of acetylsalicylic acid (e.g., a single inhaled dose) may be present at amounts ranging from about 5 to about 40 mg. The formulation may further comprise clopidogrel. The dry powders can have an emitted dose ranging from about 75% to about 95%.

In one embodiment, the pharmaceutically acceptable excipient is DSPC, the dry powders substantially comprise dry particles having a MMAD ranging from about 3 to about 4 μm and the emitted dose is greater than about 90%. In this embodiment, the mass percent of stages in an NGI testing apparatus of the dry powder yields are at, stage 1 about 10% to about 13%, stage 2, about 20% to about 23%, stage 3, about 13% to about 15%, and stage 4, about 5% to about 6% and fine particle fraction ranges from about 45% to about 55%.

In another embodiment, the pharmaceutically acceptable excipient is soy lecithin, the dry powders substantially comprise dry particles having a MMAD ranging from about 2.0 to about 3.0 μm and the emitted dose ranges from about 70% to about 85%. In this embodiment, the mass percent of stages in an NGI testing apparatus of the dry powder yields are at, stage 1 about 5% to about 10%, stage 2, about 10% to about 18%, stage 3, about 15% to about 20%, and stage 4, about 10% to about 15% and fine particle fraction ranges from about 50% to about 70%.

BRIEF DESCRIPTION OF THE DRAWINGS

The accompanying drawings, which are included to provide further understanding of the subject technology and are incorporated in and constitute a part of this specification, illustrate aspects of the subject technology and together with the description serve to explain the principles of the subject technology.

Figure 1 is a schematic view of a patient using a dry powder inhaler, in accordance with some implementations of the methods and systems disclosed herein.

Figure 2 illustrates a generic Dry Powder Inhaler Device.

Figure 3 shows laser diffraction data of Formulation 3727.

10 Figure 4 shows laser diffraction data of Formulation 3734.

Figure 5 shows DSC thermograms of raw, micronized uncoated and spray-dried DSPC/acetylsalicylic acid particles.

Figure 6 shows TGA of micronized uncoated and spray-dried DSPC/acetylsalicylic acid particles.

Figures 7A and 7B show the particle size distribution of spray-dried DSPC/acetylsalicylic acid particles based on NGI analysis. Figure 7A: 2 capsules were delivered to the NGI; Figure 7B: 1 capsule was delivered to the NGI.

20 Figures 8A and 8B show the particle size distribution of spray-dried soy lecithin/acetylsalicylic acid particles based on NGI analysis. Figure 8A: 2 capsules were delivered to the NGI; Figure 8B: 1 capsule was delivered to the NGI.

DETAILED DESCRIPTION

1. INTRODUCTION

Thromboembolic Symptoms and Events

A thromboembolic event, such as myocardial infarction, deep venous thrombosis, pulmonary embolism, thrombotic stroke, etc., can present with a group of symptoms that allow a patient or clinician to provide an initial therapy or treatment for the event, i.e., immediately, or within 1, 5, 10 or 15 minutes of onset of the thromboembolic event. In certain situations, an 81 mg, low dose, or "baby" acetylsalicylic acid or a regular acetylsalicylic acid (330 mg) may be orally administered in order to provide an initial treatment for the patient. However, oral administration may not act as quickly as necessary to provide a sufficient therapeutic effect and therefore, may lead to a less preferred outcome. Thus, the pulmonary drug delivery system and related methods of the present invention provide for an accelerated and more efficient pathway and treatment for reducing the risk of a thromboembolic event and/or providing treatment for a thromboembolic event. For example, certain embodiments provide systems and methods of administering a non-steroidal anti-inflammatory drug ("NSAID") by inhalation, such as by a dry powder inhaler ("DPI") or a metered dose inhaler ("MDI").

Delivery Mechanisms for Drugs

Drugs can be administered orally in different ways, such as by liquids, capsules, tablets, or chewable tablets. The oral route is used most often because it is convenient, safe, and inexpensive. However, oral drug delivery has limitations because of the way a drug typically moves through the digestive tract. For example, when a drug is administered orally, it is absorbed in the mouth, stomach, and the small intestine. Before the drug enters the bloodstream, it must pass through the intestinal wall and travels to the liver. While passing through the intestinal wall and liver, the drug is metabolized, which can decrease the amount of the drug that actually reaches the bloodstream. The metabolism of the drug reduces the bioavailability of the drug and is often termed the "first pass effect." The fraction of the drug lost due to the first pass effect is generally determined by absorption in the liver and gut wall, and gastrointestinal lumen enzymes, gut wall enzymes, bacterial enzymes, and hepatic (liver) enzymes.

Generally, the first pass effect on acetylsalicylic acid significantly reduces the bioavailability of the administered dosage. Due to the acidic conditions in the stomach, acetylsalicylic acid is absorbed in the stomach and the upper small intestine. After being absorbed, acetylsalicylic acid is metabolized to acetic acid and salicylate. When taken orally, generally only about one to two-thirds of the dose of acetylsalicylic acid is bioavailable due to the first pass effect.

Drugs that are administered by inhalation may also undergo a first pass effect. For drug administration by inhalation, smaller particles proceed via a nasal route, down the windpipe (trachea) and into the lungs. The size of the particles can be determinative of the overall efficacy 10 of the treatment, i.e., whether they are subject to a first pass effect. Once inside the lungs, these particles are absorbed into the bloodstream. However, in the fraction of the drug that reaches the alveolar spaces of the lung, the active pharmaceutical ingredient (e.g., acetylsalicylic acid) will be absorbed within the capillaries and delivered to the pulmonary circulation. This material will initially circulate via the pulmonary vein back to the heart with oxygenated blood, and will then be distributed systemically via output from the left ventricle. As such, upon inhalation of the pharmaceutical, a substantial portion of the acetylsalicylic acid will avoid the first pass effect due to processing in the liver, with the result that levels of acetylsalicylic acid in the region of the heart will be higher than would be possible following prior art methods of oral administration.

Few drugs are administered by inhalation because the dosage of an inhaled drug, as well 20 as the delivery timing, can often be difficult to measure. Usually, this method is used to administer drugs that act specifically on the lungs, such as aerosolized anti-asthmatic drugs in metered-dose containers, and to administer gases used for general anesthesia. In this case, the inventors have determined that it is possible to reproducibly deliver doses of acetylsalicylic acid rapidly via a dry powder device.

It has also been found that coating of the drug particles with a surfactant, such as dipalmitoyl phosphatidylcholine (DPPC), distearoyl phosphatidylcholine (DSPC) or soy lecithin reproducibly improves delivery of the drug from the dry powder inhaler device.

Pharmacokinetics of Acetylsalicylic acid

30 Acetylsalicylic acid is the acetylated form of salicylic acid. Acetylsalicylic acid is used by millions of people to achieve desirable effects, and by many people, baby acetylsalicylic acid

is often used daily. The principal effect of acetylsalicylic acid is to impair the function of cyclooxygenase enzymes (specifically, COX1 and COX2 enzymes). By inhibiting COX1, acetylsalicylic acid can irreversibly inhibit platelet aggregation, which decreases the risk of blood clots. Additionally, the impairment of the COX2 enzyme can reduce inflammation, stiffness, and pain in the body by inhibiting prostaglandins and thromboxanes. As such, individuals at high risk for heart attack, stroke, or with inflammation often take acetylsalicylic acid to address symptoms and effects of these conditions. As noted, acetylsalicylic acid can effectively reduce the likelihood of such myocardial events and reduce pain and inflammation with a dose as small as a baby acetylsalicylic acid. However, due at least in part to its inhibition of COX1, acetylsalicylic acid can increase the risk of bleeding and cause damage to organs such as the stomach and intestines, which can be painful.

Oral dosing with acetylsalicylic acid typically follows standard Michaelis-Menton kinetics. Following administration of an oral dose, peak plasma levels of salicylic acid, the primary metabolite of acetylsalicylic acid, are typically achieved after about 1-2 hours, and acetylsalicylic acid is generally undetectable within 1-2 hours after administration. The rate of absorption from the GI tract is dependent on a number of factors including the dosage form, presence or absence of food, gastric pH, as well as other factors, such as age and weight.

Dry Powder Inhaler Technology

The oral delivery of acetylsalicylic acid may create a risk of damage to the stomach wall leading to pain, indigestion and a high risk of bleeding. Furthermore, it is often difficult to orally administer a drug during emergency situations that may result in a thromboembolic event. For example, the patient may be experiencing vomiting or otherwise be unable to take the drug orally, e.g., suffered from a stroke. Additionally, oral administration of a drug may be undesirable because the drug does not reach the systemic blood stream immediately, thus delaying the important effects of the drug. Even so, due to the first pass effect in the liver and gut, the amount of drug reaching systemic circulation is much less than that administered. The alternative route of administration described herein avoids these unwanted side effect.

Delivery of a drug by inhalation in the early stages of an emergency situation can also provide a fast-acting, effective form of preliminary treatment of certain medical conditions. For example, in one embodiment, upon receiving a complaint of a symptom of a serious

thromboembolic event, a patient can be administered, by DPI, a therapeutic amount of a NSAID. The NSAID can address problems associated with or provide an initial treatment for the medical condition.

However, dry powder inhalation of drugs has generally been limited to dosages of less than a milligram. Recent developments in particle engineering, in particular the development of PulmoSphere® technology, have enabled the delivery of a larger amount of dry powder to the lungs in a single actuation. See David E. Geller, M.D., et al., Development of an inhaled dry-powder formulation of tobramycin using pulmosphere™ technology, J Aerosol Med Pulm Drug Deliv. 2011 August; 24(4), pp. 175-82. In this publication, a dose of 112 mg tobramycin (in four capsules) was effectively delivered via PulmoSpheres™®.

The body includes various particle filters that limit the efficacy of inhaled drugs. For example, the oropharynx tends to prevent passage of particles having a diameter greater than 5 μm . However, in order to reach the alveoli, particles must have a size from about 1 μm to about 5 μm . Accordingly, the preparation and use of inhalable acetylsalicylic acid with particles having a median geometric diameter of from about 1 μm to about 5 μm , and in certain embodiments, from about 1.7 μm to about 2.7 μm is disclosed. Generally, particles sizes between about 1 μm and about 3 μm effectively deposit in the alveolar spaces following inhalation. The portion of the drug formulation falling within this size range is typically referred to as the fine particle fraction (FPF) and higher FPFs are most desirable when producing an inhalable drug formulation. In some cases, the FPF in the present invention can range from about 20% to about 90%, or even higher, depending on a number of factors including the method used to micronize the acetylsalicylic acid as well as optional excipients that may be included in the formulation that modulate aerodynamic performance.

Similar results are observed for other drugs, such as pharmaceutically active proteins, for example insulin. In the case of insulin, delivery by inhalation not only provides significantly higher peak plasma levels as compared to delivery by injection (nearly double), but a substantially more rapid appearance of the molecule in the circulation (less than 30 minutes when inhaled versus about 90 minutes when injected) (Technosphere® Technology: A Platform for Inhaled Protein Therapeutics, in Pulmonary Delivery: Innovative Technologies Breathing New Life into Inhalable Therapeutics, available online at <http://www.ondrugdelivery.com>, pp. 8-11).

To date, there has been no single dose use of acetylsalicylic acid by dry powder inhaler to replace the traditional daily use of a NSAID (such as a baby acetylsalicylic acid) or emergency use of a NSAID as preventative care for symptoms of a thromboembolic event. Accordingly, in one embodiment, the methods provide for administering a NSAID by dry powder inhalation in an amount less than the dosage of a baby acetylsalicylic acid (e.g., less than 81 mg).

Therefore, a method for treating disease, e.g., by reducing the risk of a thromboembolic event, comprises administering a NSAID, such as a salicylate, by a DPI or MDI. For example, the method can comprise administering acetylsalicylic acid by a DPI or MDI. The administered dose (e.g., a single dose) can be less than 25 mg of acetylsalicylic acid. Further, in various 10 embodiments, the administered dosage (e.g., a single dose) can be less than 20 mg of acetylsalicylic acid. The administered dosage (e.g., a single dose) can be less than 15 mg of acetylsalicylic acid, less than 12 mg of acetylsalicylic acid, less than 10 mg of acetylsalicylic acid, less than 8 mg of acetylsalicylic acid, less than 5 mg of acetylsalicylic acid or less than 2 mg of acetylsalicylic acid.

In other embodiments, the dosage of acetylsalicylic acid (e.g., a single dose) can be from about 2 mg to about 30 mg, about 4 mg to about 25 mg of acetylsalicylic acid, about 6 mg to about 20 mg of acetylsalicylic acid, about 8 mg to about 15 mg of acetylsalicylic acid, about 10 mg to about 13 mg of acetylsalicylic acid, about 1 mg, about 2 mg, about 3 mg, about 4 mg, about 5 mg, about 6 mg, about 7 mg, about 8 mg, about 9 mg, about 10 mg, about 11 mg, about 20 12 mg, about 13 mg, about 14 mg, about 15 mg, about 16 mg, about 17 mg, about 18 mg, about 19 mg, or about 20 mg of acetylsalicylic acid. Such dosages can provide a bioequivalent dosage when compared to typical dosages of about 81 mg to about 325 mg, while demonstrating few negative side effects.

Thus, a NSAID, such as acetylsalicylic acid, can be administered by DPI or MDI in a single dose or multiple dose that delivers much less than a traditional oral dose of acetylsalicylic acid, which can provide an equivalent treatment while tending to avoid the negative side effects associated with certain NSAIDs, such as acetylsalicylic acid. Further, systems (devices) of administering such treatments are also provided.

The NSAID, in particular acetylsalicylic acid, can be formulated to include 30 pharmaceutically acceptable excipients that are effective to improve aerodynamic performance, bioavailability and/or pharmacokinetics as compared to prior art methods of administration.

The DPI or MDI apparatus can have a mouthpiece and an actuation member for making available the NSAID for inhalation by a patient to reduce the risk of the thromboembolic event.

For example, a method of reducing the risk of a thromboembolic event is provided and can comprise administering a dose of a non-steroidal anti-inflammatory drug by a dry powder inhaler. The dose can be effective to reduce a risk of a thromboembolic event in a patient. The dry powder inhaler can have a mouthpiece and an actuation member for making available the dose of the non-steroidal anti-inflammatory drug for inhalation by the patient to reduce the risk of the thromboembolic event.

A drug delivery system can also be provided for treating a disease, for example, by 10 reducing the risk of a thromboembolic (ischemic) event. The system can comprise a dose of a non-steroidal anti-inflammatory drug in powder form. The dose can be effective to reduce the risk of a thromboembolic event in a patient. The system can also comprise a dry powder inhaler. The dry powder inhaler can have a mouthpiece, a reservoir for receiving the dose of the non-steroidal anti-inflammatory drug, and an actuation member for making available the dose of the non-steroidal anti-inflammatory drug for inhalation by the patient through the mouthpiece.

The thromboembolic event may be a myocardial infarction, deep venous thrombosis, pulmonary embolism, or thrombotic stroke. The dose of the NSAID drug can be administered as a preliminary treatment in response to a symptom of a thromboembolic event. The NSAID may be acetylsalicylic acid and may be administered in a single dose or in multiple doses, e.g., 2, 3, 4, 20 5, 6, 7, 8, 9 or 10 or greater doses.

2. DEFINITIONS

The term "dry powder" refers to a composition contains finely dispersed respirable dry particles that are capable of being dispersed in an inhalation device and subsequently inhaled by a subject. Such dry powder or dry particle may contain up to about 15% water or other solvent, or be substantially free of water or other solvent, or be anhydrous.

The term "dry particles" refers to respirable particles that may contain up to about 15% water or other solvent, or be substantially free of water or other solvent, or be anhydrous.

The term "respirable" refers to dry particles or dry powders that are suitable for delivery 30 to the respiratory tract (e.g., pulmonary delivery) in a subject by inhalation. Respirable dry

powders or dry particles may have a mass median aerodynamic diameter (MMAD) of less than about 20 μm , less than about 15 μm , less than about 10 μm , less than about 5 μm , or less.

The term "dispersible" describes the characteristic of a dry powder or dry particles to be dispelled into a respirable aerosol. Dispersibility of a dry powder or dry particles is expressed herein as the quotient of the volume median geometric diameter (VMGD) measured at a dispersion (*i.e.*, regulator) pressure of 1 bar divided by the VMGD measured at a dispersion (*i.e.*, regulator) pressure of 4 bar, or VMGD at 0.5 bar divided by the VMGD at 4 bar as measured by HELOS/RODOS. These quotients are referred to herein as "1/4 bar," and "0.5/4 bar," respectively, and dispersibility correlates with a low quotient. For example, 1/4 bar refers to the 10 VMGD of respirable dry particles or powders emitted from the orifice of a RODOS dry powder disperser (or equivalent technique) at about 1 bar, as measured by a HELOS or other laser diffraction system, divided the VMGD of the same respirable dry particles or powders measured at 4 bar by HELOS/RODOS. Thus, a highly dispersible dry powder or dry particles will have a 1/4 bar or 0.5/4 bar ratio that is close to 1.0. Highly dispersible powders have a low tendency to agglomerate, aggregate or clump together and/or, if agglomerated, aggregated or clumped together, are easily dispersed or de-agglomerated as they emit from an inhaler and are breathed in by the subject. Dispersibility can also be assessed by measuring the size emitted from an inhaler as a function of flow rate.

The term "emitted dose" or "ED" refers to an indication of the delivery of a drug 20 formulation from a suitable inhaler device after a firing or dispersion event. More specifically, for dry powder formulations, the ED is a measure of the percentage of powder that is drawn out of a unit dose package and that exits the mouthpiece of an inhaler device. The ED is defined as the ratio of the dose delivered by an inhaler device to the nominal dose (*i.e.*, the mass of powder per unit dose placed into a suitable inhaler device prior to firing). The ED is an experimentally-measured parameter, and can be determined using the method of USP Section 601 Aerosols, Metered-Dose Inhalers and Dry Powder Inhalers, Delivered-Dose Uniformity, Sampling the Delivered Dose from Dry Powder Inhalers, United States Pharmacopeia Convention, Rockville, MD, 13th Revision, 222-225, 2007. This method utilizes an *in vitro* device set-up to mimic patient dosing.

30 The terms "FPF (<5.6)," "FPF (<5.6 μm)," and "fine particle fraction of less than 5.6 μm " refer to the fraction of a sample of dry particles that have an aerodynamic diameter of less than

5.6 μm . For example, FPF (<5.6) can be determined by dividing the mass of respirable dry particles deposited on the stage one and on the collection filter of a two-stage collapsed Andersen Cascade Impactor (ACI) by the mass of respirable dry particles weighed into a capsule for delivery to the instrument. This parameter may also be identified as "FPF_TD (<5.6)," where TD means total dose. A similar measurement can be conducted using an eight-stage ACI. The eight-stage ACI cutoffs are different at the standard 60 L/min flow rate, but the FPF_TD (<5.6) can be extrapolated from the eight-stage complete data set. The eight-stage ACI result can also be calculated by the USP method of using the dose collected in the ACI instead of what was in the capsule to determine FPF.

10 The terms "FPF (<3.4)," "FPF (<3.4 μm)," and "fine particle fraction of less than 3.4 μm " refers to the fraction of a mass of respirable dry particles that have an aerodynamic diameter of less than 3.4 μm . For example, FPF (<3.4) can be determined by dividing the mass of respirable dry particles deposited on the collection filter of a two-stage collapsed ACI by the total mass of respirable dry particles weighed into a capsule for delivery to the instrument. This parameter may also be identified as "FPF_TD (<3.4)," where TD means total dose. A similar measurement can be conducted using an eight-stage ACI. The eight-stage ACI result can also be calculated by the USP method of using the dose collected in the ACI instead of what was in the capsule to determine FPF.

20 The terms "FPF (<5.0)," "FPF (<5.0 μm)," and "fine particle fraction of less than 5.0 μm " as used herein, refer to the fraction of a mass of respirable dry particles that have an aerodynamic diameter of less than 5.0 μm . For example, FPF (<5.0) can be determined by using an eight-stage ACI at the standard 60 L/min flow rate by extrapolating from the eight-stage complete data set. This parameter may also be identified as "FPF_TD (<5.0)," where TD means total dose.

3. NON-STEROIDAL ANTI-INFLAMMATORY DRUGS (NSAIDs)

NSAIDs, such as acetylsalicylic acid, can provide various beneficial effects and contribute to reducing the risk of a cardiovascular disease (such as thrombosis). However, the use of NSAIDs, such as acetylsalicylic acid, in a clinical setting has traditionally been limited to oral administration. Oral administration of acetylsalicylic acid, for example, can result in the loss 30 or inactivation of approximately 2/3 of the oral dosage due to the first pass effect in the gut and liver. While one third of the dosage reaches the systemic blood stream and provides the desired

effect, the negative side effects created by the full dosage often deter patients from using acetylsalicylic acid on a regular or daily basis.

Further, in many situations, such as in emergencies, oral administration of NSAIDs, such as acetylsalicylic acid, may be inappropriate because it may take too long to be effective.

According to at least one aspect of some embodiments disclosed herein is the realization that an alternative administration method and systems can be implemented that utilize a lower dosage and provide a more direct delivery mechanism to the systemic blood stream. Thus, the methods and systems of the present invention allow for the beneficial effects of NSAIDs, such as acetylsalicylic acid, to be achieved on a regular basis and in emergency situations, while minimizing previous drawbacks associated with the use of NSAIDs.

Various studies have determined that acetylsalicylic acid has a significant effect on reducing the risk of myocardial infarction. These studies have used acetylsalicylic acid dosages of 325 mg. However, these studies have based their findings on oral administration of acetylsalicylic acid and have not suggested DPI or MDI administration.

Although inhaled dry powder formulations of acetylsalicylic acid have been developed, reports have stated that the formulation was not clinically feasible because it is difficult to meet the high dosage requirements of acetylsalicylic acid (~80 mg/day for low-dose prevention of coronary events and stroke, and at least 300 mg/day for pain or fever relief) via pulmonary delivery of dry powders.

In addition, these reports recognize that adverse effects of dry powder on the lungs, such as coughing, cannot be avoided unless the doses are less than a few tenths of a milligram in a single breath. Thus, prior teachings suggest that higher dosage requirements of acetylsalicylic acid would be impossible or difficult to meet using DPI (or MDI). Finally, there may be a higher incidence of acetylsalicylic acid intolerance in asthmatic patients when acetylsalicylic acid is delivered by inhalation than orally.

The use of nanoparticulate drugs for dry powder inhaler (DPI) delivery is not straightforward. Direct inhalation of nanoparticulate drugs was infeasible due to their small size. The nanometer size leads to the nanoparticulate drugs being predominantly exhaled from the lungs, without any deposition in the lungs taking place. Moreover, a severe aggregation problem arising from the small size makes their physical handling difficult for DPI delivery. Accordingly, "large hollow carrier particles" of nanoparticulate drugs has been developed for pulmonary

delivery of some drugs. See Hadinoto et al., *Drug Release Study Of Large Hollow Nanoparticulate Aggregates Carrier Particles For Pulmonary Delivery*, International Journal of Pharmaceutics 341 (2007) 195-20.

The methods and systems of the present invention provide for treating (including prophylactic treatment or reducing the risk of) a disease, for example, treating a cardiovascular disease (such as thrombosis) by administration of a low amount of a NSAID, such as a low dose of acetylsalicylic acid, by DPI. The dose can be much less than that of a baby acetylsalicylic acid (e.g., less than 81 mg). The administered dosage can be less than about 40 mg of acetylsalicylic acid. The administered dosage can be less than 25 mg of acetylsalicylic acid. Further, the 10 administered dosage can be less than 20 mg of acetylsalicylic acid. The administered dosage can be less than 15 mg of acetylsalicylic acid. The administered dosage can also be less than 12 mg of acetylsalicylic acid. The administered dosage can be less than 10 mg of acetylsalicylic acid. Furthermore, the administered dosage can be less than 8 mg of acetylsalicylic acid. The administered dosage can be less than 5 mg of acetylsalicylic acid. In some embodiments, the administered dosage can be less than 2 mg of acetylsalicylic acid.

For example, the dosage can be from about 1 mg to about 40 mg. In various embodiments, the dosage can be from about 4 mg to about 25 mg of acetylsalicylic acid, about 6 mg to about 20 mg of acetylsalicylic acid, about 8 mg to about 15 mg of acetylsalicylic acid, about 10 mg to about 13 mg of acetylsalicylic acid or about 1 mg, about 2 mg, about 3 mg, about 4 mg, about 5 mg, about 6 mg, about 7 mg, about 8 mg, about 9 mg, about 10 mg, about 11 mg, about 12 mg, about 13 mg, about 14 mg, about 15 mg, about 16 mg, about 17 mg, about 18 mg, about 19 mg, or about 20 mg of acetylsalicylic acid. Alternatively, the dose of acetylsalicylic acid can be less than about 80 mg, about 1 mg to about 75 mg, about 2 mg to about 60 mg, about 5 mg to about 40 mg, about 10 mg to about 30 mg, about 12 mg to about 25 mg, about 15 mg to about 20 mg, about 60 mg to about 95 mg, about 50 mg to about 100 mg, about 50 mg to about 80 mg, about 40 mg to about 80 mg, about 20 mg to about 30 mg, about 30 mg to about 40 mg, about 40 mg to about 50 mg, about 50 mg to about 60 mg, about 60 mg to about 70 mg, about 70 mg to about 80 mg, about 80 mg to about 90 mg, or about 90 mg to about 100 mg.

Such dosages can provide a bioequivalent dosage of acetylsalicylic acid when compared 30 to typical dosages of 81 mg to about 325 mg, while demonstrating few negative side effects.

In certain embodiments, NSAIDs can be used in various methods and systems. In some embodiments, NSAIDs can include salicylates, i.e., the salts and esters of salicylic acid, which have anti-platelet action. Further, NSAIDs can also include one or more of the following compounds listed in Table 1.

Table 1 Examples of NSAIDs

| | |
|----|---|
| | Aspirin is a brand name; the chemical is called acetylsalicylic acid |
| | Celecoxib (Celebrex) |
| 10 | Dexketoprofen (Keral) |
| | Diclofenac (Voltaren, Cataflam, Voltaren-XR) |
| | Diflunisal (Dolobid) |
| | Etodolac (Lodine, Lodine XL) |
| | Etoricoxib (Algix) |
| | Fenoprofen (Fenopron, Nalfron) |
| | Firocoxib (Equioxx, Previcox) |
| | Flurbiprofen (Urbifen, Ansaid, Flurwood, Proben) |
| | Ibuprofen (Advil, Brufen, Motrin, Nurofen, Medipren, Nuprin) |
| | Indomethacin (Indocin, Indocin SR, Indocin IV) |
| 20 | Ketoprofen (Actron, Orudis, Oruvail, Ketoflam) |
| | Ketorolac (Toradol, Sprix, Toradol IV /IM, Toradol IM) |
| | Licofelone (under development) |
| | Lomoxicam (Xefo) |
| | Loxoprofen (Loxonin, Loxomac, Oxeno) |
| | Lumiracoxib (Prexige) |
| | Meclofenamic acid (Meclofenam) |
| | Mefenamic acid (Ponstel) |
| | Meloxicam (Movalis, Melox, Recoxa, Mobic) |
| | Nabumetone (Relafen) |
| 30 | Naproxen (Aleve, Anaproxx, Midol Extended Relief, Naprosyn, Naprelan) |
| | Nimesulide (Sulide, Nimalox, Mesulid) |

Oxaprozin (Daypro, Dayrun, Duraprox)
Parecoxib (Dynastat)
Piroxicam (Feldene)
Rofecoxib (Vioxx, Ceoxxx, Ceeoxxx)
Salsalate (Mono-Gesic, Salflex, Disalcid, Salsitab)
Sulindac (Clinoril)
Tenoxicam (Mobi flex)
Tolfenamic acid (Clotam Rapid, Tufnil)
Valdecoxib (Bextra)

10

Other alternatives can also be used instead of a NSAID. Such alternatives include Plavix (clopidogrel), COX-2 inhibitors, other remedies such as Nattokinase (an enzyme (EC 3.4.21.62, extracted and purified from a Japanese food called nattō)). Further, other drugs that provide different beneficial effects, such as being effective to reduce a risk of a cardiovascular disease (such as thrombosis) in a patient, can also be used in some embodiments. Thus, the discussion of methods and systems shall apply generally to these various alternatives, although for discussion purposes, the present disclosure often refers to acetylsalicylic acid. It is contemplated that the methods, effects, pharmacokinetic data, and other considerations relating to acetylsalicylic acid 20 can be equally applied to other NSAIDs.

4. DRY POWDERS AND DRY PARTICLES

The dry particles of the subject technology are dispersible. The size of the dry particles can be expressed in a variety of ways that are conventional in the art, such as, fine particle fraction (FPF), volumetric median geometric diameter (VMGD), or mass median aerodynamic diameter (MMAD).

The dry particles of the subject technology may have a VMGD as measured by HELOS/RODOS at 1.0 bar of about 10 μm or less (e.g., about 0.1 μm to about 10 μm). Preferably, the dry particles of the subject technology have a VMGD of about 9 μm or less (e.g., 30 about 0.1 μm to about 9 μm), about 8 μm or less (e.g., about 0.1 μm to about 8 μm), about 7 μm or less (e.g., about 0.1 μm to about 7 μm), about 6 μm or less (e.g., about 0.1 μm to about 6 μm),

about 5 μm or less (e.g., less than 5 μm , about 0.1 μm to about 5 μm), about 4 μm or less (e.g., 0.1 μm to about 4 μm), about 3 μm or less (e.g., 0.1 μm to about 3 μm), about 2 μm or less (e.g., 0.1 μm to about 2 μm), about 1 μm or less (e.g., 0.1 μm to about 1 μm), about 0.5 μm to about 6 μm , about 0.5 μm to about 5 μm , about 0.5 μm to about 4 μm , about 0.5 μm to about 3 μm , or about 0.5 μm to about 2 μm as measured by HELOS/RODOS at 1.0 bar. In an exemplary embodiment, the dry particles of the subject technology have a VMGD as measured by HELOS/RODOS at 1.0 bar of about 1.3 to about 1.7 μm . In another exemplary embodiment, the dry particles of the subject technology have a VMGD as measured by HELOS/RODOS at 1.0 bar of about 0.5 μm to about 2 μm .

10 Alternatively, the dry particles may have a VMGD as measured by HELOS/RODOS at 1.0 bar of about 30 μm or less (e.g., about 5 μm to about 30 μm). Preferably, the dry particles of the subject technology have a VMGD of about 25 μm or less (e.g., about 5 μm to about 25 μm), about 20 μm or less (e.g., about 5 μm to about 20 μm), about 15 μm or less (e.g., about 5 μm to about 15 μm), about 12 μm or less (e.g., about 5 μm to about 12 μm), about 10 μm or less (e.g., about 5 μm to about 10 μm), or about 8 μm or less (e.g., 6 μm to about 8 μm) as measured by HELOS/RODOS at 1.0 bar. The dry powders can comprise a mixture of particles having different sizes.

20 The respirable dry particles can have an MMAD of about 10 μm or less, such as an MMAD of about 0.5 μm to about 10 μm . Preferably, the dry particles of the subject technology have an MMAD of about 5 μm or less (e.g. about 0.5 μm to about 5 μm , preferably about 1 μm to about 5 μm), about 4 μm or less (e.g., about 1 μm to about 4 μm), about 3.8 μm or less (e.g. about 1 μm to about 3.8 μm), about 3.5 μm or less (e.g. about 1 μm to about 3.5 μm), about 3.2 μm or less (e.g. about 1 μm to about 3.2 μm), about 3 μm or less (e.g. about 1 μm to about 3.0 μm), about 2.8 μm or less (e.g. about 1 μm to about 2.8 μm), about 2.2 μm or less (e.g. about 1 μm to about 2.2 μm), about 2.0 μm or less (e.g. about 1 μm to about 2.0 μm) or about 1.8 μm or less (e.g. about 1 micron to about 1.8 μm).

30 Alternatively, the dry powders and dry particles of the subject technology have a FPF of less than 5.0 μm ($\text{FPF_TD} < 5.0 \mu\text{m}$) of at least about 20%, at least about 30%, at least about 45%, at least about 40%, at least about 45%, at least about 50%, at least about 60%, at least about 65% or at least about 70%. Alternatively or in addition, the dry powders and dry particles of the subject technology have a FPF of less than 5.0 μm of the emitted dose ($\text{FPF_ED} < 5.0 \mu\text{m}$)

of at least about 45%, at least about 50%, at least about 60%, at least about 65%, at least about 70%, at least about 75%, at least about 80%, or at least about 85%.

In another embodiment, the dry powders and dry particles of the invention can have an FPF of less than about 5.6 μm ($\text{FPF} < 5.6 \mu\text{m}$) of at least about 20%, at least about 30%, at least about 40%, preferably at least about 45%, at least about 50%, at least about 55%, at least about 60%, at least about 65%, or at least about 70%.

In a third embodiment, embodiment, the dry powders and dry particles of the invention can have an FPF of less than about 3.4 μm ($\text{FPF} < 3.4 \mu\text{m}$) of at least about 20%, preferably at least about 25%, at least about 30%, at least about 35%, at least about 40%, at least about 45%,
10 at least about 50%, or at least about 55%.

The dry powders and dry particles may have a tap density of about 0.1 g/cm^3 to about 1.0 g/cm^3 . For example, the small and dispersible dry particles have a tap density of about 0.1 g/cm^3 to about 0.9 g/cm^3 , about 0.2 g/cm^3 to about 0.9 g/cm^3 , about 0.2 g/cm^3 to about 0.9 g/cm^3 , about 0.3 g/cm^3 to about 0.9 g/cm^3 , about 0.4 g/cm^3 to about 0.9 g/cm^3 , about 0.5 g/cm^3 to about 0.9 g/cm^3 , or about 0.5 g/cm^3 to about 0.8 g/cm^3 , greater than about 0.4 g/cc , greater than about 0.5 g/cc , greater than about 0.6 g/cc , greater than about 0.7 g/cc , about 0.1 g/cm^3 to about 0.8 g/cm^3 , about 0.1 g/cm^3 to about 0.7 g/cm^3 , about 0.1 g/cm^3 to about 0.6 g/cm^3 , about 0.1 g/cm^3 to about 0.5 g/cm^3 , about 0.1 g/cm^3 to about 0.4 g/cm^3 , about 0.1 g/cm^3 to about 0.3 g/cm^3 , less than 0.3 g/cm^3 . In a preferred embodiment, tap density is greater than about 0.4 g/cm^3 ; in another
20 preferred embodiment, tap density is greater than about 0.5 g/cm^3 . Alternatively, tap density may be less than about 0.4 g/cm^3 .

The dry powders and dry particles can have a water or solvent content of less than about 15% by weight of the dry particle. For example, the dry particles can have a water or solvent content of less than about 15% by weight, less than about 13% by weight, less than about 11.5% by weight, less than about 10% by weight, less than about 9% by weight, less than about 8% by weight, less than about 7% by weight, less than about 6% by weight, less than about 5% by weight, less than about 4% by weight, less than about 3% by weight, less than about 2% by weight, less than about 1% by weight or be anhydrous. In another embodiment, the dry particles of the subject technology can have a water or solvent content of less than about 6% and greater than about 1%, less than about 5.5% and greater than about 1.5%, less than about 5% and greater than about 2%, about 2%, about 2.5%, about 3%, about 3.5%, about 4%, about 4.5% about 5%.
30

Depending on the specific applications, the present composition (e.g., the present dry powders or dry particles) may contain a low or high percentage of active ingredient in the composition. For example, the dry particles may contain 3% or more, 5% or more, 10% or more, 15% or more, 20% or more, 25% or more, 30% or more, 35% or more, 40% or more, 50% or more, 60% or more, 70% or more, 75% or more, 80% or more, 85% or more, 90% or more, 95% or more, about 30% to about 99%, about 40% to about 99%, about 50% to about 99%, about 60% to about 99%, about 70% to about 99%, about 80% to about 99%, about 40% to about 95%, or about 50% to about 95% (weight percentage, w/w) of the active ingredient (e.g., acetylsalicylic acid).

10

5. DELIVERY OF DRY POWDERS

Although alveoli primarily function to exchange carbon dioxide for oxygen, alveoli also produce enzymes. Thus, inhaled substances, such as pathogens, drugs, or other chemicals, may be processed at the alveoli. Absorption of NSAIDs administered by DPI or MDI through the pulmonary capillaries and epithelium can provide an immediately effective treatment to address symptoms of thromboembolic events. Substantial first pass effect produced by oral administration of NSAIDs, such as acetylsalicylic acid, can be avoided through administration to the lungs by a dry powder inhaler. In addition, there has hitherto been no teaching or suggestion regarding the pharmacokinetics of dry powder delivery of a NSAID, such as acetylsalicylic acid, 20 and the possible metabolism or inactivation of the drug as it encounters the endothelial tissue of the pulmonary capillaries.

Referring to Figure 1, in a dry powder inhalation technique, a patient can use a dry powder inhaler 10 to inhale a powder formulation of a drug, such as a NSAID. The dose is effective to reduce a risk of a thromboembolic event in the patient. The lung is an efficient filter, and generally only permits entry of particles having a size of less than 5 μm . Here, after the drug enters the main stem bronchus 20, the drug will enter each lung 22, 24. The drug can then pass through the bronchial trees 26, 28 until reaching the individual alveoli 30 in the lungs 22, 24. Thus, the dry powder inhaler 10 can allow the patient to self administer a dosage of particles having a size of from about 1 μm and about 5 μm ; alternatively, the particle size can be from 30 about 2 μm to about 4 μm .

Various types of inhalers can be used to provide the drug using a DPI or MDI delivery system. The dose administered can be effective to reduce a risk of a thromboembolic event in a patient.

For example, the dry powder inhaler 10 can comprise a mouthpiece, a reservoir for receiving the NSAID, and an actuation member for making available the NSAID for inhalation by a patient through the mouthpiece.

For example, Figure 2 illustrates a generic Dry Powder Inhaler device. The methods and systems of the present invention may be adapted for use with any DPI or MDI device, including, but not limited to Aerolizer®, Diskus®, Ellipta®, Flexhaler®, Handihaler®, Neohaler®,

10 Pressair®, Rotahaler®, Turbuhaler® or Twisthaler® Plastiape, CDMHaler (see, <http://www.nationaljewish.org/healthinfo/medications/devices/dry-powder>).

In the process of breathing, the lungs are normally continuously exposed to materials present in the environment of a variety of sizes. This can include pollens (20-90 μm), bacteria (0.2- 200 μm), and smoke particulates (0.01 - 1 μm). Deposition of a particular particle depends on a number of factors, including the size and density of the particle, as well as the velocity of flow of air into and out of the lungs, and the resident time of the particle in the respiratory system. Moreover, the human body has developed systems to protect against adverse effects of some of these inhaled substances, including such processes as phagocytosis. Thus, one factor to consider when designing systems and methods for delivering a pharmaceutical compound via inhalation is 20 the effect that particle size has on the location in the respiratory tract where drug particles are likely to become deposited after inhalation.

Particles that enter the lungs are deposited along the course of the respiratory tract by impaction, sedimentation and diffusion. Often, the behavior of particles within an airflow stream can be described by aerodynamic diameter, as described in detail herein. Like the Reynold's number concept in aerodynamics, two particles having the same aerodynamic diameter will behave fundamentally the same in an airflow, regardless of their actual geometric (i.e., physical) size.

Previously it has been shown that particle size, or aerodynamic diameter, significantly affects the location within the respiratory system where particles are most likely to become 30 deposited after inspiration. For example, Heyder et al. (J. Aerosol. Sci. 17, 811-825, 1986) examined deposition of particles ranging in size from 5 nm to 15 μm in the respiratory tract.

Their studies indicated that particles with an aerodynamic diameter greater than 5 μm deposit predominantly by inertial impaction in the mouth and upper airways. Smaller particles, (aerodynamic diameter ranging from 1-5 μm) deposit deeper in the lungs by impaction and sedimentation, while very small particles (aerodynamic diameter < 1 μm), mainly remain suspended in the airflow and are exhaled.

Others have obtained similar results, suggesting that for delivery of drugs to the lungs, particles with a median aerodynamic diameter of about 2 μm are likely to be efficiently deposited in the alveolar spaces, with fractional deposition approaching 90% of the delivered particle dose (Byron, 1986, *J. Pharm. Sci.* 75(5), 433-438). In contrast, where particles have a median aerodynamic diameter ranging from 5-10 μm , only about 10% of the delivered dose will deposit in the alveoli, with about 40% depositing in the airways, and the remainder in the oral cavity and pharynx. Where median aerodynamic diameter is 15 μm or greater, particles deposit predominantly in the oral cavity and pharynx.

The methods and systems of the invention provide an apparatus and method for providing a therapeutically effective dose of an NSAID in order to reduce the risk of a thromboembolic event. As discussed above, the general approach is to deliver an NSAID in a pharmaceutically acceptable powdered form (e.g., Acetylsalicylic acid, and/or derivatives thereof) by means of an inhaler.

As discussed above, there is a general trend that deposition of particle in the alveolar spaces increases as particle size is reduced. Studies on nanoparticle distribution have shown that inhaled nanoparticles having a size < 100 nm are desirable for alveolar deposition as well as for minimizing lung phagocytosis (Hoet *et al.*, 2004, *J. Nanbiotechnol.* 2, 10.1186/1477-3155-2-12). Nanoparticles may provide additional advantages in terms of dispersion of the active compound and ultimately in the rate of uptake as compared to coarser preparations, the most obvious of which is that smaller particles tend to disperse and solubilize faster than larger ones.

With respect to the particle size distribution (PSD), the present composition may contain particles having same (or similar) size distribution, or particles having different size distributions. The particle sizes of the present composition may have a mono-modal, bimodal or multimodal distribution. As a result, the present composition may produce mono-modal, bimodal or multimodal absorption. In other words, administration of the present composition may result in a mono-modal, bimodal or multimodal concentration-time profile.

For example, the present composition may contain one, two or three groups of the following: particles with a median aerodynamic diameter in a range from about 1 μm to about 5 μm , particles with a median aerodynamic diameter in a range from about 5 μm to about 15 μm , and particles with a median aerodynamic diameter greater than about 15 μm ,

Mixing particles of the same active ingredient (e.g., acetylsalicylic acid), using batches of particles having different size distributions, can reduce bridging. For example, while a composition having a relatively uniform particle size will aggregate, providing a blended composition having some particles with a median aerodynamic diameter in a range from about 1 μm to about 5 μm , other particles with a median aerodynamic diameter in a range from about 5 μm to about 15 μm , and still other particles with a median aerodynamic diameter greater than about 15 μm , will inhibit aggregation and maintain the deposition characteristics of the preparation. In effect, the pharmaceutically active compound is used to replace the function of an excipient (such as lactose) with respect to preventing aggregation during storage of the medicament.

In addition, and unlike many other drugs, NSAIDs, and in particular acetylsalicylic acid, are able to enter the circulatory system effectively through routes other than through the epithelium of the alveoli. Notably, acetylsalicylic acid is able to enter the body by absorption through the mucosal layers of the oral cavity, as well as the pharynx and undoubtedly the epithelium of the airways. Thus, regardless of particle size, it will be appreciated that by providing an inhalable form of acetylsalicylic acid, the inhaled dosage can be substantially taken up into the systemic circulation, and be effective to reduce the risk of a thromboembolic event within a comparatively short period of time after the initial event.

In addition, by selecting the proportions of the various particle sizes, one can provide formulations that are faster or slower acting, based on the location of where the drug is ultimately deposited. For example, in some embodiments it may be desirable to provide a preparation that comprises 80% acetylsalicylic acid particles with a median aerodynamic diameter of about 1 μm to about 5 μm , and about 20% of particles with a median aerodynamic diameter of at least 15 μm . Other combinations are possible as well, and those of skill in the art will readily appreciate that faster acting preparations will comprise proportionately more smaller particles, while slower acting preparations will comprise proportionately more large particles. Thus, using the apparatus and methods described herein it is therefore possible to provide a

therapeutically effective dose of an NSAID such as acetylsalicylic acid via the respiratory tract, at least as rapidly as can be achieved by oral dosing.

Where a slower acting dosage form was desired, the formulation could include increasing fractions of particles with a median aerodynamic diameter in the range from about 5 μm to about 10 μm , or 15 μm or greater. These preparations would result in deposition in either the airways or oral cavity and pharynx and thus provide a more gradual increase in circulating levels of acetylsalicylic acid and its metabolic derivatives.

In either case, the subject technology provides formulations that can deliver acetylsalicylic acid and its pharmacologically active metabolic byproducts (e.g., salicylate) to the 10 systemic circulation at least as quickly if not more quickly than can be accomplished via oral administration. In addition, the present formulations are effective to deliver acetylsalicylic acid and its pharmacologically active metabolic byproducts to the systemic circulation at levels at least equal to that observed after oral administration of an equivalent dose of acetylsalicylic acid.

For example, pharmacokinetic studies show that after oral administration of acetylsalicylic acid peak plasma levels are achieved after about 20 minutes, after which they rapidly decline due to the relatively short elimination half-life (15-20 minutes). By comparison, plasma levels of the primary pharmacologically active metabolite salicylate, increase for a period of about 45 minutes following administration of acetylsalicylic acid, and remain elevated for much longer due to its significantly longer elimination half-life (2-3 hr) (Dressman et al., 2012, 20 Biowaiver Monograph for Immediate-Release Solid Oral Dosage Forms: Acetylsalicylic Acid, doi 10.1002/jps.2312).

Significantly, the pharmacokinetic behavior of acetylsalicylic acid has been found to be linear over a dosage range from 30-400 mg.

Accordingly, one aspect of the subject technology provides a dry powder that comprises a mixture of particles of various sizes.

For example, the dry powder can comprise particles of large sizes, as measured by VMGD (e.g., VMGD $\geq 15 \mu\text{m}$, such as $\geq 20 \mu\text{m}$ or 20-30 μm) and of small sizes, as measured by VMGD (e.g., VMGD $\leq 5 \mu\text{m}$, such as 1-3 μm) at a ratio (w:w) of: about 1:1, about 1:2, about 1:3, about 1:4, about 1:5, about 1:6, about 1:7, about 1:8, about 1:10, about 1:15, about 1:20, 30 about 1:25, about 1:30, about 1:40, about 1:50, about 1:100, about 2:1, about 3:1, about 4:1,

about 5:1, about 6:1, about 7:1, about 8:1, about 9:1, about 10:1, about 15:1, about 20:1, about 25:1, about 30:1, about 40:1, about 50:1, or about 100:1, etc.

Alternatively, the dry powder can comprise: about 1%, about 5%, about 10%, about 15%, about 20%, about 25%, about 30%, about 35%, about 40%, about 45%, about 50%, about 55%, about 50%, about 55%, about 60%, about 65%, about 70%, about 75%, about 80%, about 85%, about 90%, about 95%, or about 99% (weight percentage) of particles having VMGD of about 10 μm or less, preferably about 5 μm or less. Particles of 10 μm or less generally can reach lungs, and particles of 5 μm or less (e.g., 1-3 μm) generally can reach alveoli.

In another embodiment, the dry powder can comprise: about 1%, about 5%, about 10%,
10 about 15%, about 20%, about 25%, about 30%, about 35%, about 40%, about 45%, about 50%, about 55%, about 50%, about 55%, about 60%, about 65%, about 70%, about 75%, about 80%, about 85%, about 90%, about 95%, or about 99% (weight percentage) of particles having VMGD of between about 5 μm to about 20 μm , preferably between about 5 μm to about 15 μm , or between about 5 μm to about 10 μm .

Alternatively, the dry powder can comprise: about 1%, about 5%, about 10%, about 15%, about 20%, about 25%, about 30%, about 35%, about 40%, about 45%, about 50%, about 55%, about 50%, about 55%, about 60%, about 65%, about 70%, about 75%, about 80%, about 85%, about 90%, about 95%, or about 99% (weight percentage) of particles having VMGD of about 15 μm , 20 μm or more.

20 The above features can be combined. For example, the dry power can comprise about 50% of particles of about 5 μm or less (VMGD), about 25% of particles of about 5 to about 15 μm (VMGD), and about 25% of particles of about 15 μm or more (VMGD).

The dry powder can also comprise a mixture of particles having various mass median aerodynamic diameters (MMAD). For example, the dry powder can comprise particles of large sizes (e.g., MMAD 2: 15 μm , such as 2:20 μm or 20-30 μm) and of small sizes (e.g., MMAD: 5 μm , such as 1-3 μm) at a ratio (w:w) of: about 1:1, about 1:2, about 1:3, about 1:4, about 1:5, about 1:6, about 1:7, about 1:8, about 1:10, about 1:15, about 1:20, about 1:25, about 1:30, about 1:40, about 1:50, about 1:100, about 2:1, about 3:1, about 4:1, about 5:1, about 6:1, about 7:1, about 8:1, about 9:1, about 10:1, about 15:1, about 20:1, about 25:1, about 30:1, about 40:1,
30 about 50:1, or about 100:1, etc

Alternatively, the dry powder can comprise: about 1%, about 5%, about 10%, about 15%, about 20%, about 25%, about 30%, about 35%, about 40%, about 45%, about 50%, about 55%, about 50%, about 55%, about 60%, about 65%, about 70%, about 75%, about 80%, about 85%, about 90%, about 95%, or about 99% (weight percentage) of particles having MMAD of about 10 μm or less, preferably about 5 μm or less. Particles of 10 μm or less generally can reach lungs, and particles of 5 μm or less (e.g., 1-3 μm) generally can reach alveoli.

In another embodiment, the dry powder can comprise: about 1%, about 5%, about 10%, about 15%, about 20%, about 25%, about 30%, about 35%, about 40%, about 45%, about 50%, about 55%, about 50%, about 55%, about 60%, about 65%, about 70%, about 75%, about 80%,
10 about 85%, about 90%, about 95%, or about 99% (weight percentage) of particles having MMAD of between about 5 μm to about 20 μm , preferably between about 5 μm to about 15 μm , or between about 5 μm to about 10 μm .

In another embodiment, the dry powder can comprise: about 1%, about 5%, about 10%, about 15%, about 20%, about 25%, about 30%, about 35%, about 40%, about 45%, about 50%, about 55%, about 50%, about 55%, about 60%, about 65%, about 70%, about 75%, about 80%, about 85%, about 90%, about 95%, or about 99% (weight percentage) of particles having MMAD of about 15 μm or more, preferably 20 μm or more.

The above features can be combined. For example, the dry powder can comprise about 50% of particles of about 5 μm or less (MMAD), about 25% of particles of about 5 to about 15 μm
20 (MMAD), and about 25% of particles of about 15 μm or more (MMAD).

In certain embodiments, the dry powder may not have an excipient, which may be an anti-aggregation (or anti-bridging) excipient.

The dry powder can comprise a mixture of particles of various sizes, and is effective to substantially prevent or reduce particle bridging. In certain embodiments, at least about 40%, at least about 45%, at least about 50%, at least about 55%, at least about 60%, at least about 65%, at least about 70%, at least about 75%, at least 80%, at least about 85%, or at least about 90% of the NSAID (such as acetylsalicylic acid) in the dry powder is delivered to the alveolar spaces of a lung.

30 **6. METHODS FOR PREPARING DRY POWDERS AND DRY PARTICLES**

The dry particles and dry powders can be prepared using any suitable method. Many suitable methods for preparing dry powders and particles are conventional in the art, and include single and double emulsion solvent evaporation, spray drying, milling (*e.g.*, jet milling), blending, solvent extraction, solvent evaporation, phase separation, simple and complex coacervation, interfacial polymerization, suitable methods that involve the use of supercritical carbon dioxide (CO₂), and other suitable methods. Dry particles can be made using methods for making microspheres or microcapsules known in the art. These methods can be employed under conditions that result in the formation of dry particles having the desired aerodynamic properties (*e.g.*, aerodynamic diameter and geometric diameter). If desired, dry particles with desired properties, such as size and density, can be selected using suitable methods, such as sieving.

Spray Drying

Dry particles can be produced by spray drying. Suitable spray drying techniques are described, for example, by K. Masters in "*Spray Drying Handbook*", John Wiley & Sons, New York (1984); and spray drying techniques developed by BUCHI Laboratory Equipment or GEA Niro drying technology. Generally, during spray drying, heat from a hot gas such as heated air or nitrogen is used to evaporate a solvent from droplets formed by atomizing a continuous liquid feed. If desired, the spray drying or other instruments, *e.g.*, jet milling instrument, used to prepare the dry particles can include an inline geometric particle sizer that determines a geometric diameter of the respirable dry particles as they are being produced, and/or an inline aerodynamic particle sizer that determines the aerodynamic diameter of the respirable dry particles as they are being produced.

For spray drying, solutions, emulsions or suspensions that contain the components of the dry particles to be produced in a suitable solvent (*e.g.*, aqueous solvent, organic solvent, aqueous-organic mixture or emulsion) are distributed to a drying vessel via an atomization device. For example, a nozzle or a rotary atomizer may be used to distribute the solution or suspension to the drying vessel. For example, a rotary atomizer having a 4- or 24-vaned wheel may be used. Examples of suitable spray dryers that can be outfitted with either a rotary atomizer or a nozzle, include, Mobile Minor Spray Dryer or the Model PSD-1, both manufactured by Niro, Inc. (Denmark). Actual spray drying conditions will vary depending, in part, on the composition of the spray drying solution or suspension and material flow rates. The person of ordinary skill

will be able to determine appropriate conditions based on the compositions of the solution, emulsion or suspension to be spray dried, the desired particle properties and other factors. In general, the inlet temperature to the spray dryer is about 100°C to about 300°C, and preferably is about 220°C to about 285°C. The spray dryer outlet temperature will vary depending upon such factors as the feed temperature and the properties of the materials being dried. Generally, the outlet temperature is about 50°C to about 150°C, preferably about 90°C to about 120°C, or about 98°C to about 108°C. If desired, the respirable dry particles that are produced can be fractionated by volumetric size, for example, using a sieve, or fractionated by aerodynamic size, for example, using a cyclone, and/or further separated according to density using techniques known to those of skill in the art.

10 To prepare the dry particles of the subject technology, generally, a solution, emulsion or suspension that contains the desired components of the dry powder (i.e., a feed stock) is prepared and spray dried under suitable conditions. Preferably, the dissolved or suspended solids concentration in the feed stock is at least about 1g/L, at least about 2 g/L, at least about 5 g/L, at least about 10 g/L, at least about 15 g/L, at least about 20 g/L, at least about 30 g/L, at least about 40 g/L, at least about 50 g/L, at least about 60 g/L, at least about 70 g/L, at least about 80 g/L, at least about 90 g/L, or at least about 100 g/L. The feedstock can be provided by preparing a single solution or suspension by dissolving or suspending suitable components (e.g., salts, excipients, other active ingredients) in a suitable solvent. The solvent, emulsion or suspension can be prepared using any suitable methods, such as bulk mixing of dry and/or liquid components or static mixing of liquid components to form a combination. For example, a hydrophilic component (e.g., an aqueous solution) and a hydrophobic component (e.g., an organic solution) can be combined using a static mixer to form a combination. The combination can then be atomized to produce droplets, which are dried to form respirable dry particles. Preferably, the atomizing step is performed immediately after the components are combined in the static mixer.

30 In one example, dry particles that comprise acetylsalicylic acid can be prepared by spray drying. Spray drying is a commonly used method of drying a liquid feed through a hot gas. It is a method whereby solutions or slurries can be rapidly dried to particulate form by atomizing the liquid in a heated chamber. Typically, the hot gas can be air. When preparing chemically sensitive materials such as pharmaceuticals, or when solvents such as ethanol are used, oxygen-

free atmosphere may be required, such as nitrogen. Spray drying is frequently used in the food preparation industry and has become an important method for the dehydration of fluid foods such as milk, coffee, and egg powder. The process is also adaptable to preparations of pharmaceutical and chemical formulations.

The liquid feed varies depending on the material being dried and is not limited to food or pharmaceutical products, and may be a solution, colloid or suspension. The process is a one-step rapid method that typically eliminates additional processing. By controlling process conditions particles of the desired size can be reproducibly formed.

The spray drying can be conducted in the presence or absence of one or more excipients.

10 In some cases, excipients can be included with the active pharmaceutical ingredient such that a complex particle of active pharmaceutical ingredient (API) and excipient can be produced in a single step process. In other cases, an active pharmaceutical particulate preparation can be produced in a first spray-drying process, and that product then modified by the subsequent addition of one or more pharmaceutically acceptable excipients. In some cases it is possible to add excipients by a subsequent spray-drying process.

In some spray-drying methods, the liquid feed is pumped through an atomizer nozzle, or array of nozzles, that produce fine droplets that are introduced into the main drying chamber.

Atomizers can vary there being rotary, single fluid, two-fluid, and ultrasonic designs. These different designs provide a variety of advantages, applicability and disadvantages depending on 20 the particular spray drying process required. The hot drying gas can be passed as a concurrent or counter-current flow to the atomizer direction. The concurrent flow enables the particles to have a lower residence time within the system and the particle separator thus operates more efficiently. In some systems the particle separator is a cyclone device. The counter-current flow method enables a greater residence time of the particles in the chamber. Therefore, in general a spray-drying method will consist of the steps of pre-concentration of liquid, atomization, drying in a hot gas atmosphere, separation of the dried powder from moist gas, cooling, and then packaging of the finished product.

In one embodiment, feed solutions with acetylsalicylic acid concentrations of either 2% w/w, or 5% w/w, were prepared by adding acetylsalicylic acid to the appropriate solvent 30 followed by stirring until a homogeneous solution was obtained. A BUCHI spray dryer model B-290 Advanced was used in all experiments. The unit was equipped with a two fluid nozzle where

the nozzle and diameter were 1.4 mm and 0.7 mm, respectively. The high-performance cyclones were used to collect the dried product. The spray-drying unit was operated in open cycle, with the aspirator blowing nitrogen at 100% of capacity, corresponding to a flow rate of the dry nitrogen of approximately 40 kg per hour. The flow rate of atomization nitrogen was adjusted to 40 mm or 50 mm in the rotameter, depending on the particular trial. Before feeding the stock solution, the spray dryer was stabilized with the solvent. During the stabilization period, the solvent flow rate was adjusted in order to give the target outlet temperature. After stabilization of the outlet temperature, the feed of the spray dryer was commuted from the solvent to the product solution (inlet temperature was then readjusted to maintain the outlet temperature in the target value). At the end of the stock solution, the feed was once more commuted to solvent, in order to rinse the feed line and carry out a controlled shutdown.

10 The initial objective of these experiments was to isolate the amorphous form of acetylsalicylic acid, in order to fully characterize it. However, as was discovered from a review of the literature, acetylsalicylic acid presents a negative Tg (of -30°C), and as such the option of producing a crystalline size reduced active pharmaceutical with this technique was attempted. For that purpose, for solutions of acetylsalicylic acid in ethanol (the most suited solvent to dissolve the acetylsalicylic acid, given its high solubility and its approval for inhalation use) were prepared and spray dried as follows. Inlet temperature ranged from about 80°C to about 160°C. Outlet temperature was initially set to 65°C. In one experiment the outlet temperature 20 was increased to 100°C in an attempt to accelerate the amorphous-crystalline conversion, in the hopes that this would reduce losses that are typical of the transient glassy state of the material. However, increasing the outlet temperature did not produce any appreciable increase in overall yield of product. The rotameter was varied from about 40 mm to about 50 mm. Feed rate was typically about 5 mL per minute. Following spray drying, a number of analytical methods were used to evaluate the resulting product.

30 X-ray powder diffraction (XRPD) showed that in each of the four different batches prepared acetylsalicylic acid appeared to be crystalline in form, and the diffractogram was similar to that of the starting material. In addition, the spray dried products presented thermal grams that were identical to the input material. The overall yield ranged from about 55% to about 65%.

The melting temperature of the resulting spray dried product ranged from about 133°C to about 137°C, comparing favorably with the published melting point for acetylsalicylic acid (136°C). A measure of hygroscopic properties showed a weight change ranging from -.4% to about 1.2% when the products were exposed to an atmosphere with 95% relative humidity. These results suggest no issues with hygroscopic behavior and that with respect to this property, spray dried acetylsalicylic acid behaves in a manner similar to that of unprocessed acetylsalicylic acid.

10 Particle size distribution analysis showed that DV₁₀ ranged from about 0.9 μm to about 1.2 μm, DV₅₀ ranged from about 3 μm to about 6 μm, and DV₉₀ ranged from about 8 μm to about 24 μm. It was discovered that by reducing feed concentration of acetylsalicylic acid to 2% w/w, a smaller average particle size could be obtained, which was within typical inhalation range.

HPLC analysis showed acetylsalicylic acid purity to range from about 92% to about 98%, with the major "impurity" being salicylic acid, which ranged from about .3% to about .5%. Residual solvent ranged from about 90 ppm to about 150 ppm, well below the limits defined in the ICH Q3A guidelines.

20 The feedstock, or components of the feed-stock, can be prepared using any suitable solvent, such as an organic solvent, an aqueous solvent or mixtures thereof. Suitable organic solvents that can be employed include but are not limited to alcohols such as, for example, ethanol, methanol, propanol, isopropanol, butanols, and others. Other organic solvents include but are not limited to perfluorocarbons, dichloromethane, chloroform, ether, ethyl acetate, methyl tert-butyl ether and others. Co-solvents that can be employed include an aqueous solvent and an organic solvent, such as, but not limited to, the organic solvents as described above. Aqueous solvents include water and buffered solutions (such as phosphate buffer).

The feedstock or components of the feed stock can have any desired pH, viscosity or other properties. If desired, a pH buffer can be added to the solvent or co-solvent or to the formed mixture. Generally, the pH of the mixture ranges from about 3 to about 8.

Jet Milling

Respirable particles can also be produced by jet-milling. See, e.g., techniques developed 30 by Apex Process Technology or Jetpharma SA. Jet milling is a process of using highly compressed air or other gasses, usually in a vortex motion, to impact fine particles against each

other in a chamber. Jet mills are capable of reducing solids to particle sizes in the low-micron to submicron range. The grinding energy is created by gas streams from horizontal grinding air nozzles. Particles in the fluidized bed created by the gas streams are accelerated towards the center of the mill, colliding with slower moving particles. The gas streams and the particles carried in them create a violent turbulence and as the particles collide with one another they are pulverized.

In certain embodiments, jet-milling was able to produce acetylsalicylic acid particles with a FPF within the desired inhalable range for maximal deposition at the deepest levels of the lung. In some cases the DV₉₀ was less than about 9 μm , in some cases less than about 5 μm , and in 10 some cases less than about 3 μm . Particles produced by jet milling can be efficiently and predictably delivered from a dry powder inhaler device, and at least 25% of the particles are of a size that would be expected to deposit within the alveolar spaces of the lungs. In some cases at least 50% of the particles are of a size that would be expected to deposit within the alveolar spaces of the lungs. In one embodiment, at least 75% of the particles are of a size that would be expected to deposit within the alveolar spaces of the lungs. In another embodiment, at least 90% of the particles are of a size that would be expected to deposit within the alveolar spaces of the lungs.

Wet Polishing

20 Wet polishing is a process that combines a technology to attain a small particle size (either a bottom up technique such as controlled crystallization or nanocrystallization or top down technique such as high shear mixing or high pressure homogenization) with a suitable isolation technology (for example spray drying or filtration with a drying process). See, e.g., techniques developed by Hovione. These combinations can be used to tune the particle size and morphology to meet specific drug delivery needs. The method allows control of particle size distribution with tight spans and in-process sampling, and maintains crystalline state (little or no amorphous content).

30 Wet polishing technique can be repeated multiple times to achieve a particular size of about 500 nanometers or less. Studies were undertaken to investigate whether wet polishing could provide an appropriate method for producing acetylsalicylic acid particles of an inhalable size and which were deliverable from a dry powder inhaler device. Initially, a literature review

was conducted in order to determine the best candidate anti-solvent for use in reducing acetylsalicylic acid particle size by a wet-polishing method. Solvents were evaluated by their predicted ability with respect to minimal solubility of acetylsalicylic acid. From this review, the following candidate solvents were identified: water, benzene, toluene, hexane, n-heptane, dibutyl ether and di-isopropyl-ether.

After considering a number of factors in the end it was determined that only n-heptane and toluene fulfilled all the requirements, and therefore these were selected for further evaluation. Suspensions of acetylsalicylic acid at 5% w/w were prepared with the various anti-solvents, by charging the required amount of anti-solvent, charging the required amount of acetylsalicylic acid, and then stirring until a homogeneous suspension was obtained. The suspensions were qualitatively evaluated at room temperature and then filtered using a 0.45 μm membrane, which was then placed in an oven and dried at 60°C until the solvent had completely evaporated. A quantitative analysis was performed on the residue that remained in the membrane by weighing the membrane before and after the test. From this analysis it was determined that acetylsalicylic acid was partially soluble in toluene, and displayed a phobic behavior towards n-heptane.

Suspensions were prepared using either toluene or n-heptane at an acetylsalicylic acid concentration of 5% w/w. Each individual suspension was then subjected to a milling operation using a Microfluidics Model M-110EH-30 apparatus. Milling was conducted at a pressure of 50 bar using a 200 μm chamber for between 20 and 70 cycles. Input temperatures ranged from about 80°C to about 140°C. Output temperatures ranged from about 65°C to about 90°C. The process yield ranged from about 5% to about 25%.

Analysis of the resulting product revealed a DV₁₀ ranging from about 1.5 μm to about 3.3 μm , DV₅₀ ranging from about 3.3 μm to about 6.7 μm , and DV₉₀ ranging from about 6.3 μm to about 12.0 μm . HPLC assay of the final product revealed a composition of acetylsalicylic acid ranging from 90% to about 98%, with impurities ranging from about 1.4% to about 12%. The primary impurity was salicylic acid. It was also observed that all of the products obtained by wet polishing were more hygroscopic than the raw material, showing a water gain of about 5% when in the presence of 95% relative humidity.

In addition, when tested for aerodynamic performance, acetylsalicylic acid processed by wet polishing performed poorly in two different dry powder delivery devices. When examining devices loaded with either 15 mg or 40 mg, a significant amount of material (about 25 to 30%)

was retained within the inhalation device itself. Overall, the results suggested that wet polishing alone impacts to a significant extent the physical and chemical properties of acetylsalicylic acid, and therefore were seen to be less desirable for producing a pharmaceutical product for inhalation. Wet polishing may nonetheless be a suitable method for micronization of acetylsalicylic acid for other purposes.

In some cases it is possible to produce acetylsalicylic acid particles of a desired size using a process known as controlled crystallization. It is well known in the art that the crystalline state of most compounds is more thermodynamically stable than the amorphous state. As a result, producing acetylsalicylic acid in crystalline form is expected to improve stability of the active 10 ingredient. In addition, production of acetylsalicylic acid in crystalline form also provides the potential to modify the active ingredient to optimize various biochemical properties, such as solubility, dissolution rate and pH solubility profile (among others) in order to improve pharmacokinetic performance. In some cases, sequential crystallization steps can be used to improve the purity of the active ingredient and selectively remove undesirable impurities.

Similarly, through the proper selection of various solvents and anti-solvents, it is possible to manipulate physical characteristics such as crystal shape. It is well known that certain crystal shapes are difficult to handle both at the product development and manufacturing stages. For example, needles and flakes are widely regarded as less desirable particle shapes. It is possible however, to manipulate crystal formation in order to direct the final product to more suitable 20 crystal shapes. In some cases, it is possible to grow crystals with high aspect ratios using non-polar hydrocarbon solvents such as hexane or heptane. In contrast, crystals having a low aspect ratio can be produced using polar solvents such as methanol or ethanol. The addition of surface active "impurities" can also be used to inhibit crystal growth in certain planar forms.

The solubility of acetylsalicylic acid in a number of solvents was first evaluated prior to initiating controlled crystallization by addition of a suitable anti-solvent. The results are shown in the following Table 2.

Table 2 - Solubility of acetylsalicylic acid

| Solvent | T (°C) | g/ml | T (°C) | g/ml |
|---------|--------|-------|--------|-------|
| EtOH | 23 | 0.125 | 3 | 0.063 |
| Acetone | 23 | 0.200 | 3 | 0.143 |

| | | | | |
|---------|----|-------|---|-------|
| MeOH | 23 | 0.167 | 3 | 0.133 |
| DMF | 23 | 0.500 | - | - |
| THF | 23 | 0.500 | 3 | 0.250 |
| PEG-200 | 23 | 0.077 | - | - |

Next, several small crystallization experiments were carried out to evaluate the behavior of the acetylsalicylic acid in different systems. Each experiment consisted of dissolving 2 gm of acetylsalicylic acid in a solvent ($T = 20-25^\circ\text{C}$), and then adding this solution to the anti-solvent (100 vol. of anti-solvent at $\sim 5^\circ\text{C}$). The suspension obtained was stirred for 15 min and solid material collected by filtration and then dried. Table 3 summarizes the conditions of each experiment.

10 **Table 3 - Summary of the crystallization experiments**

| Solvent | (v/w) | T (°C) | Anti-Solvent | (v/w) | T (°C) | Crystals | Yield |
|--------------------------------|-------|--------|------------------|-------|--------|----------|-------|
| EtOH | 8 | 20-25 | H ₂ O | 100 | 4 | yes | 51.5 |
| EtOH | 8 | 20-25 | n-Hept | 100 | 4 | yes | 58.0 |
| EtOH | 8 | 20-25 | Toluene | 100 | 4 | no | - |
| EtOH | 8 | 20-25 | H ₂ O | 100 | 4 | yes | 53.0 |
| H ₂ SO ₄ | 0.05 | | | | | | |
| THF | 2.5 | 20-25 | H ₂ O | 100 | 4 | yes | 53.5 |
| THF | 2.5 | 20-25 | Toluene | 100 | 4 | yes | 45.5 |
| THF | 2.5 | 20-25 | n-Hept | 100 | 4 | yes | 89.0 |
| MeOH | 6 | 20-25 | H ₂ O | 100 | 4 | yes | 64.5 |
| MeOH | 6 | 20-25 | n-Hept | 100 | 4 | yes | 17.0 |
| MeOH | 6 | 20-25 | Toluene | 100 | 4 | no | - |
| Acetone | 7 | 20-25 | H ₂ O | 100 | 3 | yes | 43.0 |
| Acetone | 7 | 20-25 | n-Hept | 100 | 4 | yes | 71.5 |
| Acetone | 7 | 20-25 | Toluene | 100 | 4 | yes | 34.5 |

Excipients

Particles described herein can be encapsulated, e.g., by a pharmaceutical excipient such as lactose, sugar, or a polymer.

In addition, particles described herein can be mixed and/or coated with various pharmaceutically acceptable excipients. Excipients can be included in order to improve aerodynamic performance of the active drug, to improve bioavailability, increase stability, to modulate pH, to provide sustained release properties, to provide taste-masking of an irritating drug and/or to improve pharmacokinetic performance.

In terms of suitable formulations for inhalation, not only is particle size important in order to reach a desired region of the respiratory system, but also, the formulation must be acceptable to patients. In early aspects of the studies involved in the present invention, it was discovered that pure acetylsalicylic acid is quite irritating to the respiratory system and thus 10 unlikely to be acceptable to users. Consequently, it became necessary to find a suitable excipient in order to mask this property of acetylsalicylic acid.

With dry powder formulations, excipients can also provide a carrier function to reduce clumping of the active pharmaceutical ingredient and to improve suspension of the formulation in the airflow as the pharmaceutical preparation is being inhaled. Such carriers can include substances such as, but not limited to, sugars/sugar alcohols such as glucose, saccharose, lactose and fructose, starches or starch derivatives, oligosaccharides such as dextrins, cyclodextrins and their derivatives, polyvinylpyrrolidine, alginic acid, tylose, silicic acid, cellulose, cellulose derivatives, sugar alcohols such as mannitol or sorbitol, calcium carbonate, calcium phosphate, lactose, lactitol, dextrates, dextrose, maltodextrin, saccharides including monosaccharides, 20 disaccharides, polysaccharides; sugar alcohols such as arabinose, ribose, mannose, sucrose, trehalose, maltose and dextran.

In some cases, an excipient can be provided in order to coat the active pharmaceutical ingredient, thus "masking" it. Masking is especially useful when the unmodified active pharmaceutical is irritating or otherwise unpleasant to the recipient. For example, in some cases it has been shown that coating a bitter molecule with a hydrogenated oil and surfactant combination is effective to cover the otherwise unpleasant taste of the active ingredient.

One such excipient, discovered as part of the current studies to be useful in reducing acetylsalicylic acid irritation upon inhalation, are zwitterionic phospholipids, including surfactant compounds. Examples of suitable phospholipid excipients include, without limitation, 30 phosphatidylcholines, phosphatidylethanolamines, phosphatidylinositol, phosphatidylserines, sphingomyelin or other ceramides, as well as phospholipid containing oils such as lecithin oils.

Combinations of phospholipids, or mixtures of a phospholipid(s) and other substance(s), may be used. In one embodiment, the phospholipids used as excipients are soy lecithin. In another embodiment, the phospholipid is endogenous to the lung.

Non-limiting examples of the phospholipids that may be used in the present composition include, soy lecithin, dipalmitoylphosphatidylcholine (DPPC), distearoylphosphatidylcholine (DSPC), dilaurylolyphosphatidylcholine (DLPC), dimyristoylphosphatidylcholine (DMPC), dioleoylphosphatidylcholine (DOPC), dilaurylolyphosphatidylglycerol (DLPG), dimyristoylphosphatidylglycerol (DMPG), dipalmitoylphosphatidylglycerol (DPPG), distearoylphosphatidylglycerol (DSPG), dioleoylphosphatidylglycerol (DOPG), dimyristoyl phosphatidic acid (DMPA), dimyristoyl phosphatidic acid (DMPA), dipalmitoyl phosphatidic acid (DPPA), dipalmitoyl phosphatidic acid (DPPA), dimyristoyl phosphatidylethanolamine (DMPE), dipalmitoyl phosphatidylethanolamine (DPPE), dimyristoyl phosphatidylserine (DMPS), dipalmitoyl phosphatidylserine (DPPS), dipalmitoyl sphingomyelin (DPSP), and distearoyl sphingomyelin (DSSP).

In one embodiment, soy lecithin, dipalmitoyl phosphatidylcholine (DPPC), distearoyl phosphatidylcholine (DSPC) or a mixture thereof are used as an excipient. For example, DPPC and DSPC are natural constituents of pulmonary surfactant. Pulmonary surfactant is a surface-active lipoprotein complex produced by type II alveolar cells. The proteins and lipids that make up the surfactant have a hydrophilic region and a hydrophobic region. By adsorbing to the air-water interface of alveoli with the hydrophilic head groups in the water and the hydrophobic tails facing towards the airspace, surfactants are effective to reduce surface tension to near-zero levels and permit expansion of the lung with less force than would otherwise be required. Consequently, pulmonary surfactant, by reducing surface tension, allows the lung to inflate much more easily, thus reducing the effort required to inflate the lungs.

An excipient may be used to mask taste and/or improve the aerodynamic performance of the present particles. The excipient may be any substance as described herein. The excipient(s) may be present at levels ranging from about 0% to about 99% (w/w), from about 0.01% to about 80% (w/w), from about 0.05% to about 70% (w/w), from about 0.1% to about 60% (w/w), from about 0.1% to about 50% (w/w), from about 0.1% to about 40% (w/w), from about 0.1% to about 30% (w/w), from about 0.1% to about 20% (w/w), from about 0.1% to about 10% (w/w), from about 0.05% to about 8% (w/w), from about 0.1% to about 6% (w/w), from about 5% to about 10%

(w/w), from about 3% to about 8% (w/w), from about 2% to about 6% (w/w), from about 0.1% to about 5% (w/w), from about 0.1% to about 4% (w/w), from about 0.1% to about 3% (w/w), from about 0.1% to about 2% (w/w), from about 0.1% to about 1% (w/w), from about 1% to about 6% (w/w), from about 1% to about 5% (w/w), from about 1% to about 4% (w/w), or from about 1% to about 3% (w/w) of the particles. In certain embodiments, one or more excipients (e.g., one or more phospholipids) are present at levels in a range from about 0.1% to about 10% (w/w), from about 1% to about 5% (w/w), about 0.1%, about 5% (w/w), about 3%, or about 10% (w/w) of the particles.

Acetylsalicylic acid alone may be too irritating and evoke a choking response. In one 10 embodiment, after coating acetylsalicylic acid with a surfactant, the modified formulation is well tolerated when administered by inhalation either orally or nasally. Also surprising was the observation that nasally delivered surfactant coated acetylsalicylic acid was able to alleviate symptoms of headache and nasal congestion within seconds of administration. To the inventors' knowledge, this is the first demonstration that acetylsalicylic acid can provide rapid relief from headache and/or nasal congestion when delivered in this manner.

In addition, in some embodiments, the surfactant can be provided in combination with one or more additional excipients including absorbents, acidifiers, alkalizers, buffers, antimicrobial agents, antioxidants, binders, solubilizing agents, solvents, viscosity modifiers, humectants and combinations thereof. In some embodiments the formulation includes salts in 20 amounts effective to render the dissolved formulation isosmotic with the lung.

Respirable dry particles and dry powders can be fabricated and then separated, for example, by filtration or centrifugation by means of a cyclone, to provide a particle sample with a preselected size distribution. For example, greater than about 30%, greater than about 40%, greater than about 50%, greater than about 60%, greater than about 70%, greater than about 80%, or greater than about 90% of the respirable dry particles in a sample can have a diameter within a selected range. The selected range within which a certain percentage of the respirable dry particles fall can be, for example, any of the size ranges described herein, such as between about 0.1 to about 3 μm VMGD.

The diameter of the respirable dry particles, for example, their VMGD, can be measured 30 using an electrical zone sensing instrument such as a Multisizer lie, (Coulter Electronic, Luton, Beds, England), or a laser diffraction instrument such as a HELOS system (Sympatec, Princeton,

NJ). Other instruments for measuring particle geometric diameter are well known in the art. The diameter of respirable dry particles in a sample will range depending upon factors such as particle composition and methods of synthesis. The distribution of size of respirable dry particles in a sample can be selected to permit optimal deposition within targeted sites within the respiratory system.

Experimentally, aerodynamic diameter can be determined using time of flight (TOF) measurements. For example, an instrument such as the Model 3225 Aerosizer DSP Particle Size Analyzer (Amherst Process Instrument, Inc., Amherst, MA) can be used to measure aerodynamic diameter. The Aerosizer measures the time taken for individual respirable dry particles to pass

10 between two fixed laser beams.

Aerodynamic diameter also can be experimentally determined directly using conventional gravitational settling methods, in which the time required for a sample of respirable dry particles to settle a certain distance is measured. Indirect methods for measuring the mass median aerodynamic diameter include the Andersen Cascade Impactor (ACI) and the multi-stage liquid impinger (MSLI) methods. Another method of measuring the aerodynamic diameter is with a Next Generation Impactor (NGI). The NGI operates on similar principles of inertial impaction as the ACI. The NGI consists of seven stages and can be calibrated at flow rates of 30, 60, and 100 LPM. In contrast to the ACI, for which the impactor stages are stacked, the stages of the NGI are all in one plane. Collection cups are used to collect the particles below each stage of

20 the NGI. U.S. Patent No. 8,614,255. The methods and instruments for measuring particle aerodynamic diameter are well known in the art.

The dry powder of the present formulation comprises substantially dry particles having a mass median aerodynamic diameter (MMAD) within a range of about 0.5 μm to about 10 μm , wherein the dry powder further comprises one or more phospholipids in an amount ranging from about 0.1% (w/w) to about 10% (w/w) of the dry particles. The particles may have an MMAD size distribution where the particles exhibit: (i) a DV90 less than about 20 μm , a DV50 less than about 7 μm , and a DV10 less than about 2 μm ; (ii) a DV90 less than about 10 μm , a DV50 less than about 4 μm , and a DV10 less than about 1 μm ; or (iii) a DV90 less than about 6 μm , a DV50 less than about 3 μm , and a DV10 less than about 1 μm .

30 The dry powder may comprise particles coated with a pharmaceutically acceptable excipient. The pharmaceutically acceptable excipient can be a phospholipid having surfactant

properties, such as dipalmitoyl phosphatidylcholine (DPPC), distearoyl phosphatidylcholine (DSPC) or soy lecithin, in an amount ranging from about 0.1% to about 10% w/w or in an amount ranging from about 1% to about 5% w/w. In one embodiment, the weight percentage of the DSPC on the dry particles is 5% w/w. In another embodiment, the respirable dry powder of the soy lecithin of the dry particles is 0.1% w/w.

The drug delivery system of the present invention is effective in reducing the risk of a thromboembolic event or treat thrombosis. The dose of acetylsalicylic acid may be present at amounts ranging from about 5 to about 40 mg. The formulation may further comprise clopidogrel. The respirable dry powders can have an emitted dose ranging from about 75% to about 95%.

10 In one embodiment, the pharmaceutically acceptable excipient is DSPC, the respirable dry powders substantially comprise dry particles having a MMAD ranging from about 3 to about 4 μ m and an emitted dose greater than about 90%. In this embodiment, the mass percent of stages in an NGI testing apparatus of the respirable powder yields are at, stage 1 about 10% to about 13%, stage 2, about 20% to about 23%, stage 3, about 13% to about 15%, and stage 4, about 5% to about 6% and fine particle fraction ranges from about 45% to about 55%. Ranges between 80%-120% of actual percentages as set forth above are encompassed within each embodiment.

20 In another embodiment, the pharmaceutically acceptable excipient is soy lecithin, the respirable dry powders substantially comprise dry particles having a MMAD ranging from about 2.0 to about 3.0 μ m and an emitted dose ranging from about 70% to about 85%. In this embodiment, the mass percent of stages in an NGI testing apparatus of the respirable powder yields are at, stage 1 about 5% to about 10%, stage 2, about 10% to about 18%, stage 3, about 15% to about 20%, and stage 4, about 10% to about 15% and fine particle fraction ranges from about 50% to about 70%. Ranges between 80%-120% of actual percentages set forth above are encompassed within each embodiment.

30 Tap density is a measure of the envelope mass density characterizing a particle. The envelope mass density of a particle of a statistically isotropic shape is defined as the mass of the particle divided by the minimum sphere envelope volume within which it can be enclosed. Features which can contribute to low tap density include irregular surface texture and porous structure. Tap density can be measured by using instruments known to those skilled in the art such as the Dual Platform Microprocessor Controlled Tap Density Tester (Vankel, NC), a

GeoPyc™ instrument (Micrometries Instrument Corp., Norcross, GA), or SOTAX Tap Density Tester model TD2 (SOTAX Corp., Horsham, PA). Tap density can be determined using the method of USP Bulk Density and Tapped Density, United States Pharmacopoeia convention, Rockville, MD, 10th Supplement, 4950-4951, 1999.

Fine particle fraction (FPF) can be used as one way to characterize the aerosol performance of a dispersed powder. Fine particle fraction describes the size distribution of airborne respirable dry particles. Gravimetric analysis, using a Cascade impactor, is one method of measuring the size distribution, or fine particle fraction, of airborne respirable dry particles. The Andersen Cascade Impactor (ACI) is an eight-stage impactor that can separate aerosols into nine distinct fractions based on aerodynamic size. The size cutoffs of each stage are dependent upon the flow rate at which the ACI is operated. The ACI is made up of multiple stages consisting of a series of nozzles (i.e., a jet plate) and an impaction surface (i.e., an impaction disc). At each stage an aerosol stream passes through the nozzles and impinges upon the surface. Respirable dry particles in the aerosol stream with a large enough inertia will impact upon the plate. Smaller respirable dry particles that do not have enough inertia to impact on the plate will remain in the aerosol stream and be carried to the next stage. Each successive stage of the ACI has a higher aerosol velocity in the nozzles so that smaller respirable dry particles can be collected at each successive stage.

If desired, a two-stage collapsed ACI can also be used to measure fine particle fraction.

The two-stage collapsed ACI consists of only the top two stages of the eight-stage ACI and allows for the collection of two separate powder fractions. Specifically, a two-stage collapsed ACI is calibrated so that the fraction of powder that is collected on stage one is composed of respirable dry particles that have an aerodynamic diameter of less than 5.6 μm and greater than 3.4 μm . The fraction of powder passing stage one and depositing on a collection filter is thus composed of respirable dry particles having an aerodynamic diameter of less than 3.4 μm . The airflow at such a calibration is approximately 60 L/min. Formulation produced by the methods described herein can be effectively delivered at airflow rates ranging from about 20L/min to about 60 L/min.

The FPF (<5.6) has been demonstrated to correlate to the fraction of the powder that is

able to make it into the lung of the patient, while the FPF (<3.4) has been demonstrated to

correlate to the fraction of the powder that reaches the deep lung of a patient. These correlations provide a quantitative indicator that can be used for particle optimization.

An ACI can be used to approximate the emitted dose, which herein is called gravimetric recovered dose and analytical recovered dose. "Gravimetric recovered dose" is defined as the ratio of the powder weighed on all stage filters of the ACI to the nominal dose. "Analytical recovered dose" is defined as the ratio of the powder recovered from rinsing all stages, all stage filters, and the induction port of the ACI to the nominal dose. The FPF TD (<5.0) is the ratio of the interpolated amount of powder depositing below 5.0 μm on the ACI to the nominal dose. The FPF RD (<5.0) is the ratio of the interpolated amount of powder depositing below 5.0 μm on the

10 ACI to either the gravimetric recovered dose or the analytical recovered dose.

Another way to approximate emitted dose is to determine how much powder leaves its container, *e.g.* capsule or blister, upon actuation of a dry powder inhaler (DPI). This takes into account the percentage leaving the capsule, but does not take into account any powder depositing on the DPI. The emitted dose is the ratio of the weight of the capsule with the dose before inhaler actuation to the weight of the capsule after inhaler actuation. This measurement can also be called the capsule emitted powder mass (CEPM).

A Multi-Stage Liquid Impinger (MSLI) is another device that can be used to measure particle size distribution or fine particle fraction. The Multi-stage liquid Impinger operates on the same principles as the ACI, although instead of eight stages, MSLI has five. Additionally, each

20 MSLI stage consists of an ethanol-wetted glass frit instead of a solid plate. The wetted stage is used to prevent particle bounce and re-entrainment, which can occur when using the ACI. U.S. Patent No. 8,614,255.

The subject technology also relates to a respirable dry powder or respirable dry particles produced using any of the methods described herein.

The dry particles of the subject technology can also be characterized by the chemical stability of the salts or the excipients that the dry particles comprise. The chemical stability of the constituent salts can affect important characteristics of the particles including shelf-life, proper storage conditions, acceptable environments for administration, biological compatibility, and effectiveness of the salts. Chemical stability can be assessed using techniques well known in the art. One example of a technique that can be used to assess chemical stability is reverse phase high performance liquid chromatography (RP-HPLC).

Respirable dry particles of the subject technology include salts that are generally stable over a long period time.

If desired, the dry particles and dry powders described herein can be further processed to increase stability. An important characteristic of pharmaceutical dry powders is whether they are stable at different temperature and humidity conditions. Unstable powders will absorb moisture from the environment and agglomerate, thus altering particle size distribution of the powder.

Excipients, such as maltodextrin, may be used to create more stable particles and powders. The maltodextrin may act as an amorphous phase stabilizer and inhibit the components from converting from an amorphous to crystalline state. Alternatively, a post-processing step to 10 help the particles through the crystallization process in a controlled way (e.g., on the baghouse at elevated humidity) can be employed with the resultant powder potentially being further processed to restore their dispersibility if agglomerates formed during the crystallization process, such as by passing the particles through a cyclone to break apart the agglomerates. Another possible approach is to optimize around process conditions that lead to manufacturing particles that are more crystalline and therefore more stable. Another approach is to use different excipients, or different levels of current excipients to attempt to manufacture more stable forms of the salts.

The respirable dry particles and dry powders described herein are suitable for inhalation therapies. The respirable dry particles may be fabricated with the appropriate material, surface 20 roughness, diameter, and tap density for localized delivery to selected regions of the respiratory system such as the deep lung or upper or central airways. For example, higher density or larger respirable dry particles may be used for upper airway delivery, or a mixture of varying size respirable dry particles in a sample, provided with the same or a different formulation, may be administered to target different regions of the lung in one administration.

In order to relate the dispersion of powder at different inhalation flow rates, volumes, and from inhalers of different resistances, the energy required to perform the inhalation maneuver can be calculated. Inhalation energy can be calculated from the equation $E=R^2Q^2V$ where E is the inhalation energy in Joules, R is the inhaler resistance in $kPa^{1/2}/LPM$, Q is the steady flow rate in L/min and V is the inhaled air volume in L.

30 Healthy adult populations are predicted to be able to achieve inhalation energies ranging from 2.9 to 22 Joules by using values of peak inspiratory flow rate (PIFR) measured by Clarke et

al. (Journal of Aerosol Med, 6(2), p.99-110, 1993) for the flow rate Q from two inhaler resistances of 0.02 and 0.055 kPa1/2/LPM, with a inhalation volume of 2L based on both FDA guidance documents for dry powder inhalers and on the work of Tiddens *et al.* (Journal of Aerosol Med, 19, (4), p.456-465, 2006) who found adults averaging 2.2L inhaled volume through a variety of DPIs.

Dry powder particles can also be prepared using cone-jet mode of electrohydrodynamic atomization, as described by Li *et al.*, Chemical Engineering Science 61 (2006) 3091 - 3097. For example, an acetylsalicylic acid solution flowing through a needle can be subjected to an electric field to generate droplets. The method is said to generate a near-monodispersed distribution of droplet relicts, leading to form acetylsalicylic acid particulate crystals.

7. METHODS OF TREATMENT

In other aspects, the subject technology is a method for treating (including prophylactic treatment or reducing the risk) of a cardiovascular disease (such as thrombosis), comprising administering to the respiratory tract of a subject in need thereof an effective amount of respirable dry particles or dry powder, as described herein.

Cardiovascular diseases include, for example, atherosclerosis, coronary artery disease (CAD), angina pectoris (commonly known as "angina"), thrombosis, ischemic heart disease, coronary insufficiency, peripheral vascular disease, myocardial infarction, cerebrovascular disease (such as stroke), transient ischemic attack, arteriolosclerosis, small vessel disease, elevated cholesterol, intermittent claudication or hypertension.

The respirable dry particles and dry powders can be administered to the respiratory tract of a subject in need thereof using any suitable method, such as instillation techniques, and/or an inhalation device, such as a dry powder inhaler (DPI) or metered dose inhaler (MDI). A number of DPis are available, such as, the inhalers disclosed is U. S. Patent No. 4,995,385 and 4,069,819, Spinhaler® (Fisons, Loughborough, U.K.), Rotahalers®, Diskhaler® and Diskus® (GlaxoSmithKline, Research Triangle Technology Park, North Carolina), FlowCapss®, XCaps (Hovione, Loures, Portugal), Inhalators® (BoehringerIngelheim, Germany), Aerolizer® (Novartis, Switzerland), and others known to those skilled in the art.

Generally, inhalation devices (*e.g.*, DPis) are able to deliver a maximum amount of dry powder or dry particles in a single inhalation, which is related to the capacity of the blisters,

capsules (*e.g.* size 000, 00, OE, 0, 1, 2, 3, and 4, with respective volumetric capacities of 1.37ml, 950 μ l, 770 μ l, 680 μ l, 480 μ l, 360 μ l, 270 μ l, and 200 μ l) or other means that contain the dry particles or dry powders within the inhaler. Accordingly, delivery of a desired dose or effective amount may require two or more inhalations. Preferably, each dose that is administered to a subject in need thereof contains an effective amount of respirable dry particles or dry powder and is administered using no more than about 4 inhalations. For example, each dose of respirable dry particles or dry powder can be administered in a single inhalation or 2, 3, or 4 inhalations. The respirable dry particles and dry powders are preferably administered in a single, breath-activated step using a breath-activated DPI. When this type of device is used, the energy of the subject's 10 inhalation both disperses the respirable dry particles and draws them into the respiratory tract.

The respirable dry particles or dry powders can be delivered by inhalation to a desired area within the respiratory tract, as desired. It is well known that particles with an MMAD of about 1 μ m to about 3 μ m, can be effectively delivered to the deep lung regions such as the alveolar spaces. Larger aerodynamic diameters, for example, from about 3 μ m to about 5 μ m can be delivered to the central and upper airways.

For dry powder inhalers, oral cavity deposition is dominated by inertial impaction and so characterized by the aerosol's Stokes number (DeHaan *et al.* *Journal of Aerosol Science*, 35 (3), 309-331, 2003). For equivalent inhaler geometry, breathing pattern and oral cavity geometry, the Stokes number, and so the oral cavity deposition, is primarily affected by the aerodynamic size 20 of the inhaled powder. Hence, factors that contribute to oral deposition of a powder include the size distribution of the individual particles and the dispersibility of the powder. If the MMAD of the individual particles is too large, *e.g.* above 5 μ m, then an increasing percentage of powder will deposit in the oral cavity. Likewise, if a powder has poor dispersibility, it is an indication that the particles will leave the dry powder inhaler and enter the oral cavity as agglomerates. Agglomerated powder will perform aerodynamically like an individual particle as large as the agglomerate, therefore even if the individual particles are small (*e.g.*, MMAD of about 5 μ m or less), the size distribution of the inhaled powder may have an MMAD of greater than about 5 μ m, leading to enhanced oral cavity deposition.

Therefore, it is desirable to have a powder in which the particles are small (*e.g.*, MMAD 30 of 5 μ m or less, *e.g.* between about 1 μ m to 5 μ m), and are highly dispersible (*e.g.* 1/4 bar or alternatively, 0.5/4 bar of 2.0, and preferably less than 1.5). More preferably, the respirable dry

powder is comprised of respirable dry particles with an MMAD between 1 to 4 μm or 1 to 3 μm , and have a 1/4 bar less than 1.4, or less than 1.3, and more preferably less than 1.2.

The absolute geometric diameter of the particles measured at 1 bar using the HELOS system is not critical provided that the particle's envelope density is sufficient such that the MMAD is in one of the ranges listed above, wherein MMAD is VMGD times the square root of the envelope density (MMAD = VMGD*sqrt (envelope density)). If it is desired to deliver a high unit dose of salt using a fixed volume-dosing container, then, particles of higher envelop density are desired. High envelope density allows for more mass of powder to be contained within the fixed volume-dosing container. Preferable envelope densities are greater than 0.1 g/cm^3 , greater than 0.25 g/cm^3 , greater than 0.4 g/cm^3 , greater than 0.5 g/cm^3 , and greater than 0.6 g/cm^3 .

10 The respirable dry powders and particles of the subject technology can be employed in compositions suitable for drug delivery via the respiratory system. For example, such compositions can include blends of the respirable dry particles of the subject technology and one or more other dry particles or powders, such as dry particles or powders that contain another active agent, or that consist of or consist essentially of one or more pharmaceutically acceptable excipients.

Respirable dry powders and dry particles suitable for use in the methods of the subject technology can travel through the upper airways (i.e., the oropharynx and larynx), the lower airways, which include the trachea followed by bifurcations into the bronchi and bronchioli, and 20 through the terminal bronchioli which in turn divide into respiratory bronchiole leading then to the ultimate respiratory zone, the alveoli or the deep lung. In one embodiment of the subject technology, most of the mass of respirable dry powders or particles deposit in the deep lung. In another embodiment of the subject technology, delivery is primarily to the central airways. In another embodiment, delivery is to the upper airways.

The respirable dry particles or dry powders of the subject technology can be delivered by inhalation at various parts of the breathing cycle (e.g., laminar flow at mid-breath). An advantage of the high dispersibility of the dry powders and dry particles of the subject technology is the ability to target deposition in the respiratory tract. For example, breath controlled delivery of nebulized solutions is a recent development in liquid aerosol delivery 30 (Dalby *et al.* in Inhalation Aerosols, edited by Hickey 2007, p. 437). In this case, nebulized droplets are released only during certain portions of the breathing cycle. For deep lung delivery,

droplets are released in the beginning of the inhalation cycle, while for central airway deposition, they are released later in the inhalation.

The dry powders of this subject technology provide advantages for targeting the timing of drug delivery in the breathing cycle and also location in the human lung. Because the respirable dry powders of the subject technology can be dispersed rapidly, such as within a fraction of a typical inhalation maneuver, the timing of the powder dispersal can be controlled to deliver an aerosol at specific times within the inhalation.

With a highly dispersible powder, the complete dose of aerosol can be dispersed at the beginning portion of the inhalation. While the patient's inhalation flow rate ramps up to the peak 10 inspiratory flow rate, a highly dispersible powder will begin to disperse already at the beginning of the ramp up and could completely disperse a dose in the first portion of the inhalation. Since the air that is inhaled at the beginning of the inhalation will ventilate deepest into the lungs, dispersing the most aerosol into the first part of the inhalation is preferable for deep lung deposition. Similarly, for central deposition, dispersing the aerosol at a high concentration into the air which will ventilate the central airways can be achieved by rapid dispersion of the dose near the mid to end of the inhalation. This can be accomplished by a number of mechanical and other means such as a switch operated by time, pressure or flow rate that diverts the patient's inhaled air to the powder to be dispersed only after the switch conditions are met.

Aerosol dosage, formulations and delivery systems may be selected for a particular 20 therapeutic application, as described, for example, in Gonda, I. "Aerosols for delivery of therapeutic and diagnostic agents to the respiratory tract," in Critical Reviews in Therapeutic Drug Carrier Systems, 6: 273-313 (1990); and in Moren, "Aerosol Dosage Forms and Formulations," in Aerosols in Medicine, Principles, Diagnosis and Therapy, Moren, *et al.*, Eds., Elsevier, Amsterdam (1985).

Suitable intervals between doses that provide the desired therapeutic effect can be determined based on the severity of the condition, overall well-being of the subject and the subject's tolerance to respirable dry particles and dry powders and other considerations. Based on these and other considerations, a clinician can determine appropriate intervals between doses. Respirable dry particles and dry powders may be administered once, twice or three times a day 30 or on an as needed basis.

In various embodiments the amount of NSAID, such as acetylsalicylic acid, delivered to the respiratory tract (e.g., lungs, respiratory airway) is about 0.001 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.002 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.005 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.01 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.02 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.05 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.075 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.1 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.2 mg/kg body weight/dose to about 2 mg/kg body weight/dose, about 0.5 mg/kg body weight/dose to about 2 mg/kg body weight/dose, or about 0.75 mg/kg body weight/dose to about 2 mg/kg body weight/dose.

10

In certain embodiments, at least about 50%, at least about 60%, at least about 70%, at least about 75%, at least about 80%, at least about 85%, at least about 90%, at least about 95%, or at least about 99%, of the administered acetylsalicylic acid reaches the systemic circulation of a subject within about 60 minutes upon administration, or within about 40 minutes upon administration, or within about 30 minutes upon administration, or within about 20 minutes upon administration, or within about 15 minutes upon administration, or within about 5 minutes upon administration.

The dosing of acetylsalicylic acid may be adjusted so that PGI2 synthesis capacity of the nasal, bronchial or pulmonary epithelial or endothelial cells, including nasal mucosa cells, is not inhibited.

20

In certain embodiments, the method and delivery devices described herein can deliver acetylsalicylic acid, and pharmacologically active metabolic byproducts of acetylsalicylic acid thereof, to the systemic circulation, at levels that are substantially the same, or higher as compared to those delivered by oral administration of about 30 mg of acetylsalicylic acid, about 40 mg of acetylsalicylic acid, about 50 mg of acetylsalicylic acid, about 80 mg of acetylsalicylic acid or about 160 mg of acetylsalicylic acid.

The doses of acetylsalicylic acid administered in order to achieve a level (or an average level among a population of patients) that is substantially the same, or higher as compared to those delivered by oral administration of about 30 mg, about 40 mg, about 50 mg, about 80 mg, or about 160 mg of acetylsalicylic acid can be determined by conventional methods. The dosing,

30

administration techniques and schedules are known in the art and are within the ability of the skilled clinician. For example, the serum level of acetylsalicylic acid, or a metabolite thereof, in a subject (or average serum level among a population of subjects) can be determined by conventional pharmacokinetic or pharmacodynamics studies.

In certain embodiments, the method and delivery devices described herein can deliver acetylsalicylic acid to the systemic circulation such that the circulating plasma level of acetylsalicylic acid is at least about 1 μ g/mL, at least about 2 μ g/mL, at least about 3 μ g/mL, at least about 4 μ g/mL, at least about 5 μ g/mL, at least about 6 μ g/mL, within about 60 minutes upon administration, or within about 40 minutes upon administration, or within about 30 minutes upon administration, or within about 20 minutes upon administration, or within about 15 minutes upon administration, or within about 5 minutes upon administration.

In other embodiments, the method and delivery devices described herein can deliver acetylsalicylic acid to the systemic circulation such that circulating plasma level of salicylate is about 8 μ g/mL, about 9 μ g/mL, about 10 μ g/mL, about 11 μ g/mL, about 12 μ g/mL, or about 15 μ g/mL, within about 60 minutes upon administration, or within about 40 minutes upon administration, or within about 30 minutes upon administration, or within about 20 minutes upon administration, or within about 15 minutes upon administration, or within about 5 minutes upon administration.

If desired or indicated, the respirable dry particles and dry powders described herein can be administered with one or more other therapeutic agents. The other therapeutic agents can be administered by any suitable route, such as orally, parenterally (e.g., intravenous, intraarterial, intramuscular, or subcutaneous injection), topically, by inhalation (e.g., intrabronchial, intranasal or oral inhalation, intranasal drops), rectally, vaginally, and the like. The respirable dry particles and dry powders can be administered before, substantially concurrently with, or subsequent to administration of the other therapeutic agent. Preferably, the respirable dry particles and dry powders and the other therapeutic agent are administered so as to provide substantial overlap of their pharmacologic activities.

The following examples of specific aspects for carrying out the present invention are offered for illustrative purposes only, and are not intended to limit the scope of the present invention in any way.

Example 1 Development of Acetylsalicylic acid Particles for Inhalation

This study was designed to develop phospholipid coated acetylsalicylic acid particles with particle size less than 2.0 μm for deep lung tissue delivery. This development work was carried out to achieve following target particle size: Dv50 of 0.5 nm to 2.0 μm ; Dv90 of 1.5 to 2.0 μm .

Jet milling was selected as a method for micronization of acetylsalicylic acid particles to achieve target particle size. Jet milling operation was successfully reproduced with Dv50s of 0.4 μm and Dv90s of 1.3 μm and 1.6 μm for the two batches manufactured. Micronized particles were then spray-dried using DSPC (1,2-distearoyl-(sn)-glycero-3-phosphocholine) or soy lecithin to reduce particle agglomeration and irritation on inhalation. 79% yield for 10 DSPC/acetylsalicylic acid and 54% yield for soy lecithin/acetylsalicylic acid were obtained.

Particle size analysis at each step was carried out. Spray-dried DSPC/acetylsalicylic acid particles ranged from 1.8 to 3.6 μm and lecithin/acetylsalicylic acid particles ranged from 1.7 to 3.3 μm .

DSC studies showed no change in the crystalline structure of acetylsalicylic acid before and after spray-drying with DSPC. TGA showed 0.6% moisture content for both pre- and post-spray-dried particles indicating absence of any residual solvent after spray-drying.

Formulation Development

20 1. Acetylsalicylic Acid

Rhodine 3040 US obtained from Rhodia Inc. was used for all experiments. Particles were dispersed in 0.1% w/w docusate sodium in water and observed under a light microscope to confirm particle size. Particles ranging from 66 to 280 μm were observed confirming data from certificate of analysis, though observed “rounding” of the particulates is indicative of partial dissolution in the aqueous dispersant.

2. Particle Size Analysis

Particle size analysis was carried out using laser diffraction and light microscopy.

30 2.1. Laser Diffraction

Horiba LA-950 V2 with fraction cell was used for laser diffraction studies using the following parameters: dispersion media: 0.05% w/w soy lecithin dissolved in n-hexane; refractive index of media: 1.334; refractive index of acetylsalicylic acid particles: 1.5623; i-value: 0.01. The i-value is an imaginary component that is used by the laser diffraction algorithm to account for the absorption of light by the particles. A stock dispersion of particles in the same media was prepared and added drop-wise to the fraction cell containing a magnetic stirrer bar until the intensity meter showed red laser between 80% and 90%, while blue laser was between 70% and 90%. Once stabilized, the volumetric mean diameter Dv10, Dv50 and Dv90 were measured. This laser diffraction method developed for uncoated particles was not used for spray-dried phospholipid/acetylsalicylic acid particles as they did not disperse well in the selected media.

2.2 Light Microscopy

Photomicrographs of the pre- and post-micronized uncoated particles were taken by dispersing them in a solution of 0.1% w/w docusate sodium in purified water USP, and using a digital imaging light microscope (Olympus BX51 with Clemex ST-2000 controller) at 400-times or 1000-times magnification. As spray-dried phospholipid/acetylsalicylic acid particles were found to not disperse well in the selected media, photomicrographs were taken after spreading them in the dry state over glass slides.

20

3. Jet Milling Trials using Sturtevant Qualification Mill

Initial work was carried out using a Sturtevant Qualification mill with venturi #1 using nitrogen as the carrier gas. Material was fed through vibratory feeder at controlled rate and at predetermined feed and grind pressure. The effect of grinding pressure, feed rate, and second pass were studied on particle size reduction and the conditions are reported in Table 4.

30

Table 4: Jet Milling Trials using Sturtevant Qualification Mill

| Formulation | 3694 | 3695 | 3701 | 3702 |
|---------------|--------|--------|--------|----------------------------|
| Milling run | Pass#1 | Pass#1 | Pass#1 | Pass#2 of Formulation 3695 |
| P_Feed (bar) | 7 | 7 | 7 | 7 |
| P_Grind (bar) | 3.5 | 5 | 3.5 | 5 |
| F_Flow (g/hr) | 17 | 17 | 54 | 7 |

3.1 Effect of Grind Pressure

Formulations 3694 and 3695 were compared to study effect of grind pressure on PSD (particle size distribution). Laser diffraction and microscopy results were obtained are presented in Table 5. Microscopy and laser diffraction data were found to correlate very well. When grinding pressure was increased from 3.5 to 5 bar, a measurable decrease in particle size was observed as would be expected.

10

Table 5: Effect of Grind Pressure on Acetylsalicylic acid Particle Size

| | Average (%RSD)/n=3 | |
|-----------------------|--------------------|-----------|
| Formulation | 3694 | 3695 |
| D _{v10} (μm) | 1.9 (1.0) | 1.2 (2.2) |
| D _{v50} (μm) | 3.3 (1.8) | 2.4 (1.9) |
| D _{v90} (μm) | 5.9 (2.8) | 4.9 (2.6) |
| Microscopy (μm) | 1.7-5.1 | 2.0-3.5 |

3.2 Effect of Feed Rate

Formulations 3694 and 3701 were compared to study the effect of material feed rate on particle size using microscopy (Table 6). Clearly, as the flow rate was increased from 17 g/hr to 54 g/hr, significantly larger particles were obtained. This is likely the result of new material entering the milling chamber and pushing out particles to the collection bag before they undergo sufficient attrition.

20

Table 6: Effect of Feed Rate on Particle Size

| Formulation | 3694 | 3701 |
|---------------------------------|---------|----------|
| Microscopy (μm) | 1.7-5.1 | 5.2-42.1 |

3.3 Effect of Second Milling Pass

In order to achieve the target particle size of D_{v50} of 1.5 μm and D_{v90} of 2 μm , formulation 3695 was passed through mill for a second pass. Particle size analysis was carried out using laser diffraction and microscopy (Table 7). Significant particle size reduction was achieved on the second pass through the jet mill which implies that acetylsalicylic acid particles undergo first order size reduction, and that final particle size obtained depends upon initial particle size used.

10

Table 7: Effect of Second Milling Pass on Particle Size

| Formulation | 3695 | 3702 |
|---------------------------------|---------|---------|
| Microscopy (μm) | 2.0-3.5 | 0.8-2.4 |

4. Jet Milling Using Sturtevant Sanitary Design Mill

In order to achieve higher feed rate with better control as well as to increase batch size, the larger 2" sanitary design mill was used according to parameters listed in Table 8. Material was processed on a second pass as well to reduce particle size to target. Formulations 3727 and 20 3734 were compared with 3705 and 3725 processed using the Qualification mill respectively to study reproducibility in PSD. An antistatic device was necessary to feed the powder for the second pass to minimize the effects of the static electricity imparted during the first pass.

Table 8: Jet Milling using Sturtevant Sanitary Design Mill

| Mill Used | Q-Mill | | 2" Mill | |
|-----------------|--------|---------------------------------|---------|---------------------------------|
| Formulation | 3705 | 3725 | 3727 | 3734 |
| Milling run | Pass#1 | Pass#2 (Formulation 3705) | Pass#1 | Pass#2 (Formulation 3727) |
| P_Venturi (bar) | 4.1 | 2.8 | 4.1 | 2.9 |
| P_Grind (bar) | 2.8 | 2.1 | 2.8 | 2.1 |
| F_Flow (g/hr) | 132 | 78 | 142 | 59 |
| Batch size (g) | 80 | 50 | 200 | 120 |

Aggregated particles with high static charge were obtained in all cases.

4.1 Particle Size Analysis

Particle size analysis of above formulations was carried out using laser diffraction and microscopy (Table 9, Figure 3 and Figure 4). Reproducible results in particle size reduction were obtained with comparable Dv10, Dv50 and Dv90 values between the two mill sizes, even with batch size increased from 80 g to 200 g for the first pass and from 50 g to 120 g for the second pass. Monomodal PSD was obtained for the first pass while a bimodal distribution was observed for the second pass.

Table 9: Particle Size Analysis of Jet Milled Acetylsalicylic acid Formulations Prepared Using Sanitary Design Mill

| Formulation | Average (%RSD)/n=3 | | | |
|-------------|--------------------|------------|------------|------------|
| | 3705 | 3725 | 3727 | 3734 |
| Dv10 (µm) | 0.9 (5.2) | 0.1 (1.9) | 0.8 (10.0) | 0.1 (1.6) |
| Dv50 (µm) | 1.5 (3.3) | 0.4 (6.3) | 1.3 (5.2) | 0.4 (12.9) |
| Dv90 (µm) | 2.6 (4.2) | 1.8 (6.0) | 2.2 (6.7) | 1.5 (5.1) |
| Microscopy | 1.2-2.6 µm | 0.9-1.8 µm | 1.1-3.2 µm | 0.9-2.3 µm |

5. Coating

Spray-drying was used for coating. Jet milled formulation 3734, processed two passes on the 2" sanitary mill, was used further to coat with either DSPC or soy lecithin. Particles were dispersed in n-hexane containing lipid and spray-drying was selected as a method to remove solvent. In order to achieve coating around all individual particles, it was required to disperse Jet milled particles completely without settling, and therefore, continuous stirring was employed throughout the spray-drying operation.

5% w/w DSPC was used as it was found from previous work to mitigate irritation when inhaled. Additionally, soy lecithin was also used in the concentration of 0.1% w/w. As acetylsalicylic acid is insoluble in n-hexane, it was selected as a dispersion media for the micronized particles. Also, it has boiling point of 70°C which is much below the melting point of acetylsalicylic acid (~135°C) and therefore, an inlet temperature of 85°C should remove solvent without affecting the acetylsalicylic acid particles.

A Buchi-290 spray dryer equipped with nozzle of 0.7 mm diameter was used for the study. Spray-drying was performed using nitrogen as the carrier gas with the aspirator set at 100% capacity. The flow rate of nitrogen was adjusted to 1052 L/hr (50 mm in rotameter). Before 10 feeding the stock dispersion, feed rate was adjusted using dispersing media alone to achieve targeted outlet temperature and stabilization of the system.

5.1 Spray-Drying using DSPC

DSPC (Lipoid PC 18:0/18:0) is an endogenous lung phospholipid with a phase transition temperature of 55 °C. On heating at this temperature, DSPC transforms into a liquid crystalline phase from the gel phase, and the phospholipid layer is dispersed in n-hexane as a monolayer with a random and non-rigid structure. When jet milled acetylsalicylic acid particles are dispersed in the DSPC/Hexane solution, a well dispersed colloidal suspension was formed without noticeable settling. From this, it was hypothesized that spray-drying should be able to 20 coat individual acetylsalicylic acid particles on solvent removal. Details of the processing are reported in Table 10.

Table 10: Spray-Drying Parameters for DSPC/Acetylsalicylic acid Formulation

| Formulation | 3739 |
|--------------------------------|------|
| Suspension preparation | |
| DSPC (g) | 0.50 |
| n-hexane (g) | 490 |
| Jet milled ASA (g) | 9.50 |
| %Solid in feed | 2 |
| Suspension temperature (°C) | 55 |
| Spray-drying parameters | |
| Inlet temperature (°C) | 85 |
| Outlet temperature (°C) | 56 |
| Flow rate (g/min) | 3.9 |
| Flow meter (mm) | 50 |
| Suspension temperature (°C) | 55 |
| %Yield | 79 |

No excessive sticking to the spray-drying chamber was observed during processing and a yield of 79% was obtained. Also, the coated particles obtained were observed to be denser and less static than uncoated particles.

5.1.1 Particle Size Analysis

The spray-dried DSPC coated particles were found to not disperse as well in the 0.05% w/w soy lecithin/ n-hexane solution used for particle size analysis of the uncoated acetylsalicylic acid particles. Some agglomeration was observed by microscopy compared to uncoated, though PSD ranges of the primary particles was collated from the microscopic images (Table 11).

Table 11: Particle Size of Micronized Uncoated and Spray-Dried DSPC/Acetylsalicylic acid Particles

| Formulation | 3734 | 3739 |
|-----------------|---------------------|--------------------------|
| Description | Micronized uncoated | Spray-dried DSPC/aspirin |
| Microscopy (μm) | 0.9-2.3 | 1.8-3.6 |

5.1.2 Differential Scanning Calorimetry (DSC)

A DSC study was carried out on raw acetylsalicylic acid, uncoated milled particles of formulation 3734 and spray-dried DSPC/acetylsalicylic acid particles of formulation 3739 to study any change in the crystallinity of the acetylsalicylic acid induced from processing.

Samples were sealed in 40 μ L aluminum pans with pierced lids and analyzed using a differential scanning calorimeter (Mettler-Toledo DSC equipped with STAR® software V10.00). The samples were heated from 25°C to 160°C at a rate of 10°C per minute. An empty pan served as the reference.

In all samples, a sharp endothermic peak corresponding to acetylsalicylic acid melting was observed. No other polymorphic conversion was observed. Also, no significant shift in peak was observed confirming no change in crystallinity of the acetylsalicylic acid on processing (Figure 5, Table 12).

Table 12: DSC Analysis of Raw, Micronized Uncoated and Spray-Dried DSPC/Acetylsalicylic acid

Particles

| Sample | Onset temperature (°C) | Peak temperature (°C) |
|-----------------------------|------------------------|-----------------------|
| Rhodine 3040 US raw | 142.5 | 144.7 |
| Micronized uncoated aspirin | 141.6 | 142.8 |
| Spray-dried DSPC/aspirin | 139.7 | 141.3 |

5.1.3 Thermogravimetric Analysis (TGA)

TGA was carried out for the micronized uncoated acetylsalicylic acid particles of formulation 3734 and spray-dried DSPC/acetylsalicylic acid particles of formulation 3739 to evaluate those for residual solvent and change in moisture content of the particles on spray-drying.

TGA of spray-dried powder carried out in 40 μ L aluminum open pans by heating them from 25°C to 160°C at a rate of 10°C per minute using Mettler-Toledo TGA/DSC1 equipped

with STAR® software V10.00. The % weight loss was measured from 25°C to 120°C and compared between pre- and post-spray-drying.

TGA data suggests that there is no residual hexane in spray-dried particles, as the % weight loss before and after spray drying show similar values. The 0.57% weight loss is likely indicative of the moisture content of pre- and post-spray-dried acetylsalicylic acid particles (Figure 6, Table 13).

Table 13: % Weight Loss for Micronized Uncoated and Spray-Dried DSPC/Acetylsalicylic acid Particles

| Formulation | %weight loss (g) |
|-----------------------------|------------------|
| Micronized uncoated aspirin | 0.58 |
| Spray-dried DSPC/aspirin | 0.57 |

10

5.2 Spray-Drying Acetylsalicylic acid Particles using Soy Lecithin

Soy lecithin was selected as an excipient as it is also approved for inhalation drug delivery and it was able to disperse jet milled acetylsalicylic acid particles well. Therefore, it was expected to be able to coat individual acetylsalicylic acid particle on solvent removal. Soy lecithin was dissolved in n-hexane and jet milled acetylsalicylic acid particles dispersed in it with stirring. However, unlike the dispersion of acetylsalicylic acid in DSPC, the soy lecithin in 0.1% w/w concentration was not able to form colloidal dispersion, and some settling was observed. Therefore, continuous stirring of the feed suspension during spray-drying was used to maintain the dispersion of the acetylsalicylic acid particles. Spray-drying was carried out to remove n-hexane and coat acetylsalicylic acid particles using the parameters in Table 14. A 54% yield was obtained.

20

Table 14: Spray-Drying Parameters for Soy Lecithin/Acetylsalicylic acid Formulation

| Formulation | 3740 |
|--------------------------------|------------|
| Suspension preparation | |
| Soy lecithin (g) | 0.01 |
| n-hexane (g) | 490 |
| Jet milled ASA (g) | 9.99 |
| %Solid in feed | 2 |
| Suspension temperature (°C) | RT |
| Spray-drying parameters | |
| Inlet temperature (°C) | 85 |
| Outlet temperature (°C) | 59 |
| Flow rate (g/min) | 3.9 |
| Flow meter (mm) | 50 |
| Suspension temperature | RT |
| Yield | 54% |

5.2.1 Particle Size Analysis

Particle size analysis was carried out using powder microscopy and compared with micronized uncoated and spray-dried DSPC/acetylsalicylic acid. Particle size of both spray-dried formulations suggest satisfactory results (Table 15).

Table 15: Particle Size Analysis of Spray-dried Soy Lecithin/Acetylsalicylic acid Particles

| Formulation | 3734 | 3739 | 3740 |
|-----------------|-----------------------------|--------------------------|----------------------------------|
| Description | Micronized uncoated aspirin | Spray-dried DSPC/aspirin | Spray-dried soy lecithin/Aspirin |
| Microscopy (μm) | 0.9-2.3 | 1.8-3.6 | 1.7-3.3 |

10

Conclusions

The micronization of acetylsalicylic acid yielded an approximately 70 fold reduction in the starting particle size. Spray-drying with DSPC or soy lecithin resulted in satisfactory particle size for deep lung tissue drug delivery with maximum size of 3.6 μm. Spray-dried DSPC/acetylsalicylic acid particles were found to be less static than soy lecithin/acetylsalicylic acid particles, and even less static than micronized uncoated acetylsalicylic acid particles. The

crystalline structure of acetylsalicylic acid did not change during milling or spray drying as observed by DSC study. DSC studies also suggested absence of any other event such as polymorph conversion during processing. No traces of residual solvent found in spray dried DSPC/acetylsalicylic acid during TGA analysis.

Example 2 Emitted dose analysis of DSPC/acetylsalicylic acid particles and soy lecithin/acetylsalicylic acid particles

DPI devices, e.g. Plastiape, were used to evaluate emitted doses of the DSPC/acetylsalicylic acid particles and soy lecithin/acetylsalicylic acid particles.

10

Table 16

| Powder Type | DPI Device | Flow Rate (slpm) | Fill Weight (mg) | Initial Device Weight (g) | Final Device Weight (g) | Total Emitted Dose (mg) | % Emitted (based on fill weight) |
|-------------------------------------|------------------|-------------------------|------------------|---------------------------|-------------------------|-------------------------|----------------------------------|
| Acetylsalicylic acid (Soy Lecithin) | Plastiape | 100.0 | 20.11 | 10.4966 1 | 10.48124 | 15.37 | 76.4 |
| Acetylsalicylic acid (Soy Lecithin) | Plastiape | 100.0 | 32.81 | 10.5542 1 | 10.53301 | 21.20 | 64.6 |
| Acetylsalicylic acid (DSPC) | Plastiape | (to be provided) | | | | | |

Lactose was used to test proper device setup.

Example 3 Particle Size Distribution (PSD) Analysis of Inhaled Acetylsalicylic acid by Dry Dispersion and Laser Diffraction

Particle size analysis was carried out using laser diffraction analysis of dry dispersed spray-dried DSPC/acetylsalicylic acid particles of formulation 3739 (Table 17), and spray-dried soy lecithin/acetylsalicylic acid particles formulation 3740 (Table 18) (see Example 1 for the preparation of DSPC/acetylsalicylic acid particles and soy lecithin/acetylsalicylic acid particles).

Table 17

| Lens | Primary Pressure (bar) | Replicate | Particle Size (μm) | | | | GSD | Optical Conc. |
|------|------------------------|-----------|--------------------|------|------|------|------|---------------|
| | | | X10 | X50 | X90 | VMD | | |
| R3 | 1.0 | 1 | 0.96 | 2.29 | 4.47 | 2.56 | 1.82 | 8.71 |
| R2 | 1.0 | 1 | 0.83 | 2.31 | 4.49 | 2.56 | 1.90 | 7.56 |
| | | 2 | 0.79 | 2.24 | 4.44 | 2.49 | 1.93 | 7.10 |
| R1 | 0.7 | 1 | 0.66 | 2.63 | 5.27 | 2.88 | 2.07 | 9.21 |
| | | 2 | 0.64 | 2.59 | 5.18 | 2.84 | 2.07 | 5.72 |
| | 0.9 | 1 | 0.62 | 2.34 | 4.63 | 2.57 | 2.01 | 4.78 |
| | | 2 | 0.57 | 2.31 | 4.68 | 2.55 | 2.09 | 5.98 |
| | 1.0 | 1 | 0.58 | 2.31 | 4.69 | 2.54 | 2.11 | 10.98 |
| | | 2 | 0.60 | 2.34 | 4.69 | 2.56 | 2.08 | 6.53 |
| | | 3 | 0.56 | 2.31 | 4.76 | 2.57 | 2.14 | 6.74 |
| | | 4 | 0.57 | 2.26 | 4.51 | 2.48 | 2.07 | 7.97 |
| | | 5 | 0.58 | 2.28 | 4.53 | 2.49 | 2.06 | 8.20 |
| | 1.2 | 1 | 0.56 | 2.13 | 4.17 | 2.32 | 2.03 | 4.38 |
| | | 2 | 0.55 | 2.12 | 4.17 | 2.32 | 2.05 | 12.60 |
| | 2.0 | 1 | 0.60 | 2.03 | 4.03 | 2.54 | 1.97 | 6.03 |
| | | 2 | 0.54 | 2.03 | 4.25 | 2.57 | 2.12 | 7.63 |
| | 3.0 | 1 | 0.55 | 1.84 | 3.68 | 2.13 | 2.03 | 8.88 |
| | | 2 | 0.52 | 1.81 | 3.63 | 2.01 | 2.07 | 8.49 |
| | 4.0 | 1 | 0.47 | 1.79 | 3.64 | 2.00 | 2.12 | 6.41 |
| | | 2 | 0.52 | 1.80 | 3.58 | 2.00 | 2.03 | 7.56 |

Table 18

| Lens | Primary Pressure (bar) | Replicate | Particle Size (μm) | | | | GSD | Optical Conc. |
|------|------------------------|-----------|--------------------|------|------|------|------|---------------|
| | | | X10 | X50 | X90 | VMD | | |
| R1 | 1.0 | 1 | 0.50 | 1.91 | 3.90 | 2.12 | 2.11 | 11.28 |
| | | 2 | 0.50 | 1.89 | 3.73 | 2.07 | 2.03 | 3.89 |
| | | 3 | 0.49 | 1.90 | 3.83 | 2.11 | 2.09 | 9.58 |
| | | 4 | 0.52 | 1.90 | 3.66 | 2.06 | 2.00 | 4.70 |
| | | 5 | 0.49 | 1.90 | 3.83 | 2.10 | 2.08 | 6.84 |
| | | Average | 0.50 | 1.90 | 3.79 | 2.09 | 2.06 | |
| | | | 2 | 0 | 2 | 1 | 2 | |

RSD: relative standard deviation.

Example 4 NGI (next-generation impactor) Analysis of Spray Dried Acetylsalicylic acid/DPSC Particles

The dry powders of Example 1 were evaluated for aerodynamic performance. The DPI device used was a monodose inhaler. The NGI test conditions ranged between 20°C and 25°C, and between 40% and 50% RH (relative humidity) (Table 19).

Table 19

| | NGI 1 | NGI 2 | NGI 3 | NGI 4 | NGI 5 |
|----------------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| Controlled condition | 21.83C/46.7 %RH | 22.66C/47.3% RH | 21.93C/46.9% RH | 21.93C/46.9% RH | 21.99C/43.1% RH |
| Measured Flow | 99.1 SLPM | 98.4 SLPM | 97.6 SLPM | 100.0 SLPM | 100.5 SLPM |

Table 20 shows the aerodynamic properties of DSPC/acetylsalicylic acid particles.

Table 20

| | 2 capsules | | 1 capsule | | |
|--|------------|---------|-----------|--------|--------|
| | NGI 1 | NGI 2 | NGI 3 | NGI 4 | NGI 5 |
| Device, μg | 7876.4 | 9010.6 | 4267.0 | 4118.0 | 5115.8 |
| Capsule 1, μg | 653.6 | 717.9 | 484.6 | 464.5 | 670.5 |
| Capsule 2, μg | 616.2 | 560.3 | NA | NA | NA |
| Induction Port, μg | 11611.6 | 14550.8 | 7253.0 | 7454.4 | 6792.6 |
| Stage 1, μg | 10232.0 | 9393.6 | 3704.8 | 4257.6 | 5481.2 |
| Stage 2, μg | 17402.0 | 16198.0 | 8284.4 | 8136.4 | 8758.4 |
| Stage 3, μg | 10882.4 | 9993.6 | 5600.8 | 4976.4 | 5087.6 |
| Stage 4, μg | 4884.0 | 4864.4 | 2791.2 | 2387.2 | 2273.6 |
| Stage 5, μg | 1670.0 | 1514.8 | 983.2 | 757.6 | 891.2 |
| Stage 6, μg | 983.8 | 1076.6 | 619.8 | 471.9 | 530.3 |
| Stage 7, μg | 575.6 | 498.2 | 318.9 | 262.9 | 284.1 |
| MOC, μg | 320.8 | 292.4 | 134.0 | 158.0 | 201.7 |
| Nozzles, μg | 5364.8 | 6363.2 | 2546.4 | 2833.6 | 3280.8 |
| Nominal loaded mass (mg) | 74 | 74 | 37 | 37 | 37 |
| ED (mg) | 63.93 | 64.75 | 32.24 | 31.70 | 33.58 |
| Nominal % ED (mg) | 86% | 88% | 87% | 86% | 91% |
| FPD (mg) | 32.2 | 30.8 | 16.6 | 15.1 | 16.0 |
| FPF (%) | 50.4 | 47.5 | 51.5 | 47.7 | 47.5 |
| MMAD (μm) | 3.94 | 3.93 | 3.62 | 3.91 | 4.12 |
| GSD | 1.91 | 1.94 | 1.91 | 1.94 | 2.00 |
| Recovery (%) | 99.8 | 100.0 | 101.2 | 100.1 | 103.7 |

Example 5 NGI Analysis of Spray Dried Acetylsalicylic acid/Soy Lecithin Particles

The dry powders of Example 1 were evaluated for aerodynamic performance. The DPI device used was a monodose inhaler. The NGI test conditions ranged between 20°C and 25°C, and between 40% and 50% RH (relative humidity) (Table 21).

Table 21

| | NGI 1 | NGI 2 | NGI 3 | NGI 4 | NGI 5 |
|----------------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| Controlled condition | 22.57C/49.6 %RH | 22.16C/48.7% RH | 22.14C/47.9% RH | 21.76C/45.1% RH | 21.66C/45.1% RH |
| Measured Flow | 98.7 SLPM | 97.6 SLPM | 99.0 SLPM | 100.0 SLPM | 97.5 SLPM |

10 Table 22 shows the aerodynamic properties of soy lecithin/acetylsalicylic acid particles.

Table 22

| | 2 capsules | | 1 capsule | | |
|--------------------------|------------|---------|-----------|--------|--------|
| | NGI 1 | NGI 2 | NGI 3 | NGI 4 | NGI 5 |
| Device, μ g | 13139.2 | 15032.8 | 7664.0 | 6554.6 | 8382.0 |
| Capsule 1, μ g | 1259.1 | 1607.1 | 1595.1 | 1078.2 | 916.0 |
| Capsule 2, μ g | 2893.7 | 1050.2 | NA | NA | NA |
| Induction Port, μ g | 5834.4 | 5586.6 | 3008.0 | 3604.4 | 3795.8 |
| Stage 1, μ g | 4378.4 | 5104.0 | 1962.0 | 2274.8 | 2266.0 |
| Stage 2, μ g | 12060.0 | 12890.8 | 5726.0 | 6028.0 | 6028.0 |
| Stage 3, μ g | 15818.4 | 16041.6 | 7544.0 | 7687.2 | 7712.0 |
| Stage 4, μ g | 11276.8 | 11301.6 | 5556.8 | 5345.6 | 5485.6 |
| Stage 5, μ g | 3305.2 | 3182.0 | 1692.4 | 1622.4 | 1694.4 |
| Stage 6, μ g | 1272.6 | 1161.2 | 749.5 | 728.2 | 658.7 |
| Stage 7, μ g | 708.4 | 605.2 | 436.4 | 414.9 | 366.2 |
| MOC, μ g | 340.8 | 375.8 | 231.8 | 228.6 | 236.9 |
| Nozzles, μ g | 4105.6 | 4928.0 | 1812.8 | 2306.4 | 2121.6 |
| <hr/> | | | | | |
| Nominal loaded mass (mg) | 74 | 74 | 37 | 37 | 37 |
| ED (mg) | 59.10 | 61.18 | 28.72 | 30.24 | 30.37 |
| Nominal % ED (mg) | 80% | 83% | 78% | 82% | 82% |
| FPD (mg) | 42.7 | 43.7 | 20.9 | 21.2 | 21.2 |

| | | | | | |
|---------------------|-------|-------|-------|-------|-------|
| FPP (%) | 72.3 | 71.5 | 72.7 | 70.0 | 69.7 |
| MMAD (μm) | 2.71 | 2.79 | 2.65 | 2.72 | 2.72 |
| GSD | 1.72 | 1.73 | 1.75 | 1.75 | 1.75 |
| Recovery (%) | 104.6 | 104.0 | 104.4 | 103.1 | 104.5 |

HPLC analysis of acetylsalicylic acid in NGI and Delivered Dose samples was carried out as follows.

Equipment

The HPLC column was Phenomenex Luna C18(2) 5μm, 4.6 x 100mm. Shimadzu HPLC Equipment was used, including Shimadzu SIL-HTC Autosampler, Shimadzu CTO-10ASVP Column Oven, Shimadzu LC-10ADVP Binary HPLC Pump, Shimadzu DGU-14A Inline Degasser, Shimadzu UV Detector, and Computer with Shimadzu Class VP software.

10 Materials

Mobile Phase A was 69:28:3 Water : Methanol : Glacial Acetic Acid. Mobile Phase B was 97:3 Methanol : Glacial Acetic Acid. Diluent was 95:5 Methanol : Glacial Acetic Acid. Needlewash was 50:50 Water : Methanol. The working standard was 750 μg/mL acetylsalicylic acid (working standard A “WSA” and working standard B “WSB”).

HPLC Conditions and Analysis

Flow rate was 2.0 mL/min. The sample injection volume was 10 μL. The gradient was run according to the timing scheme in Table 23.

Table 23 HPLC Gradient Program

| Time (min) | %B |
|------------|-------|
| 0.00 | 0.0 |
| 3.80 | 0.0 |
| 3.81 | 100.0 |
| 5.80 | 100.0 |
| 5.81 | 0.0 |
| 8.00 | STOP |

20

The analysis of the samples was in the following sequence:

| | |
|-----------------------|--------------------|
| A. Blank | (2 injections) |
| B. Working Standard A | (6 injections) |
| C. Working Standard B | (2 injections) |
| D. Blank | (1 injection) |
| E. Sample | (1 injection each) |
| F. WSB (QC Standard) | (1 injection) |

Repeat steps E-F as necessary ensuring that the last injection of a sequence is a QC standard.

10 The standard agreement between WSA and WSB must be within 97.0 - 103.0%. The QC standard agreement between the ongoing standard analysis and initial analysis (n=2) for WSB must between 97.0-103.0%.

The standard agreement between WSA and WSB was calculated according to the equation below.

$$SA = \frac{A_{WSA}}{A_{WSB}} \times \frac{C_{WSB}}{C_{WSA}} \times 100$$

Where:

| | | |
|------------------|---|---------------------------------------|
| SA | = | Standard Agreement (%) |
| A _{WSA} | = | WSA Average Area (n=6) |
| A _{WSB} | = | WSB Average Area (n=2) |
| C _{WSA} | = | WSA Theoretical Concentration (µg/mL) |
| C _{WSB} | = | WSB Theoretical Concentration (µg/mL) |
| 100 | = | Conversion to % |

The % recovery of the QC standard(s) was calculated according to the equation below.

$$QC = \frac{A_{QC}}{A_{WSB}} \times 100$$

Where:

| | | |
|------------------|---|--------------------------------|
| QC | = | QC % Recovery |
| A _{QC} | = | QC Area |
| A _{WSB} | = | Initial WSB Average Area (n=2) |
| 100 | = | Conversion to % |

The concentration of samples was calculated according to the equation below.

$$C_{SX} = \frac{A_{SX}}{A_{WSA}} \times C_{WSA}$$

Where:

| | | |
|-----------|---|--|
| C_{SX} | = | Sample Concentration ($\mu\text{g/mL}$) |
| A_{SX} | = | Sample Area |
| A_{WSA} | = | WSA Average Area (n=6) Area |
| C_{WSA} | = | Theoretical WSA Concentration ($\mu\text{g/mL}$) |

Example 6 Spray Pattern and Plume Geometry

Spray pattern and plume geometry characterization of spray pattern and plume geometry of the formulation will be evaluated using standard methodology (see, 10 <http://www.proveris.com/products/sprayview/>; see also, <http://www.oxfordlasers.com/imaging/spray-pattern-plume-geometry-measurement/>).

Various factors can affect the spray pattern and plume geometry, including the size and shape of the actuator orifice, the design of the actuator, the size of the metering chamber, the size of the stem orifice of the valve, the vapor pressure in the container, and the nature of the formulation. Spray pattern testing will be tested on all formulations under a variety of different temperature and humidity conditions.

Example 7 – Dry Powder Stability Testing

20 The following test parameters will be analyzed for all dry powder formulations.

i. Appearance and Color

The appearance of the content of the container and the appearance of the container and closure system (i.e., the valve and its components and the inside of the container) will be analyzed. To determine whether there is any color is associated with the formulation (either present initially or from degradative processes occurring during shelf life).

ii. Microbial Limits

30 The microbial quality will be controlled for the total aerobic count, total yeast and mold count, and freedom from designated indicator pathogens. Appropriate testing will be done to show that the drug product does not support the growth of microorganisms and that microbial quality is maintained throughout the expiration period.

iii. Water or Moisture Content

Water or moisture content will be analyzed. Changes in particle size distribution,

morphic form, and other changes such as crystal growth or aggregation will be evaluated.

iv. Dose Content Uniformity

Dose content uniformity will be evaluated across multiple batches, formulations and under stability testing condition. The amount of active ingredient per determination will be evaluated to show that the emitted dose is 80–120 percent of label claim for more than one of ten containers. The dose content uniformity over container life will also be evaluated.

v. Particle Size Distribution

Particle size distribution will be evaluated across all batches, formulations as well as under stability testing conditions. For particular formulations, the total mass of drug collected on 10 all stages and accessories will be shown to lie between 85 and 115 percent of label claim on a per actuation basis.

vi. Stability studies will be performed on all batches as well as each formulation. The test storage conditions in the stability protocol for a drug product intended for storage under controlled room temperature conditions will include (1) accelerated ($40\pm2^\circ\text{C}/75\pm5\%$ Relative Humidity (RH)), (2) intermediate ($30\pm2^\circ\text{C}/60\pm5\%$ RH), if applicable, and (3) long-term ($25\pm2^\circ\text{C}/60\pm5\%$ RH) conditions.

The foregoing description is provided to enable a person skilled in the art to practice the various configurations described herein. While the subject technology has been particularly 20 described with reference to the various figures and configurations, it should be understood that these are for illustration purposes only and should not be taken as limiting the scope of the subject technology.

There may be many other ways to implement the subject technology. Various functions and elements described herein may be partitioned differently from those shown without departing from the scope of the subject technology. Various modifications to these configurations will be readily apparent to those skilled in the art, and generic principles defined herein may be applied to other configurations. Thus, many changes and modifications may be made to the subject technology, by one having ordinary skill in the art, without departing from the scope of the subject technology.

30 It is understood that the specific order or hierarchy of steps in the processes disclosed is an illustration of exemplary approaches. Based upon design preferences, it is understood that the

specific order or hierarchy of steps in the processes may be rearranged. Some of the steps may be performed simultaneously. The accompanying method claims present elements of the various steps in a sample order, and are not meant to be limited to the specific order or hierarchy presented.

It is to be understood that, while the subject technology has been described in conjunction with the detailed description, thereof, the foregoing description is intended to illustrate and not limit the scope of the subject technology. The citation of any references herein is not an admission that such references are prior art to the present invention.

Those skilled in the art will recognize, or be able to ascertain using no more than routine experimentation, many equivalents to the specific embodiments of the invention described herein. Such equivalents are intended to be encompassed by the following embodiments.

CLAIMS

1. A dry powder composition comprising dry particles that comprise acetylsalicylic acid or a pharmaceutically acceptable salt thereof, in an amount greater than 30% (w/w) of the composition, wherein said dry particles have a mass median aerodynamic diameter (MMAD) within a range of about 0.5 μm to about 10 μm , wherein the composition further comprises one or more phospholipids in an amount ranging from about 0.1% (w/w) to about 10% (w/w) of the composition.

2. The dry powder composition of claim 1, wherein the composition comprises particles having an MMAD size distribution such that said particles exhibit a DV90 less than about 20 μm , a DV50 less than about 7 μm , and a DV10 less than about 2 μm .

3. The dry powder composition of claim 1, wherein the composition comprises particles having an MMAD size distribution such that said particles exhibit a DV90 less than about 10 μm , a DV50 less than about 4 μm , and a DV10 less than about 1 μm .

4. The dry powder composition of claim 1, wherein the composition comprises particles having an MMAD size distribution such that said particles exhibit a DV90 less than about 6 μm , a DV50 less than about 3 μm , and a DV10 less than about 1 μm .

20

5. The dry powder composition of claim 1, wherein the composition comprises particles having an MMAD size distribution such that said particles exhibit a DV50 less than about 5 μm , and a DV10 less than about 2 μm .

6. The dry powder composition of claim 1, wherein the composition comprises particles having an MMAD size distribution such that said particles exhibit a DV50 ranging from about 2.5 μm to about 4 μm , and a DV10 ranging from about 0.8 μm to about 1.5 μm .

30

7. The dry powder composition of claim 1, wherein acetylsalicylic acid or a pharmaceutically acceptable salt thereof is in an amount greater than 40% (w/w) of the composition.

8. The dry powder composition of claim 1, wherein acetylsalicylic acid or a pharmaceutically acceptable salt thereof is in an amount greater than 50% (w/w) of the composition.

9. The dry powder composition of any one of the claims 1-8, wherein the composition comprises particles coated with a pharmaceutically acceptable excipient.

10. The dry powder composition of claim 9, wherein the pharmaceutically acceptable excipient is a phospholipid having surfactant properties.

10

11. The dry powder composition of claim 9, wherein the pharmaceutically acceptable excipient comprises at least one of dipalmitoyl phosphatidylcholine (DPPC), distearoyl phosphatidylcholine (DSPC) or soy lecithin, in an amount ranging from about 0.1% to about 10% (w/w) of the composition.

12. The dry powder composition of claim 9, wherein the pharmaceutically acceptable excipient comprises at least one of dipalmitoyl phosphatidylcholine, distearoyl phosphatidylcholine or soy lecithin, in an amount ranging from about 1% to about 5% w/w (w/w) of the composition.

20

13. The dry powder composition of claim 11, wherein the composition comprises DSPC in an amount of about 5% (w/w) of the composition.

14. The dry powder composition of claim 11, wherein the composition comprises soy lecithin in an amount of about 0.1% (w/w) of the composition.

15. A drug delivery system effective to reduce the risk of a thromboembolic event or treat thrombosis, wherein the system comprises the dry powder composition of any one of claims 1-14, and wherein acetylsalicylic acid is present at a dose ranging from about 5 mg to about 40 mg.

30

16. The drug delivery system of claim 15, further comprising clopidogrel.

17. The drug delivery system of claim 15, wherein the dry powder composition has an emitted dose ranging from about 75% to about 95%.

18. The drug delivery system of claim 15, comprising particles coated with a pharmaceutically acceptable excipient comprising at least one of dipalmitoyl phosphatidylcholine (DPPC), distearoyl phosphatidylcholine (DSPC) or soy lecithin, in an amount ranging from about 0.1% to about 10% (w/w) of the composition.

10 19. The drug delivery system of claim 18, wherein the pharmaceutically acceptable excipient is DSPC.

20. The drug delivery system of claim 15, wherein the composition substantially comprises dry particles having a MMAD ranging from about 3 to about 4 μm .

21. The drug delivery system of claim 15, wherein the emitted dose is greater than about 90%.

22. The drug delivery system of claim 20, wherein mass percent of stages in an NGI testing apparatus of the respirable powder yields are at, stage 1 about 10% to about 13%, stage 2, about 20% to about 23%, stage 3, about 13% to about 15%, and stage 4, about 5% to about 6% and fine particle fraction ranges from about 45% to about 55%.

23. The drug delivery system of claim 18, wherein the pharmaceutically acceptable excipient is soy lecithin.

24. The drug delivery system of claim 15, wherein the composition substantially comprises dry particles having a MMAD ranging from about 2.0 to about 5.0 μm .

25. The drug delivery system of claim 24, wherein the emitted dose ranges from about 70%
30 to about 85%.

26. The drug delivery system of claim 25, wherein mass percent of stages in an NGI testing apparatus of the respirable powder yields are at, stage 1 about 5% to about 10%, stage 2, about 10% to about 18%, stage 3, about 15% to about 20%, and stage 4, about 10% to about 15% and fine particle fraction ranges from about 50% to about 70%.

27. The drug delivery system of claim 15, further comprising an excipient.

28. The drug delivery system of claim 27, wherein the excipient is SLS, lactose, starch, cellulose, leucine, sodium citrate, maltodextrin and/or mannitol.

10 29. A method of treating an ischemic event or reducing the risk of a thromboembolic event or treat thrombosis comprising, administrating to a subject in need thereof a therapeutically effective dose of a dry powder composition comprising dry particles that comprise acetylsalicylic acid, or a pharmaceutically acceptable salt thereof, wherein said dry powder comprises substantially dry particles having a mass median aerodynamic diameter (MMAD) within a range of about 0.5 μ m to about 10 μ m, wherein the dry powder further comprises one or more phospholipids in an amount ranging from about 0.1% (w/w) to about 10% (w/w) of the composition.

20 30. The method of claim 29, wherein the thromboembolic event is a myocardial infarction.

31. The method of claim 29, wherein the thromboembolic event is a transient ischemic event.

32. The method of claim 29, wherein the thromboembolic event is a stroke.

33. The method of claim 29, wherein the thromboembolic event is treated within about 5 minutes of onset of the ischemic event.

34. The method of claim 29, wherein the thromboembolic event is treated within about 10 minutes of onset of the ischemic event.

35. The method of claim 29, wherein the thromboembolic event is treated within about 15 minutes of onset of the ischemic event.

36. The method of claim 29, wherein the phospholipid is DSPC.

37. The method of claim 36, wherein the composition substantially comprises dry particles having a MMAD ranging from about 3 to about 4 μm .

38. The method of claim 37, wherein the emitted dose is greater than about 90%.

10

39. The method of claim 38, wherein mass percent of stages in an NGI testing apparatus of the respirable powder yields are at, stage 1 about 10% to about 13%, stage 2, about 20% to about 23%, stage 3, about 13% to about 15%, and stage 4, about 5% to about 6% and fine particle fraction ranges from about 45% to about 55%.

40. The method of claim 29, wherein the phospholipid is soy lecithin.

41. The method of claim 40, wherein the composition substantially comprises dry particles having a MMAD ranging from about 2.0 to about 3.0 μm .

20

42. The method of claim 41, wherein the emitted dose ranges from about 70% to about 80%.

43. The method of claim 42, wherein mass percent of stages in an NGI testing apparatus of the respirable powder yields are at, stage 1 about 5% to about 10%, stage 2, about 10% to about 18%, stage 3, about 15% to about 20%, and stage 4, about 10% to about 15% and fine particle fraction ranges from about 50% to about 70%.

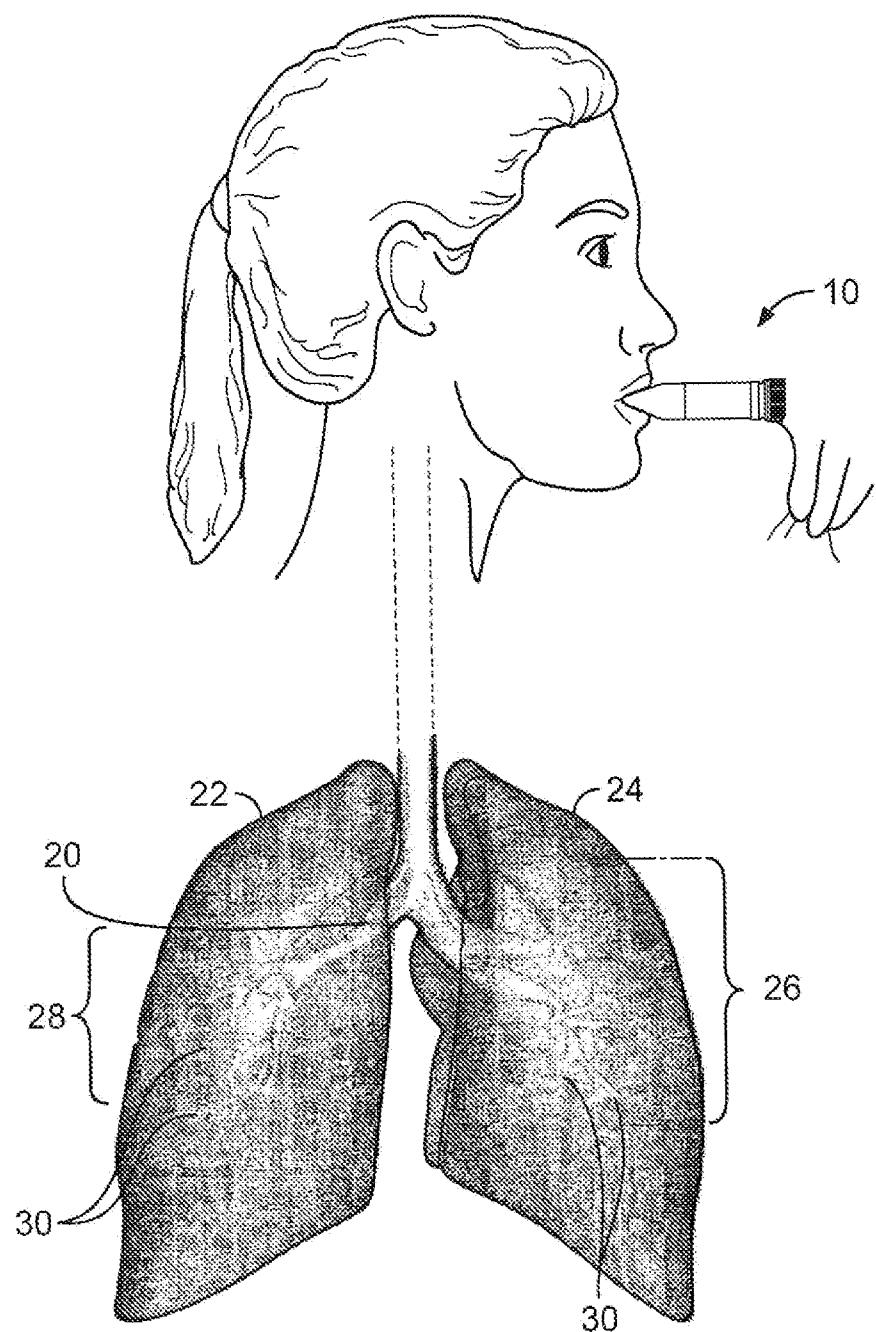
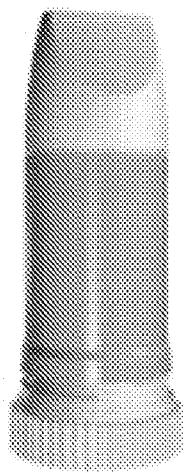


FIG. 1

Figure 2



| Data Name | Graph Type | Sample Name | Median Size | D10 | D90 |
|----------------------|------------|-------------------------------|-------------|-------------|-----|
| GNL 1177 FID#3727 n1 | □—□ | GNL 1177 FID#3727 1.22522(μm) | 0.69581(μm) | 2.03158(μm) | |
| GNL 1177 FID#3727 n2 | —□—□ | GNL 1177 FID#3727 1.35906(μm) | 0.82782(μm) | 2.31122(μm) | |
| GNL 1177 FID#3727 n3 | □—□—□ | GNL 1177 FID#3727 1.29054(μm) | 0.83492(μm) | 2.10566(μm) | |

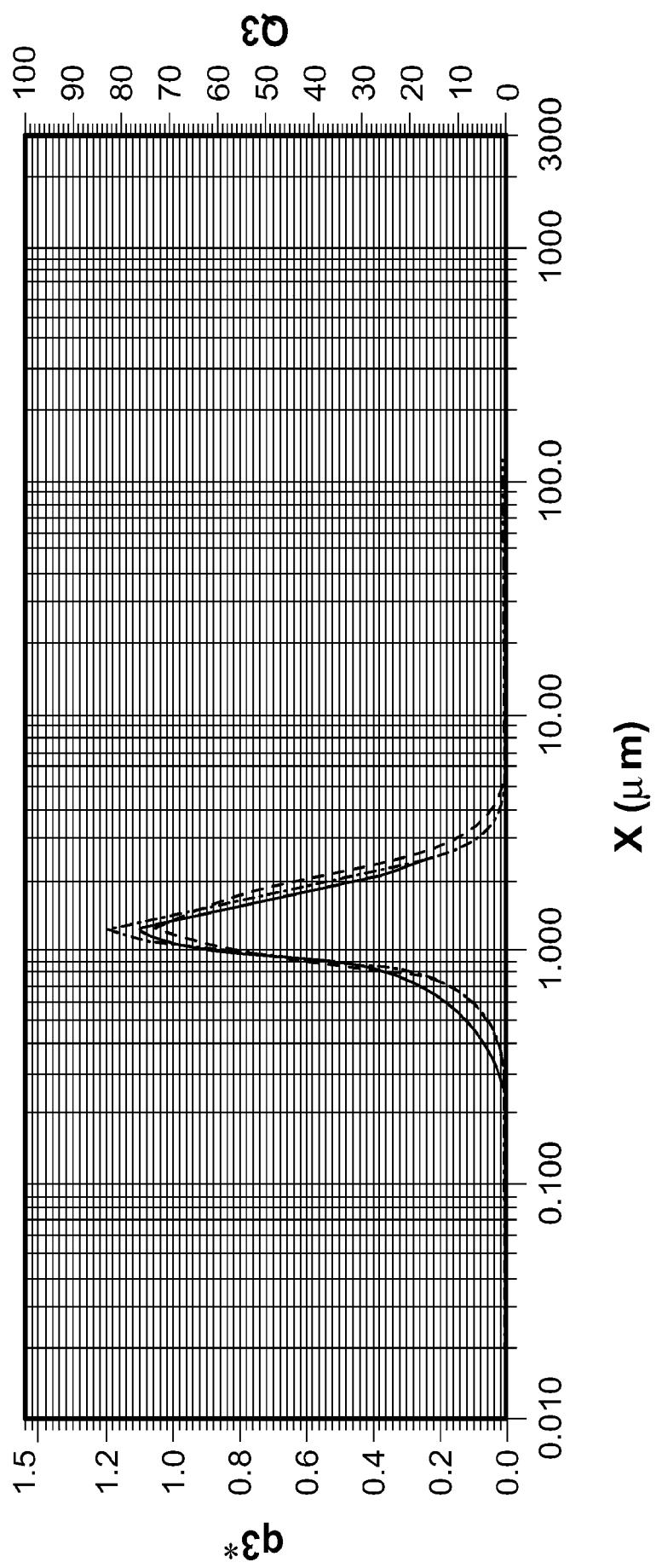


FIG. 3

| Data Name | Graph Type | Sample Name | Median Size | D10 | D90 |
|----------------------|------------|-------------------------------|-------------|-------------|-------------|
| GNL 1177 FID#3734 n3 | ----- | GNL 1177 FID#3734 0.37630(μm) | 0.37630(μm) | 0.12112(μm) | 1.57581(μm) |
| GNL 1177 FID#3734 n2 | ---- | GNL 1177 FID#3734 0.48479(μm) | 0.48479(μm) | 0.12289(μm) | 1.52823(μm) |
| GNL 1177 FID#3734 n1 | --- | GNL 1177 FID#3734 0.41551(μm) | 0.41551(μm) | 0.12513(μm) | 1.42528(μm) |

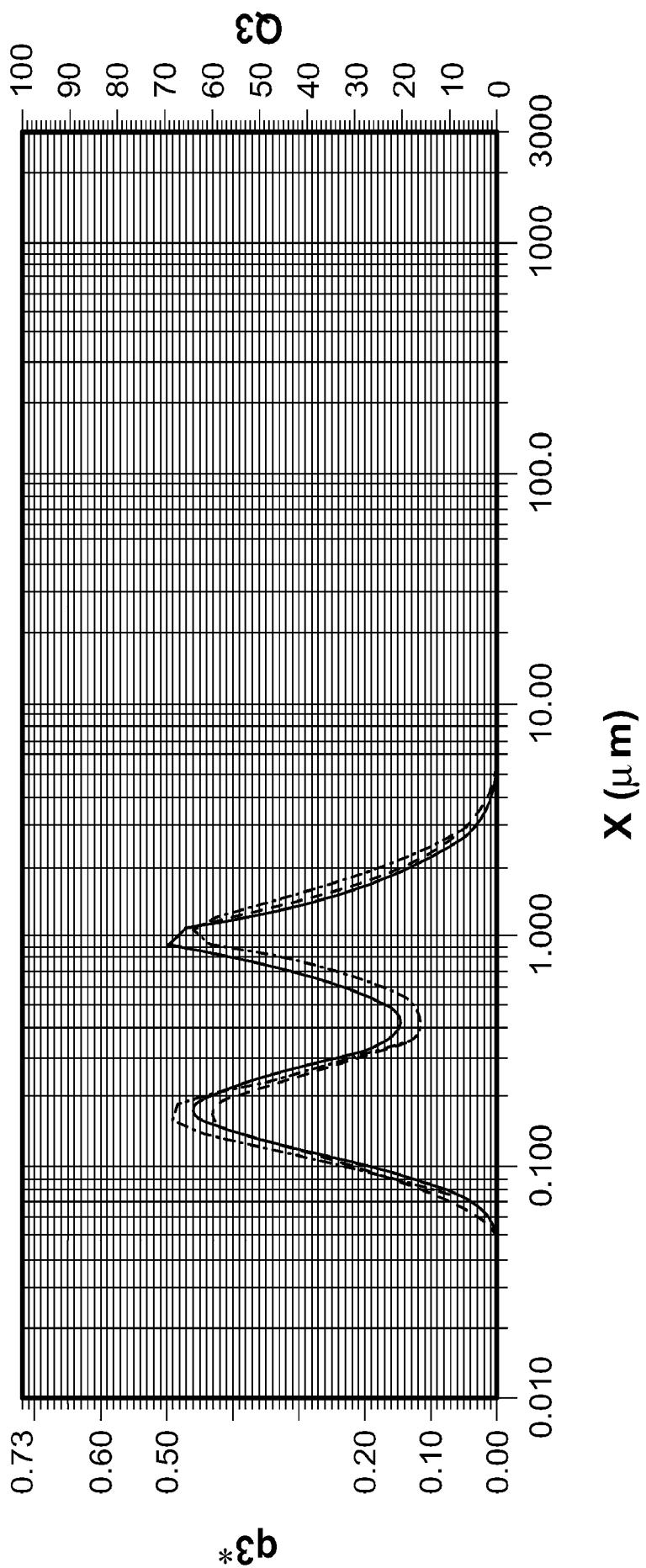
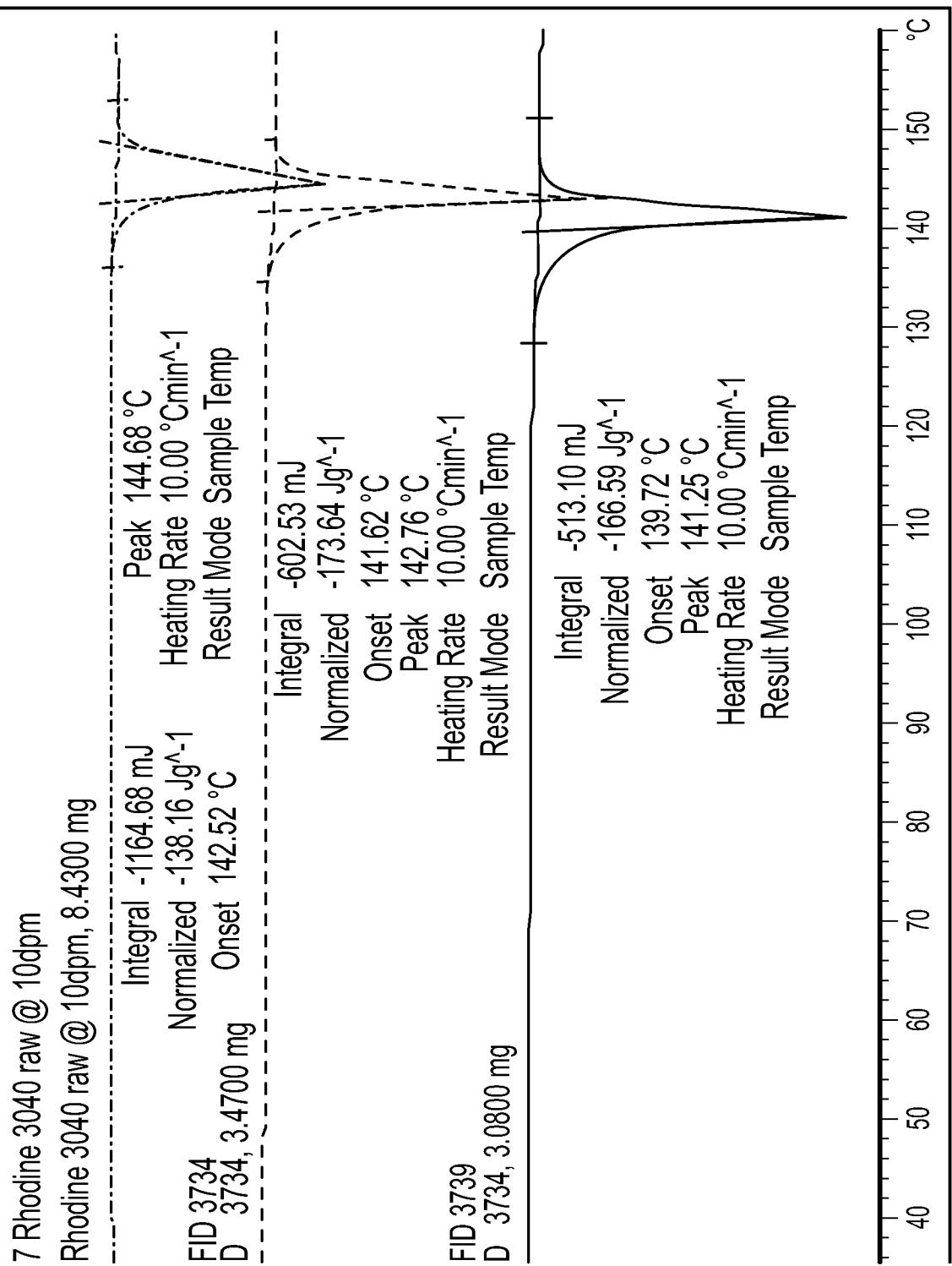


FIG. 4

03.09.2014 12:15:42

GNL1177 FD#3734 VS 3739



ances Inc. : METTLER

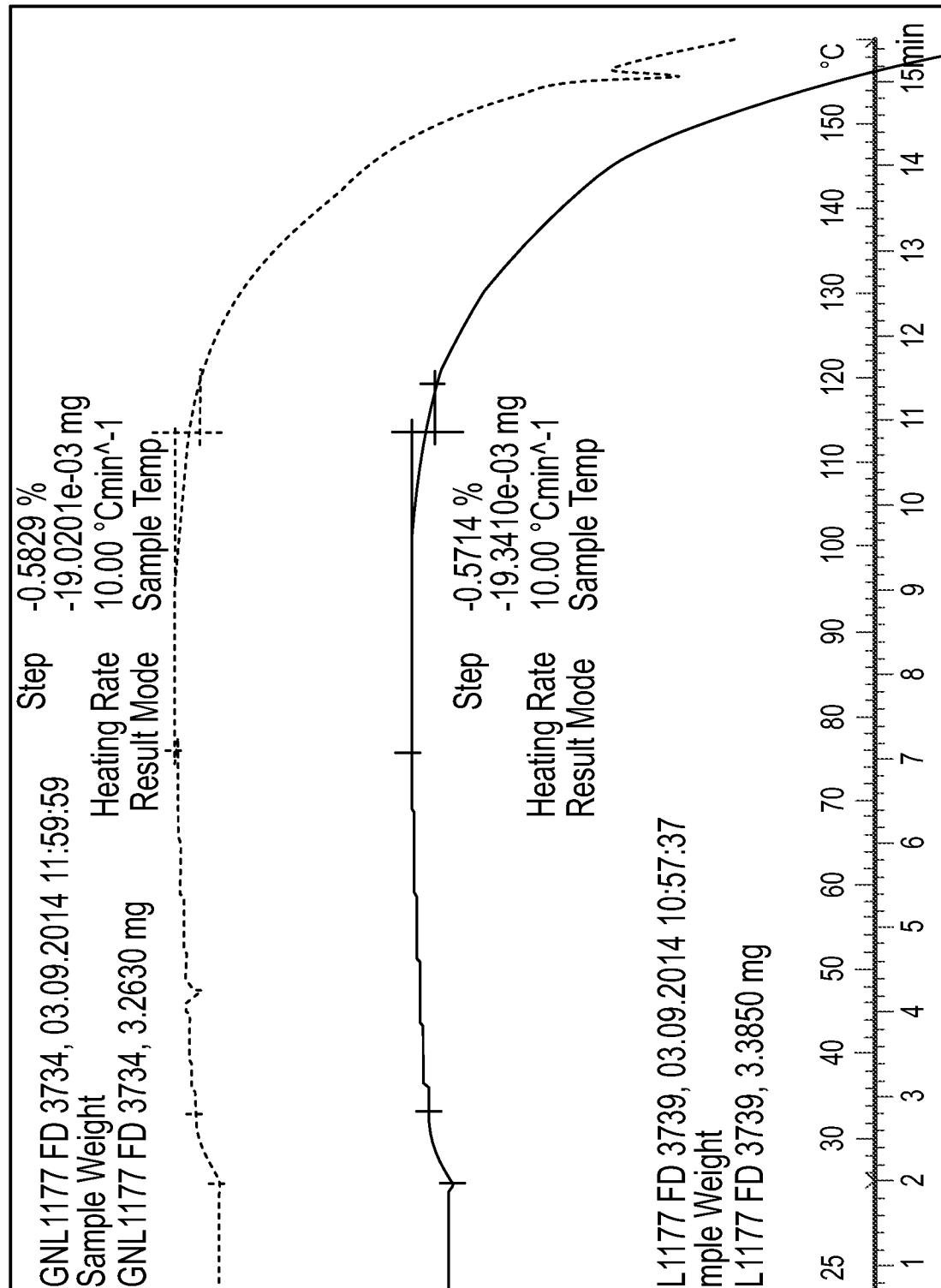
Not signed

STAR SW 10.00

FIG. 5

TGA FID#3734 Vs 3739

04.09.2014 10:40:23

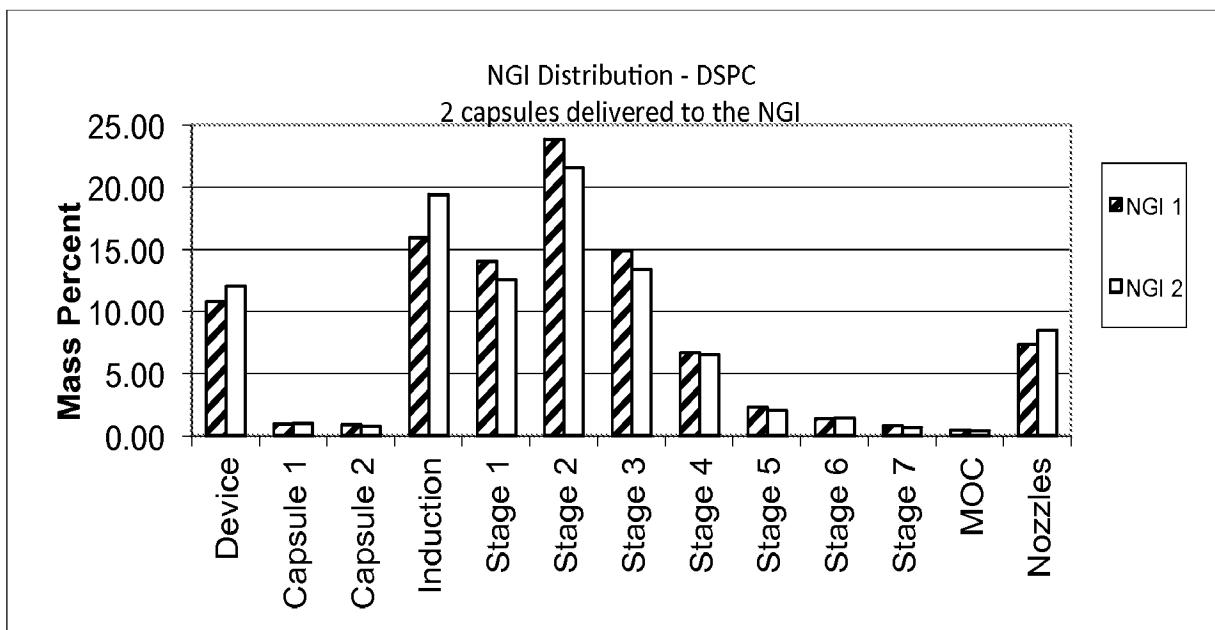


ences Inc.: METTLER

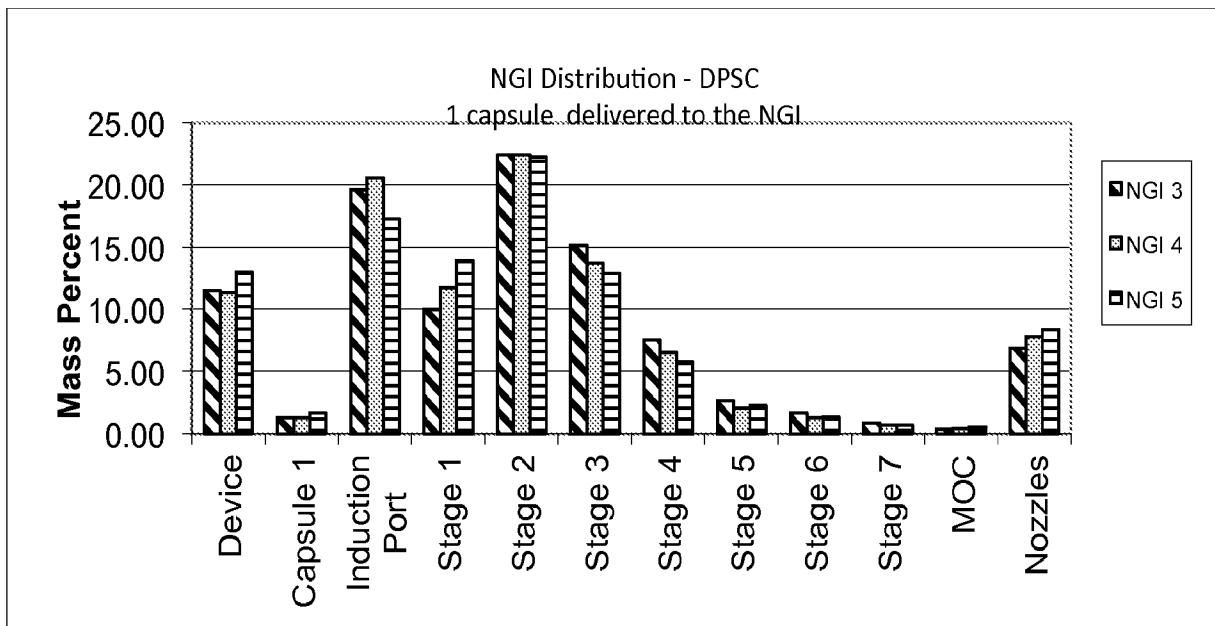
Not signed

Star SW 10.00

FIG. 6

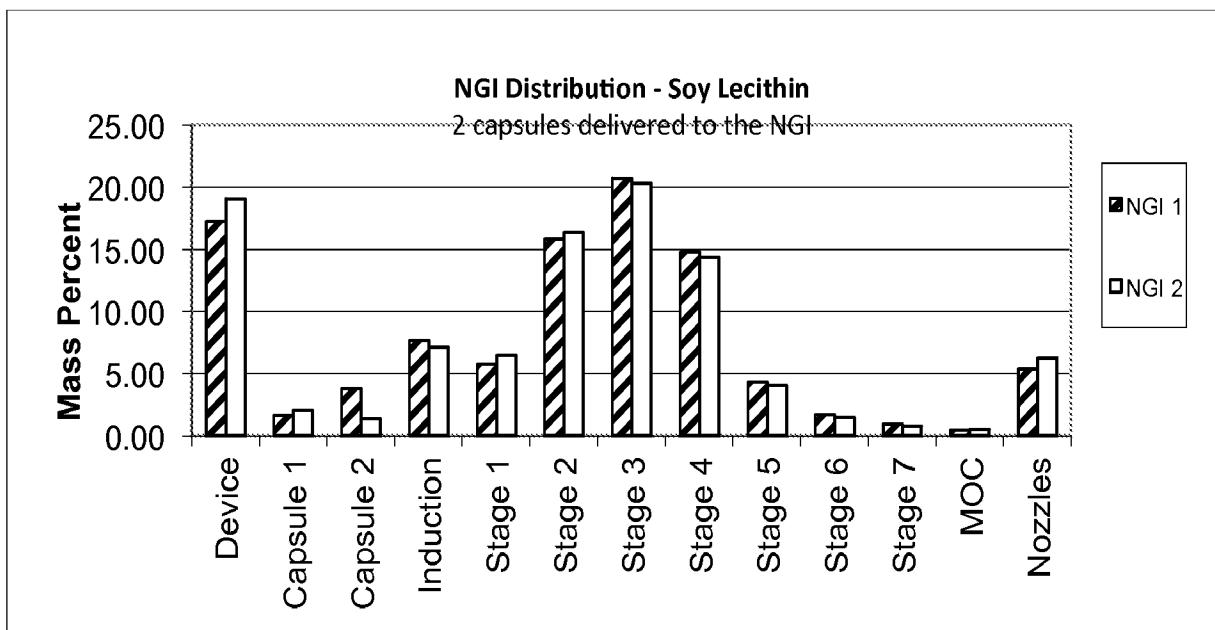


A.

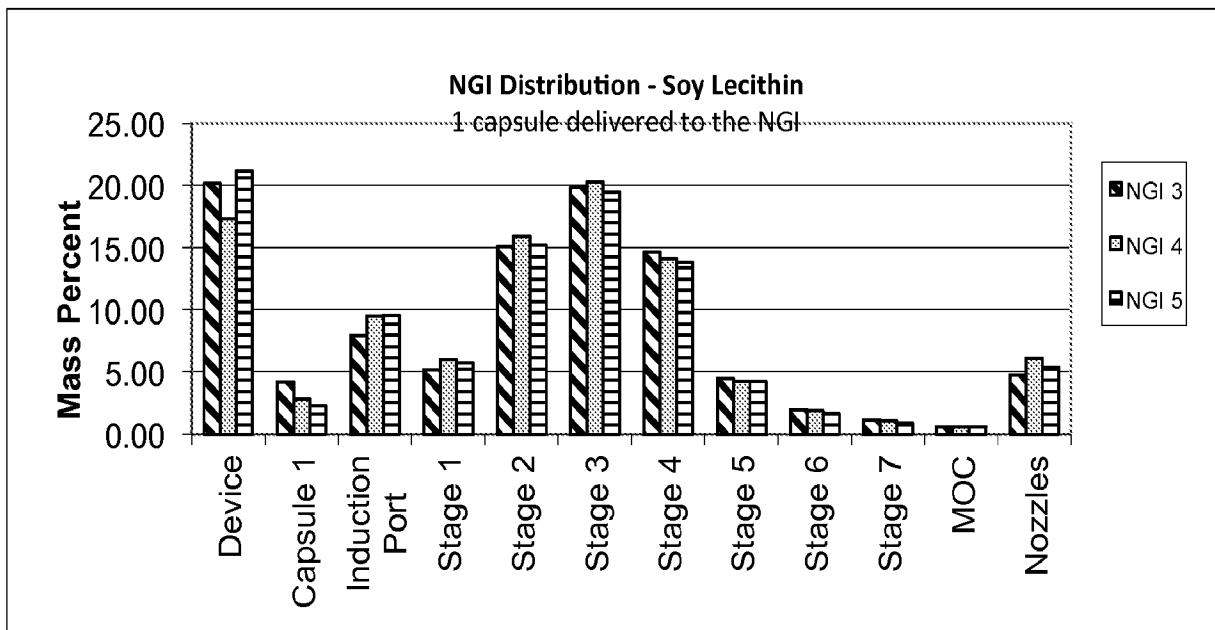


B.

Figure 7



A.



B.

Figure 8