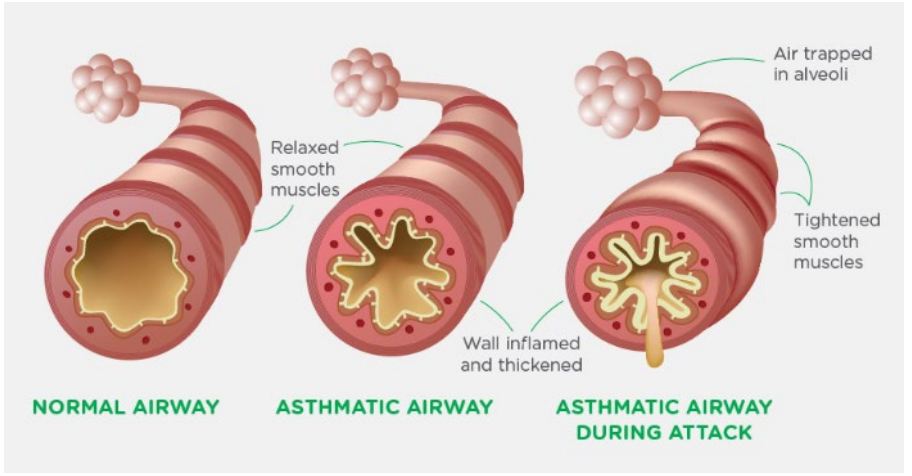
UAB SCHOOL OF ENGINEERING
The University of Alabama at Birmingham



UAB HEERSINK SCHOOL OF MEDICINE
The University of Alabama at Birmingham

Materials for Inhaled Aerosol Treatment of Disease:

Optical Photothermal Infrared Microscopy (O-PTIR) as an Advanced Characterization Method for Assessing the Emitted Dose of Complex Dry Powder Inhaler Formulations

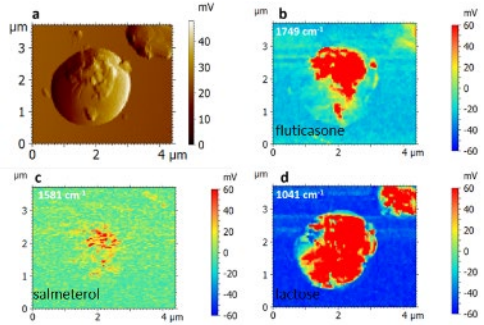
Mark Banaszak Holl

Department of Mechanical and Materials – School of Engineering
Division of Pulmonology, Allergy, and Critical Care Medicine – Heersink School of Medicine

September 2025

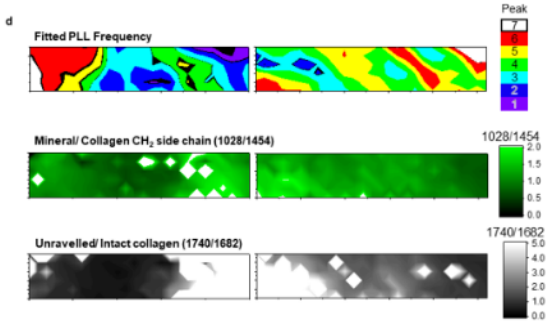
Banaszak Holl Lab Photothermal Infrared Applications

Drug Delivery



Analytical Chemistry 2025, *97,* 16421.
International Journal of Pharmaceutics 2024, 656, 124116.
International Journal of Pharmaceutics 2023, *646,* 123505
Advanced Drug Delivery Reviews 2023 *192*, 114646
International Journal of Pharmaceutics 2023 *632*, 122563
Analytical Chemistry 2020 *92*, 8328-8322

Bone and Ligament



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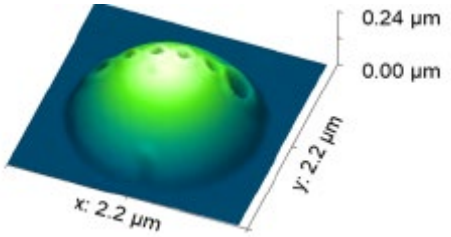

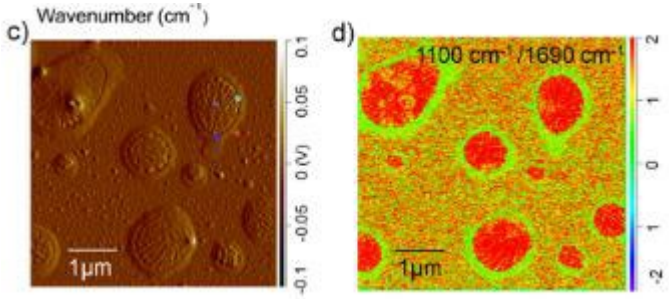

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J. Env. Management, 2023 *343*, 118205
Analytical Chemistry 2022 94, 11973
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ACS Earth and Space Chemistry 2022 6, 871

Chemical Engineering Journal 2022 428, 132614
ACS Applied Materials & Interfaces 2021 *13*, 46033
ACS Nano 2021 *15*, 13721
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Polymer 2021 *222*, 123643
Frontiers in Marine Science 2020 *7*, 576170
Global Challenges 2019 1800104
Analytical Chemistry 2017 *89*, 8594

Bruker IconIR
AFM-IR

Photothermal mIRage-LS
O-PTIR

Engineering for the Human Process - Aerosols for Drug Delivery

University of Alabama at Birmingham
School of Engineering Heersink School of Medicine

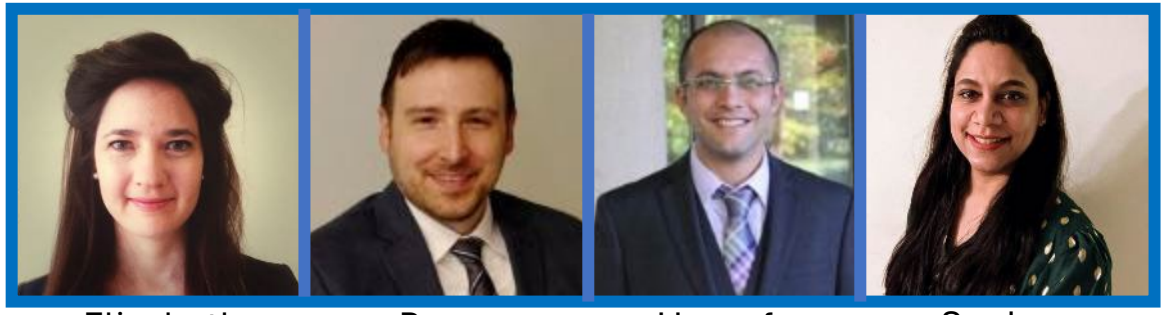
Department of Mechanical and Materials Engineering

Division of Pulmonology, Allergy, and Critical Care Medicine

Lung Health Center

Mark Banaszak Holl | Sheikh Tanzina Haque | Blessy Joseph | Terri Ross | Thanh Kien Ta

U.S. Food and Drug Administration



Elizabeth Bielski | Bryan Newman | Huzeyfe Yilmaz | Snober Ahmed

University of Sydney
School of Pharmacy

Hak-Kim Chan | Dipesh Khanal | Eric Cao

$$$ U.S. FDA Contract 75F40122C00202 Identification of Drug Distribution in Aerosols A Nanospectroscopy and NanoThermal Analysis

Asthma

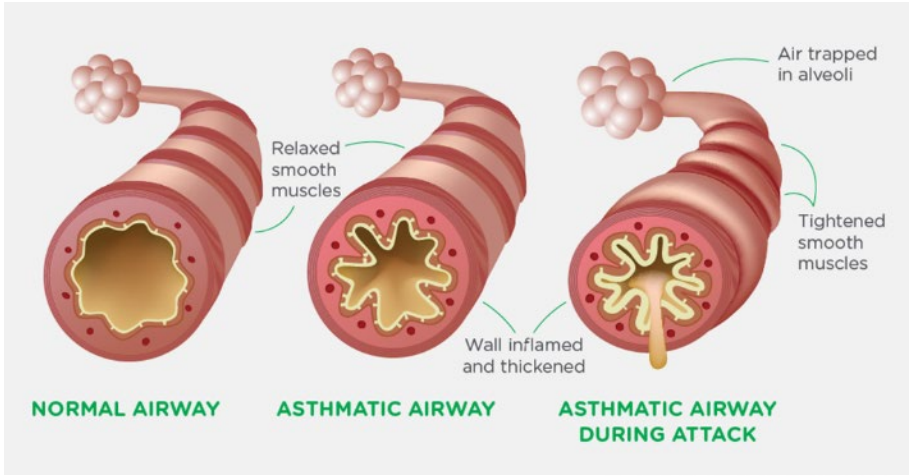
Chronic, inflammatory disease of conducting airways affecting 300 million people worldwide. Affluent societies have occurrence in 1 in 10 children and 1 in 12 adults. Estimated economic impact is estimated to exceed $18B. A mixture of genetic susceptibility and environmental risk factors lead to a variety of endotypes and consideration of asthma as a syndrome.

Both innate and adaptive immune systems act with epithelial cells to cause:

- Bronchial hyper-reactivity (non-specific response to exercise and cold air)
- Mucus overproduction
- Airway wall remodeling
- Airway narrowing

Leading to:

- Shortness of breath
- Wheezing
- Chest tightness



Lambrecht, Hammad *Nature* 2015, *16*, 45-56.

Asthma & COPD: A Legacy of the Past in Birmingham, Alabama

Sloss Furnaces, operated 1882-1970,
longest continually running blast furnace.
Museum and Community Center

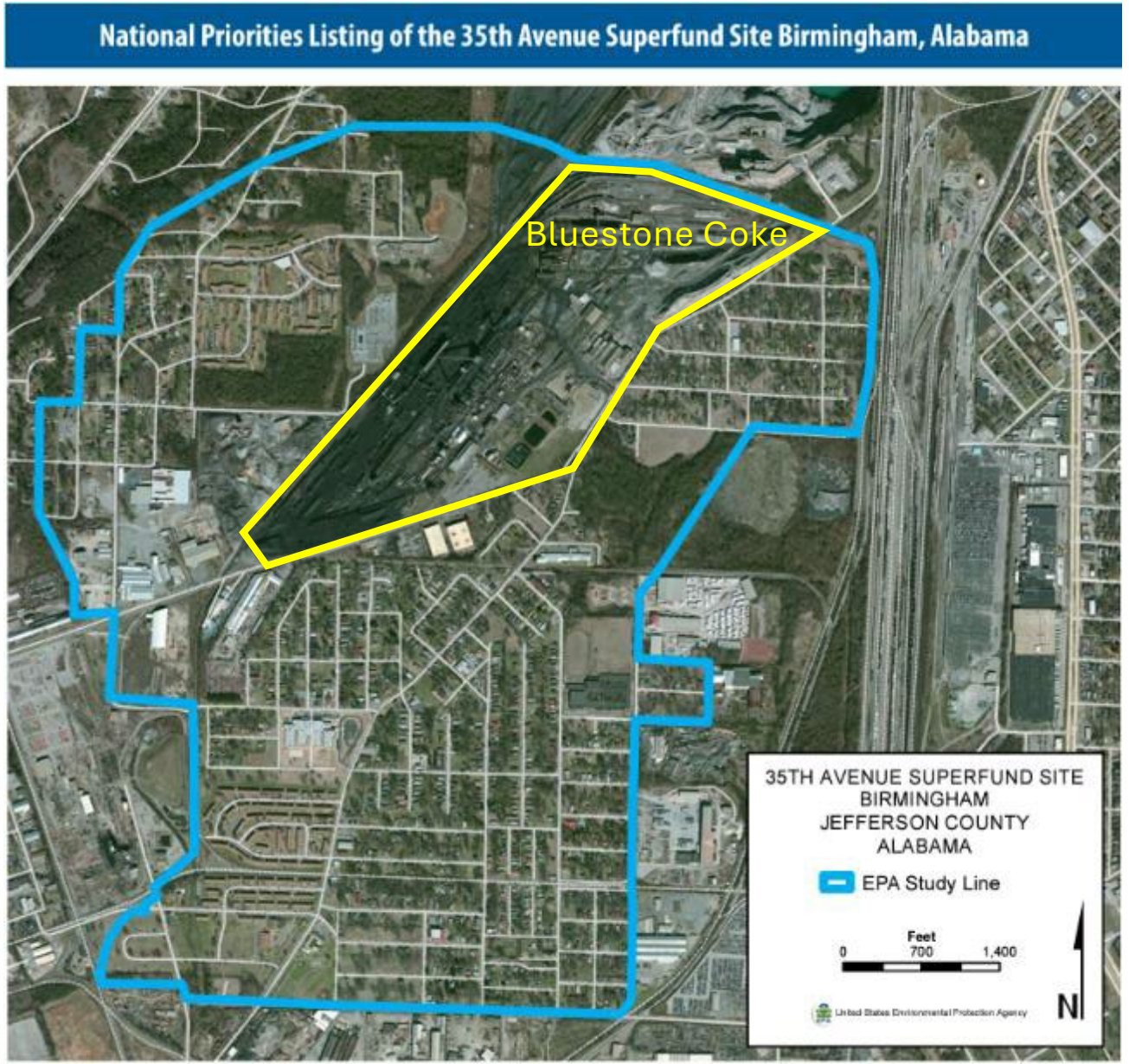
[The Environmental Injustices of Coke Plants in Birmingham, AL — ProPublica]

COPD: Chronic Obstructive Pulmonary Disease

EPA 35th Avenue Superfund Site

Birmingham, Alabama

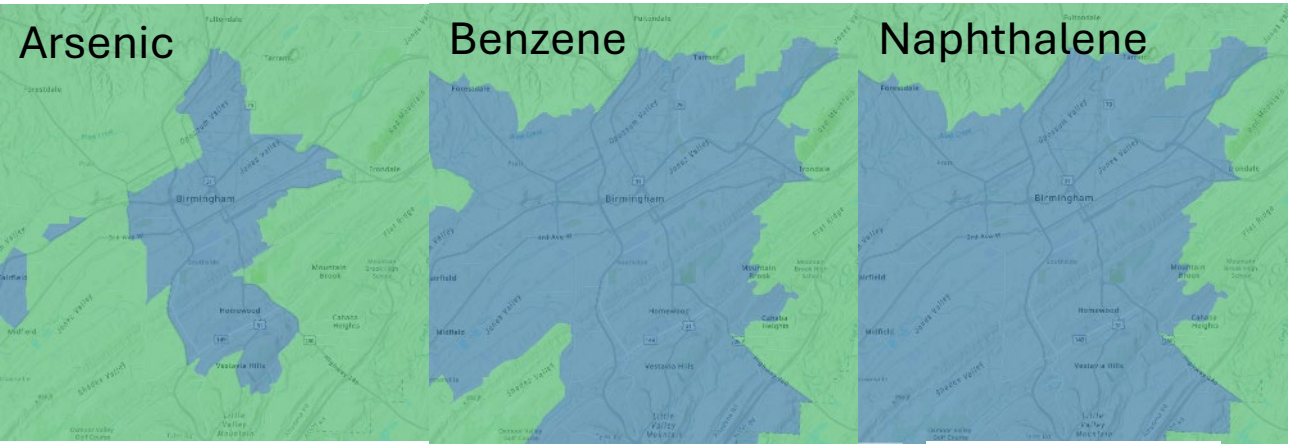
Airborne exposures to heavy metals contribute to multiple and overlapping lung diseases such as COPD, asthma, and lower respiratory tract infection.



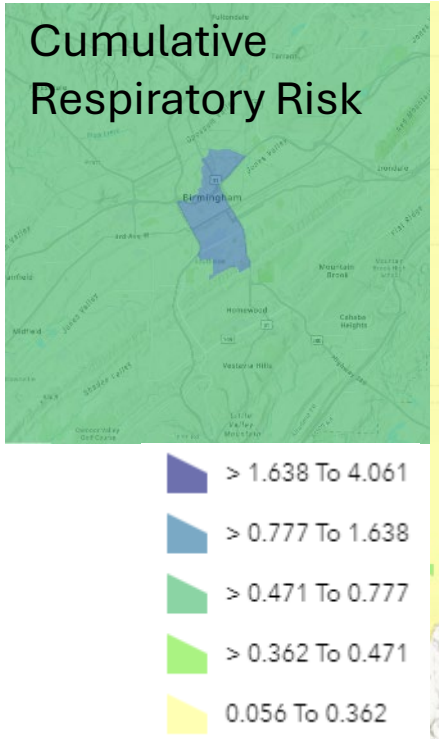
EPA: U. S. Environmental Protection Agency

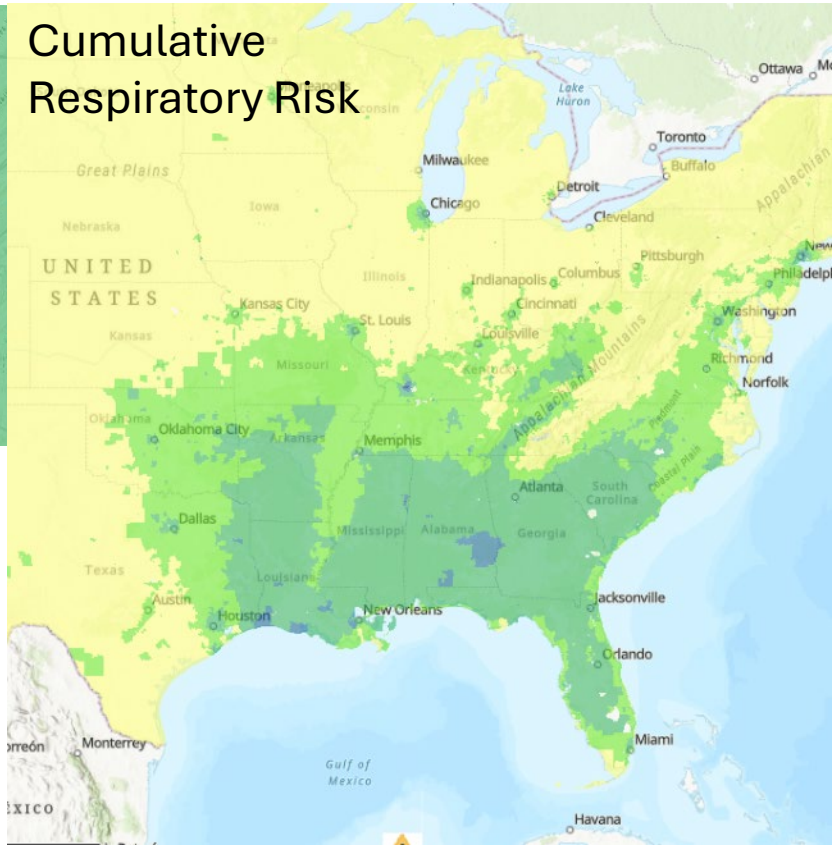
The Present

Birmingham, Alabama



µg/m^3



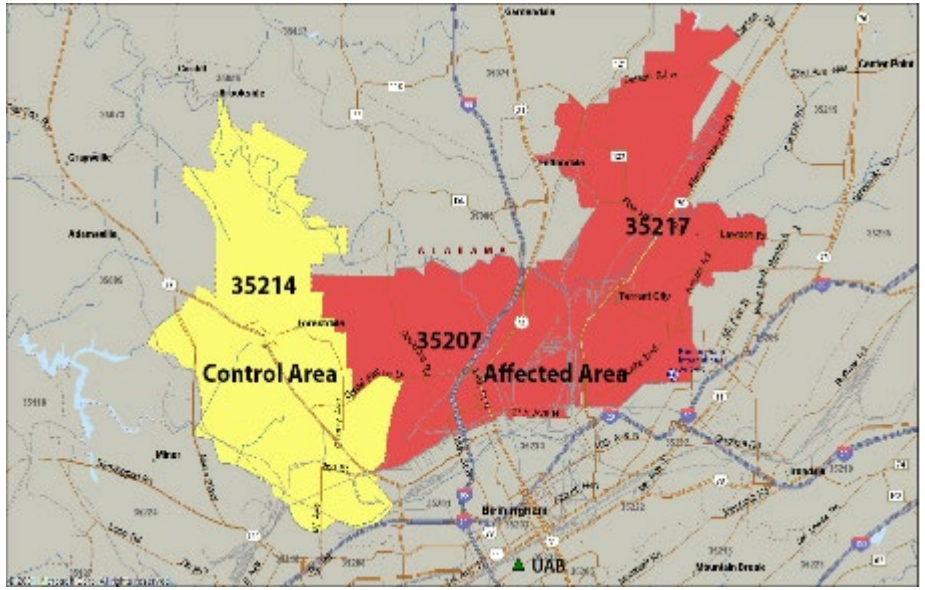


- Some of the worst air pollution in nation
- Not just urban - extends into Black Belt region and rural Alabama
- Pollution equivalent to largest petrochemical production sites in world (TX, LA)

https://enviroatlas.epa.gov/

The Present

Birmingham, Alabama



LUNG DISEASE IS HIGHLY PREVALENT

The Affected Area (EPA superfund site) has twice the prevalence of COPD than Control Area.

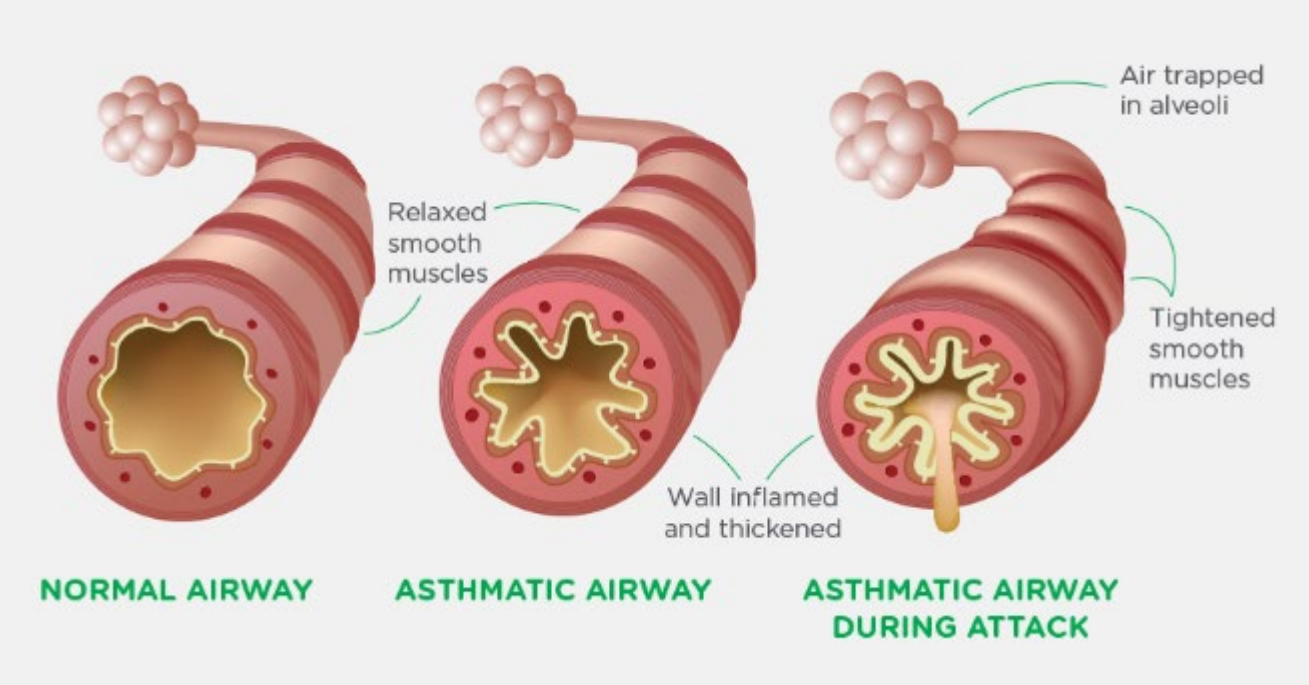
74% Children in K-8th grade have physician diagnosed Asthma (7x national average!).

Current and Ongoing Health Crisis!

Major plants currently operating in the city including ABC Coke, ACIPCO, NUCOR.

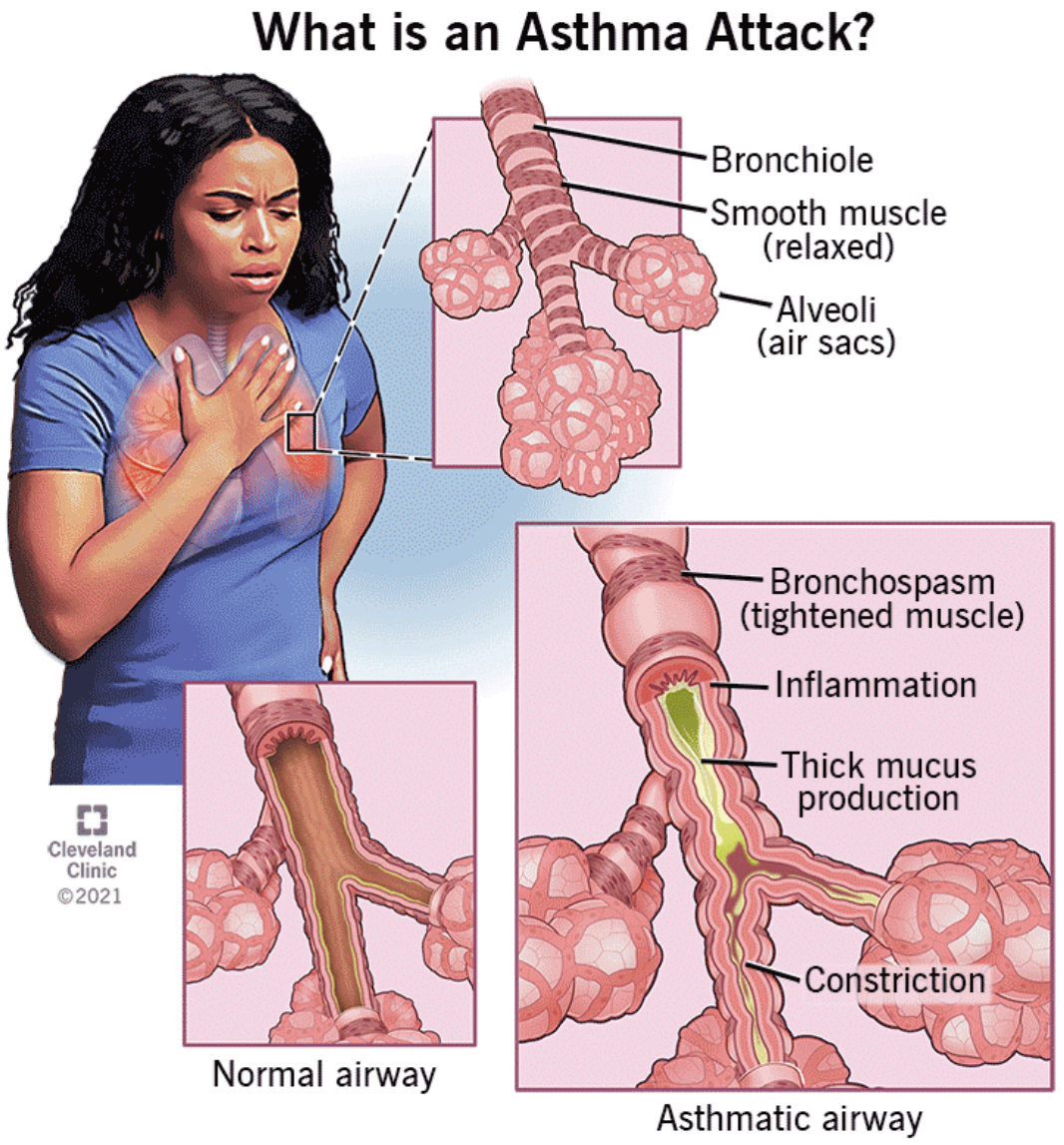
ACIPCO in process of switching from coke to electric induction furnaces.

Asthma flare-ups are caused by triggers, which are irritants like high smog levels, colds and flu, cigarette smoke, scented cleaning products or even seasonal changes in the weather.



What happens during an asthma attack?

The airways get swollen and inflamed.
Thick, sticky mucus fills up the airways.
The muscles that wrap around the airways squeeze tight.
Air becomes trapped in alveoli.



https://www.choa.org/parent-resources/asthma/asthma-attacks

https://my.clevelandclinic.org/health/diseases/6424-asthma

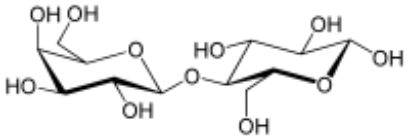
Inhaled Corticosteroid (ICS) and β2-agonist for Asthma Treatment

Corticosteroids (i.e., steroids): anti-inflammatory medicine. Synthetic version of hormones normally produced by the adrenal glands (two small glands that sit on top of the kidneys).

Effective for ~90% of Asthma cases

Fluticasone Propionate (FP): Flovent HFA, Flonase, and Flixotide

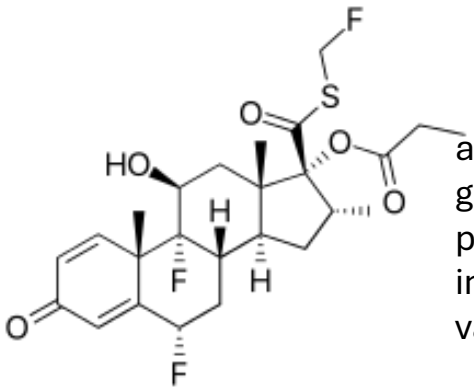
Mixed with lactose excipient



Fluticasone Propionate Metered Inhalation Aerosol (U.S. Marketed)

Fluticasone Propionate Metered Nasal Spray (U.S. Marketed)

Fluticasone Propionate Metered Inhalation Aerosol (EU, AUS, NZ marketed)

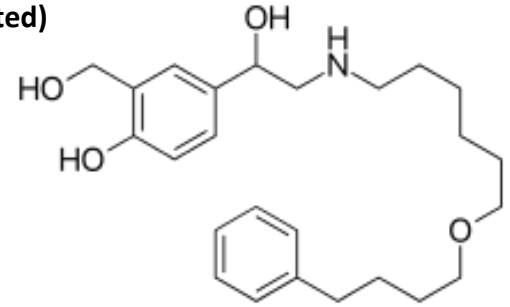


agonist at the glucocorticoid receptor producing anti-inflammatory and vasoconstriction effects.

β2-agonist:

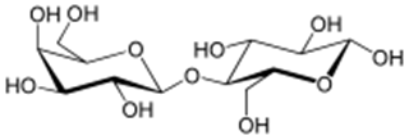
Salmeterol Xinafoate (SX): Serevent

Salmeterol Xinafoate Inhalation Powder (U.S. Marketed)

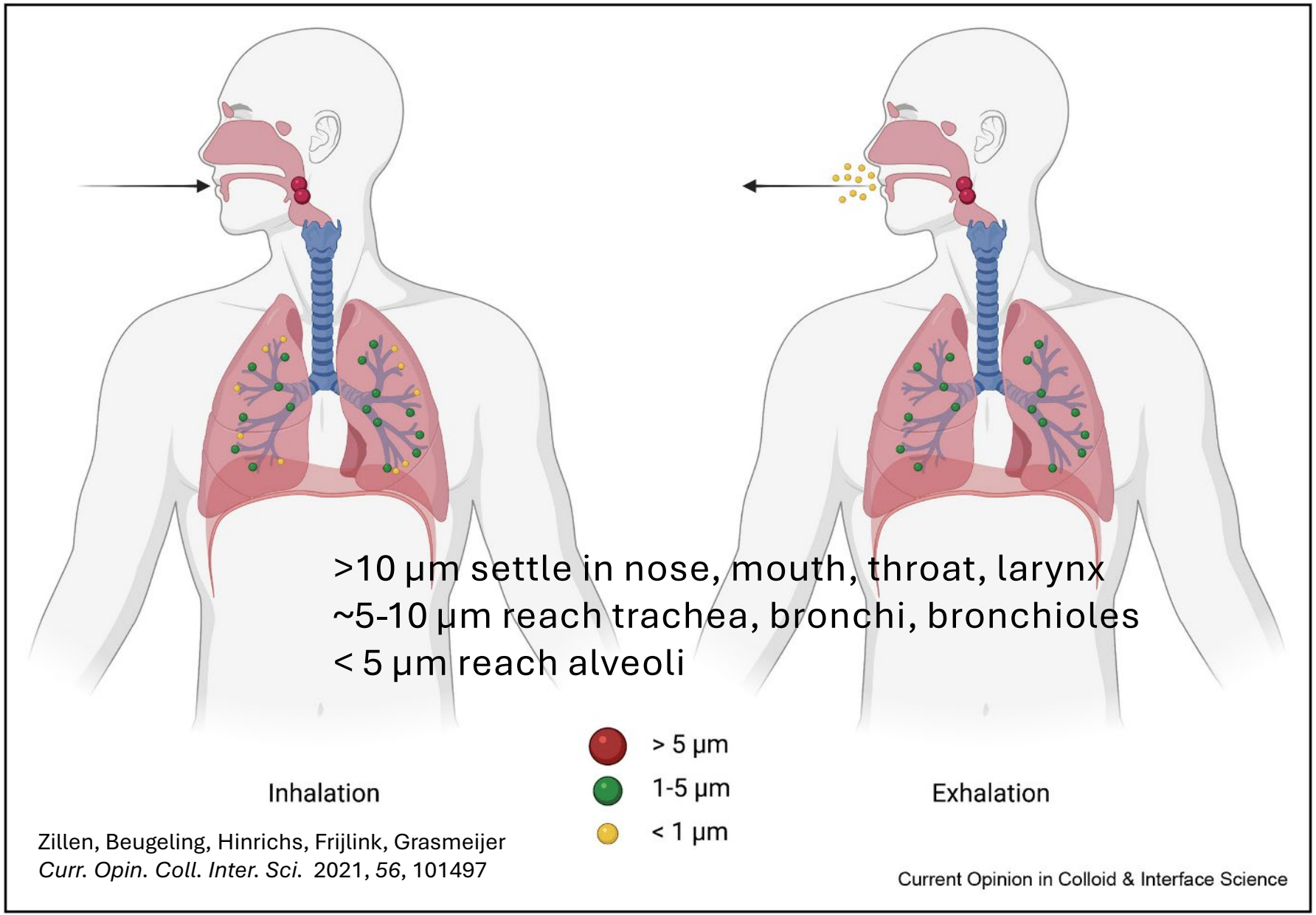


β2-agonist that relaxes bronchial muscles in the lungs – 12 hour action

Jetzer, M. W.; Morrical, B. D.; Fergenson, D. P.; Imanidis, G. *Int. J. Pharm.* 2017, *532*, 218–228.



Dispersion into lungs with inhaler achieved by absorbing drugs on micron-size lactose particles.



Advair Diskus (fluticasone propionate; salmeterol xinafoate) Inhalation Powder

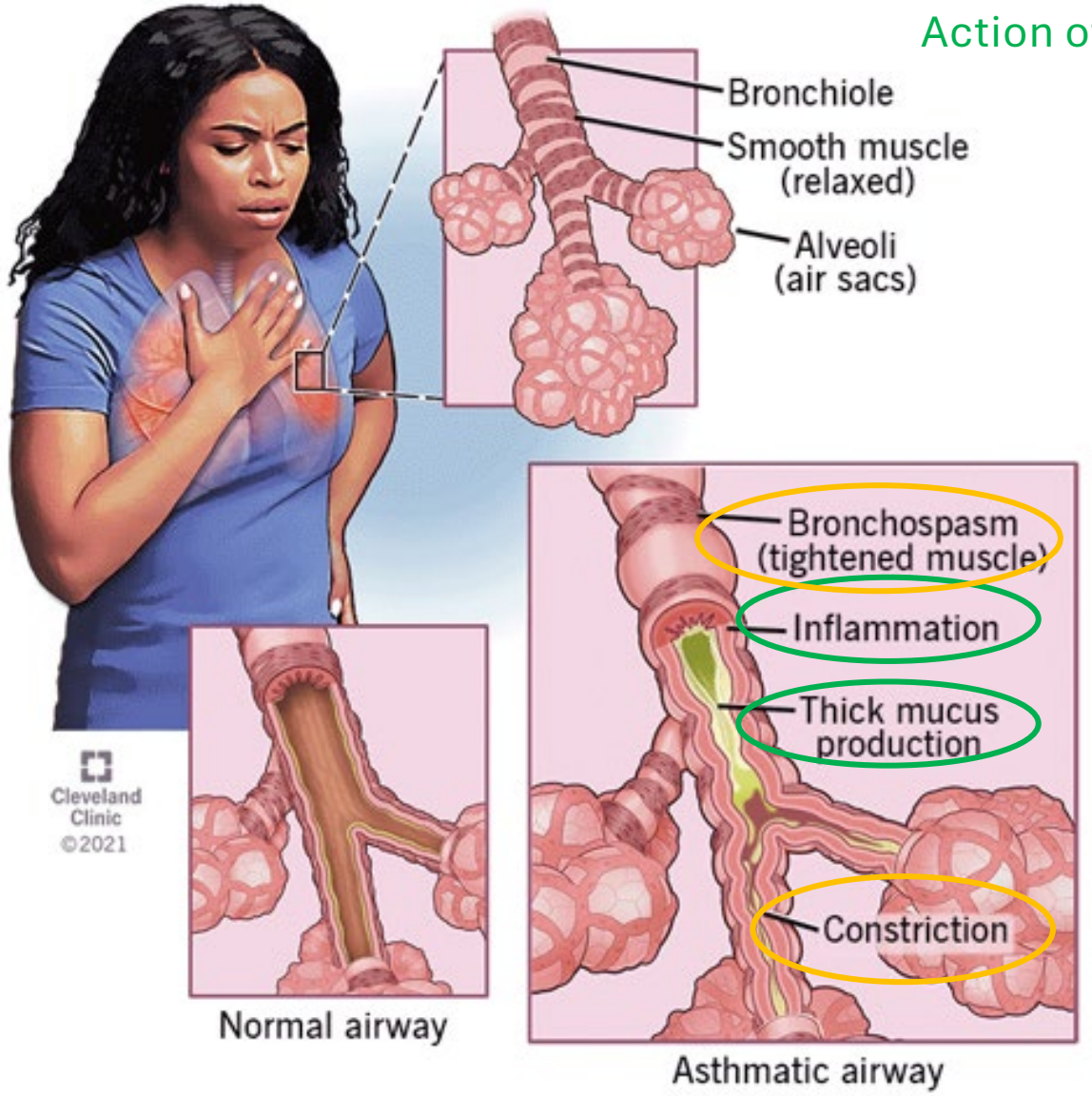
Deliver directly to lung cells to obtain therapeutic dose

while

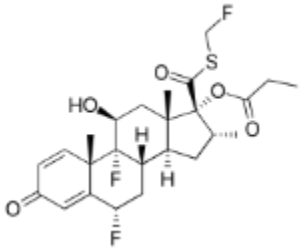
Reducing systemic delivery and associated side effects

Common: Headaches, sore throat, cramps, increased heart rate

Rare: allergic reaction, bone density loss, glaucoma, slowed growth (children)



Action of inhaled corticosteroid fluticasone

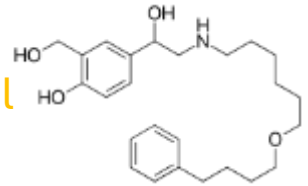


"significant clinical and in-vitro evidence to suggest that the co-association of steroid AND β2-agonist at the level of the single administered aerosol particle leads to a synergetic effect with improved clinical outcomes"

Synergistic effects operating at both the molecular receptor and cellular levels

Important to deliver BOTH drugs to the SAME cell in the lung tissue

Action of β-2 agonist salmeterol

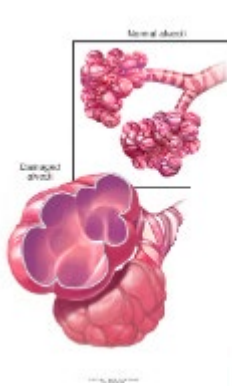


Jetzer, M. W.; Morrical, B. D.; Fergenson, D. P.; Imanidis, G. *Int. J. Pharm.* 2017, *532*, 218–228.

Treatments also relevant to

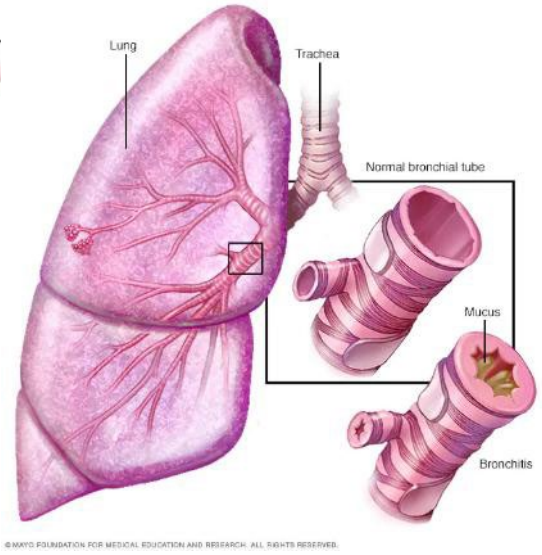
Chronic Obstructive Pulmonary Disease (COPD)

Affects about 10% of people over 45 years of age with a similar prevalence in males and females in developed counties reflecting smoking pattern.



Emphysema

Destruction of the fragile walls and elastic fibers of the alveoli. Small airways collapse upon exhalation.



Bronchitis

Bronchial tubes become inflamed and narrowed, and the lungs produce more mucus.

In developing countries, over half of the COPD patients are non-smokers, particularly women exposed to biomass smoke in poorly ventilated homes.

COPD is now ranked as the 4th most common cause of death in the world. Although, in developed countries, it has risen to 3rd most common cause of death, is ranked as the 5th commonest cause of morbidity, and is a leading cause for emergency hospital admission with acute exacerbations.

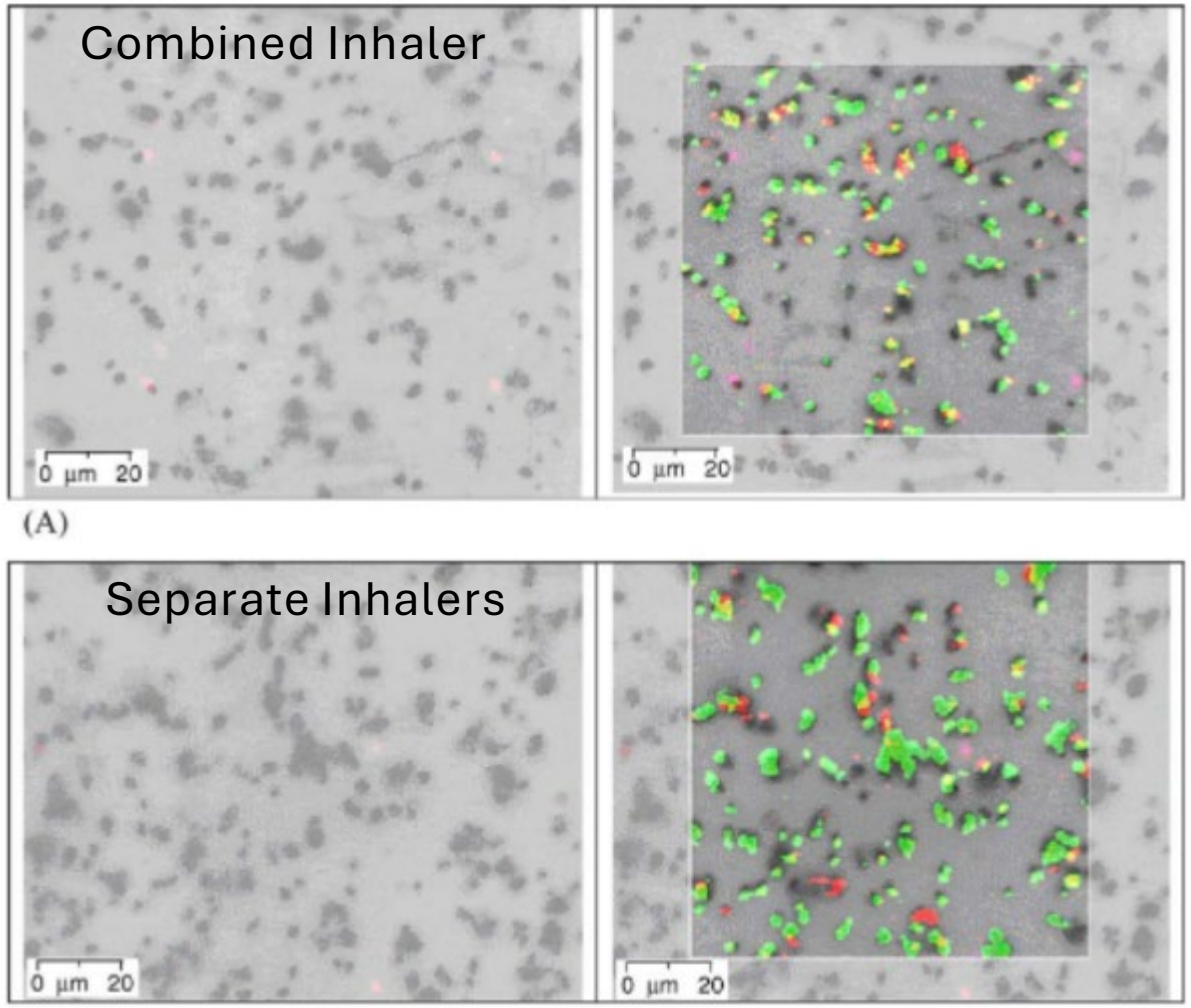
Substantial co-morbidity in Adults with Asthma

- Respiratory infection
- Heart Problems
- Lung Cancer
  - Depression

https://www.mayoclinic.org/diseases-conditions/copd/symptoms-causes/syc-20353679

Barnes *Allergy* 2019, *74*, 1249-1256

What is known about Aerosol Particle Content and Structure?



Fluticasone and Salmeterol Particle Co-Association by Raman Microscopy

Fluticasone Propionate
Salmeterol Xinafoate

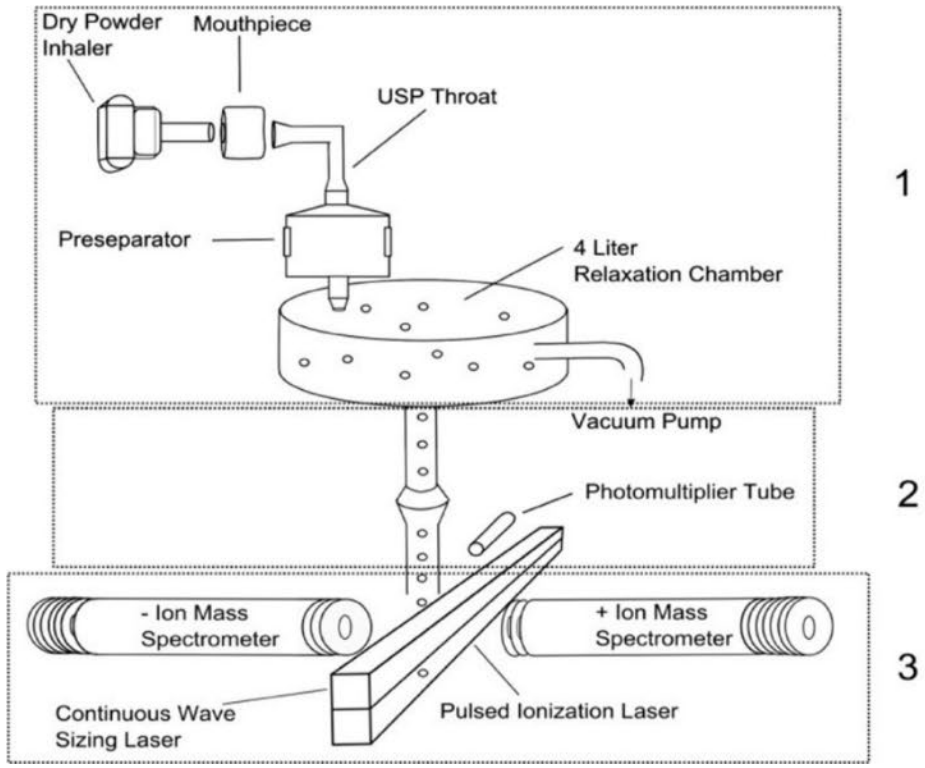
Drugs combined in particles Yellow

Spatial resolution ~ 3 µm or about size of many single particles

Theophilus, A.; Moore, A.; Prime, D.; Rossomanno, S.; Whitcher, B.; Chrystyn, H. *Int. J. Pharm*. 2006, *313*, 14–22.

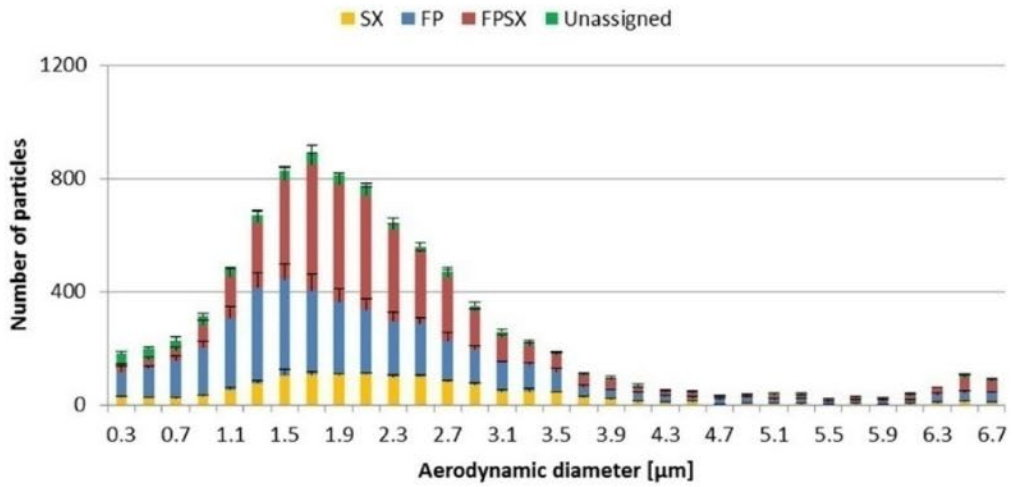
Fluticasone and Salmeterol Particle Co-Association by Mass Spectrometry

Novartis/University of Basel



Fluticasone Propionate : Salmeterol Xinafoate

10-Fluticasone/ 1-Salmeterol co-associated particles

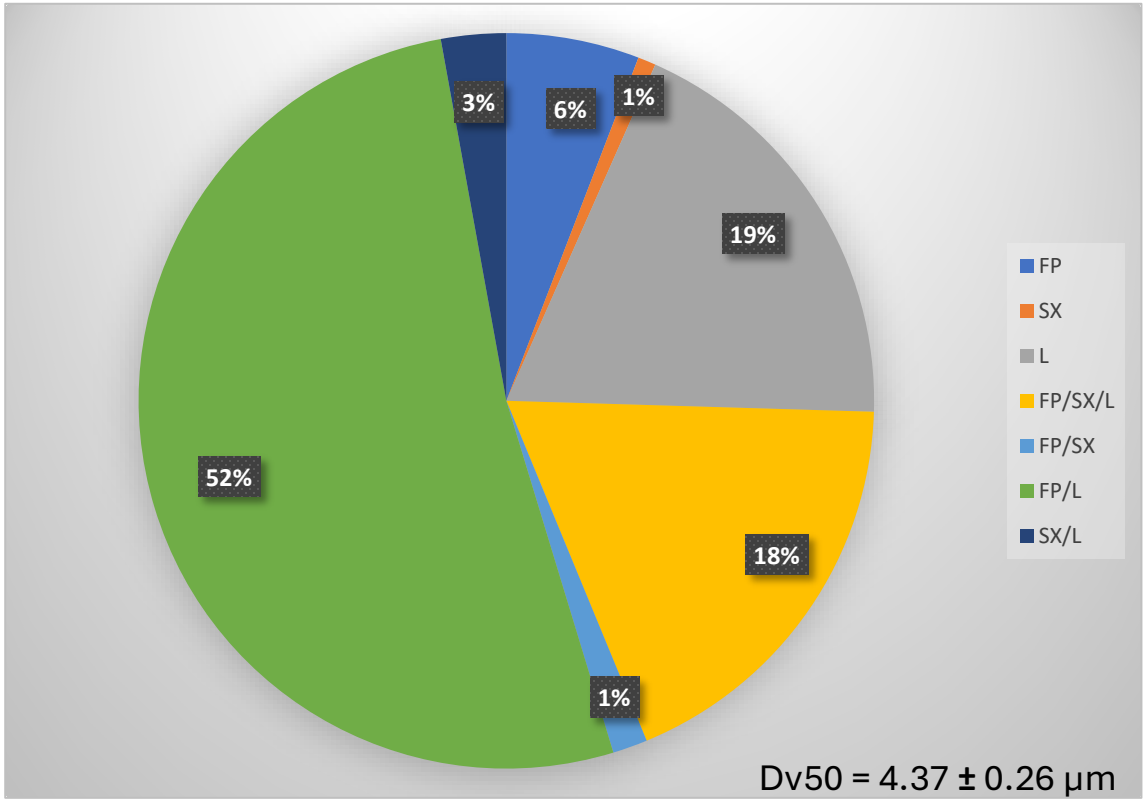


- 45% of particles contain fluticasone and salmeterol
- 70% of particles containing salmeterol also contain fluticasone

Jetzer, M. W.; Morrical, B. D.; Fergenson, D. P.; Imanidis, G. *Int. J. Pharm.* 2017, *532*, 218–228.

Characterization of Advair Diskus (fluticasone propionate; salmeterol xinafoate inhalation powder) using Morphologically-Directed Raman Spectroscopy (MDRS)

Single Actuation, Next Generation Impactor (NGI) Stage 2

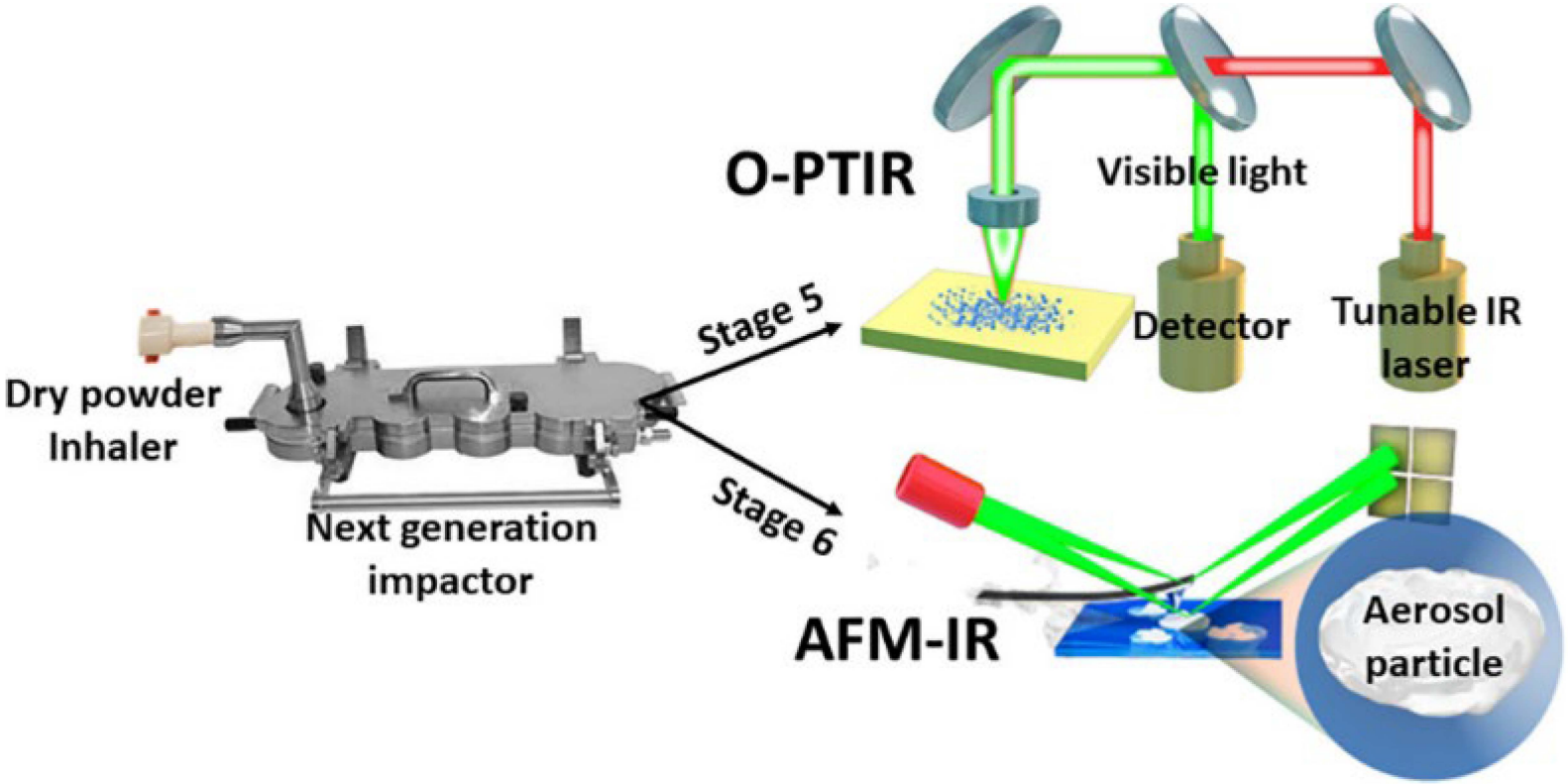


Chemical classification of agglomerates of Advair Diskus 100/50 (fluticasone propionate; salmeterol xinafoate inhalation powder, 0.1 mg/inh; EQ 0.05 mg base/inh) collected at 60 L min^{-1} with a USP inlet and pre-separator connected to the ADC apparatus. Quantities are a percentage by number and an average of 6 independent measurements of at least 3000 particles.

Dv50 is the mean diameter by volume

Farias G, Ganley WJ, Price R, Conti DS, Mangal S, Bielski E, Newman B, Shur J., *Pharmaceutical Research*. **2024** Oct;41(10):2015-29.

Analytical Challenge: Solid-State Characterization of Pharmaceutical Inhalation Aerosols



- Nanometer to Micrometer Particles
- Complex Drug/Excipient Mixtures
- Assess variation at particle level?
- Use O-PTIR and AFM-IR

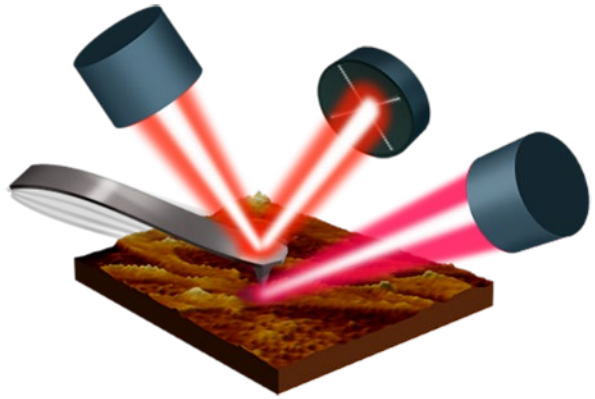
Optical Photothermal Infrared Microscopy (O-PTIR) - 0.5 µm (500 nm) spatial resolution on individual particles

Atomic Force Microscopy – Infrared Spectroscopy (AFM-IR) - 0.01 µm (10 nm) resolution on individual particles

D. Khanal; J. Zhang, M. M.; Banaszak Holl; H.K. Kim, *Analytical Chemistry* 2020, *92*, 12, 8323-8332.

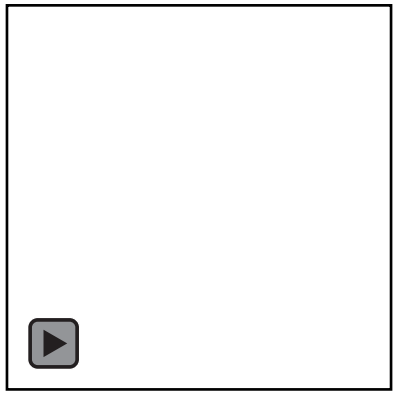
Photothermal Infrared Spectroscopy

AFM-IR: Bruker IconIR



- Nano-scale (10 nm) thermal expansion detected via AFM tip for IR spectrum
- Simultaneous acquisition of mechanical data including modulus, adhesion, dissipation, and deformation)
- nanoThermal analysis (T_g, T_m)

O-PTIR: Photothermal mIRage-LS



- Micro-scale (0.5 μm) thermal expansion detected via green laser for IR spectrum
- Simultaneous Raman spectrum
- Rapid all-optical system
- Co-located fluorescence microscopy

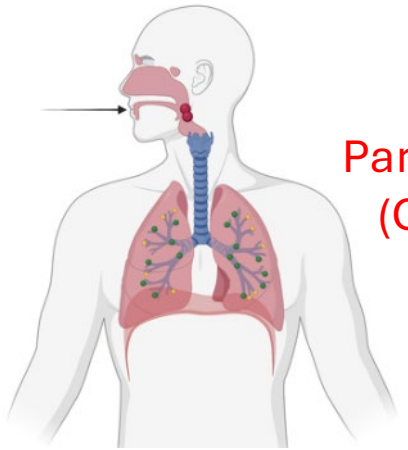
Conventional Raman and FTIR microscopy probe chemical information from the aggregates of aerosol particles in the size range greater than a few micrometers.

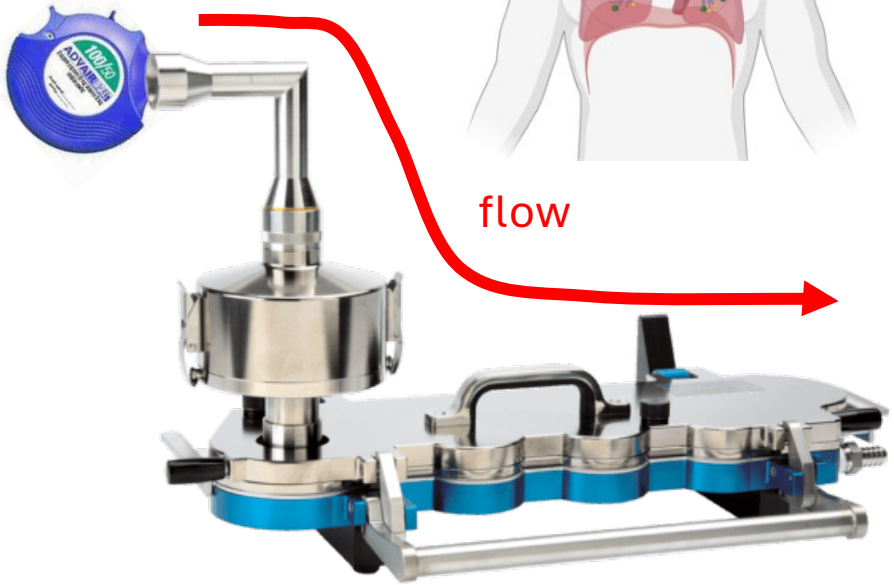
https://www.bruker.com/en/products-and-solutions/infrared-and-raman/nanoscale-infrared-spectrometers/dimension-iconir.html

https://www.photothermal.com/home/co-located-fluorescence-optir/

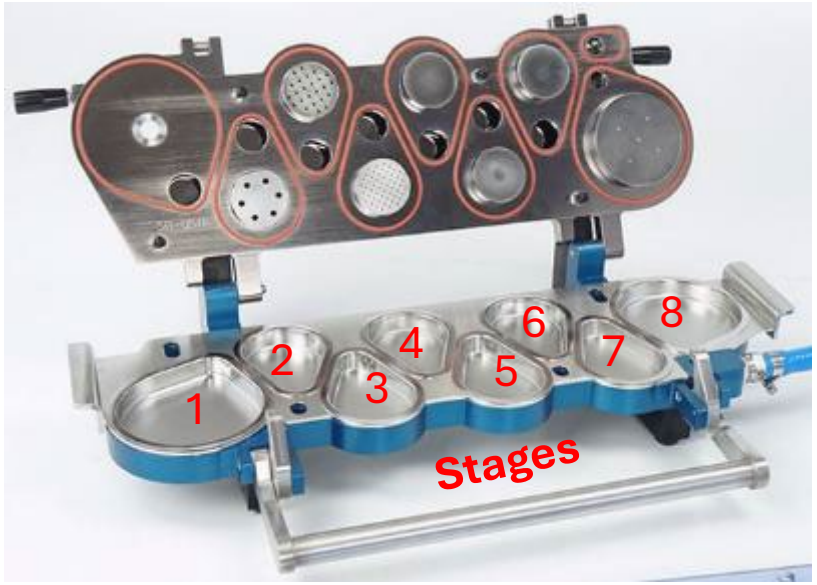
Evaluating Aerodynamics - Next Generation Impactor

Particles In Here





Particles collected on silicon (Si) wafers or calcium fluoride (CaF_2) cover slips at different stages as a function of size



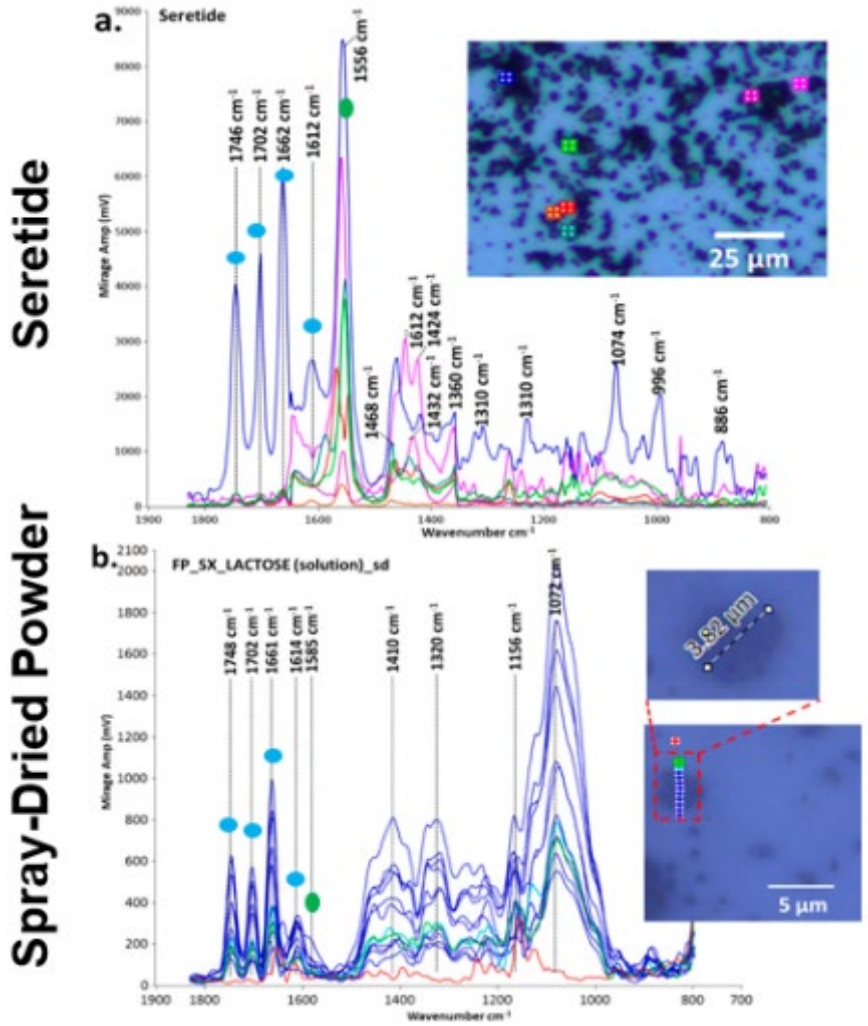
Analysis

O-PTIR: mIRage-LS

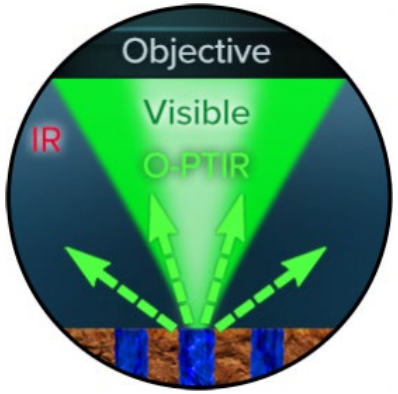
AFM-IR: IconIR

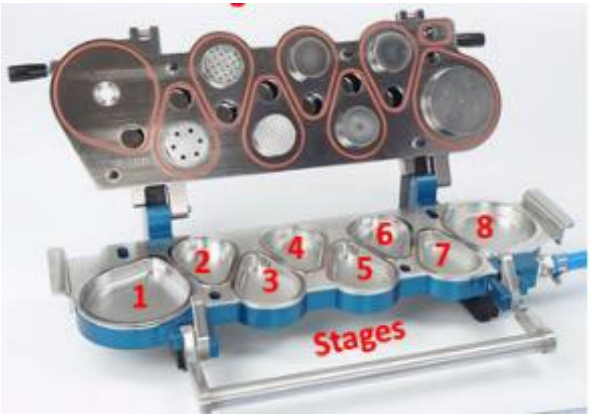
Stage	1	2	3	4	5	6	7	8
Cutoff Diameter in microns	6.1	3.4	2.2	1.3	0.72	0.4	0.24	< 0.24

O-PTIR spectra of sample collected from stage 5 of NGI
(a) Seretide (fluticasone propionate [FP]; salmeterol xinafoate [SX] inhalation powder), 500 µg/inh; 50 µg base/inh; (b) spray-dried FP; SX powder from solution.



O-PTIR





Flow Rate: 100 L/min; Time: 1.2 s
Stage 5: 0.72 µm cutoff

Both Drugs and Lactose Identified in IR spectra.

Rapid drug specific imaging of ~100,000 µm^3 ROIs and thousand of particles at ~ 0.3 µm resolution.

Comparison of micronized aerosol powder inhaler drug particles to spray-dried particles.

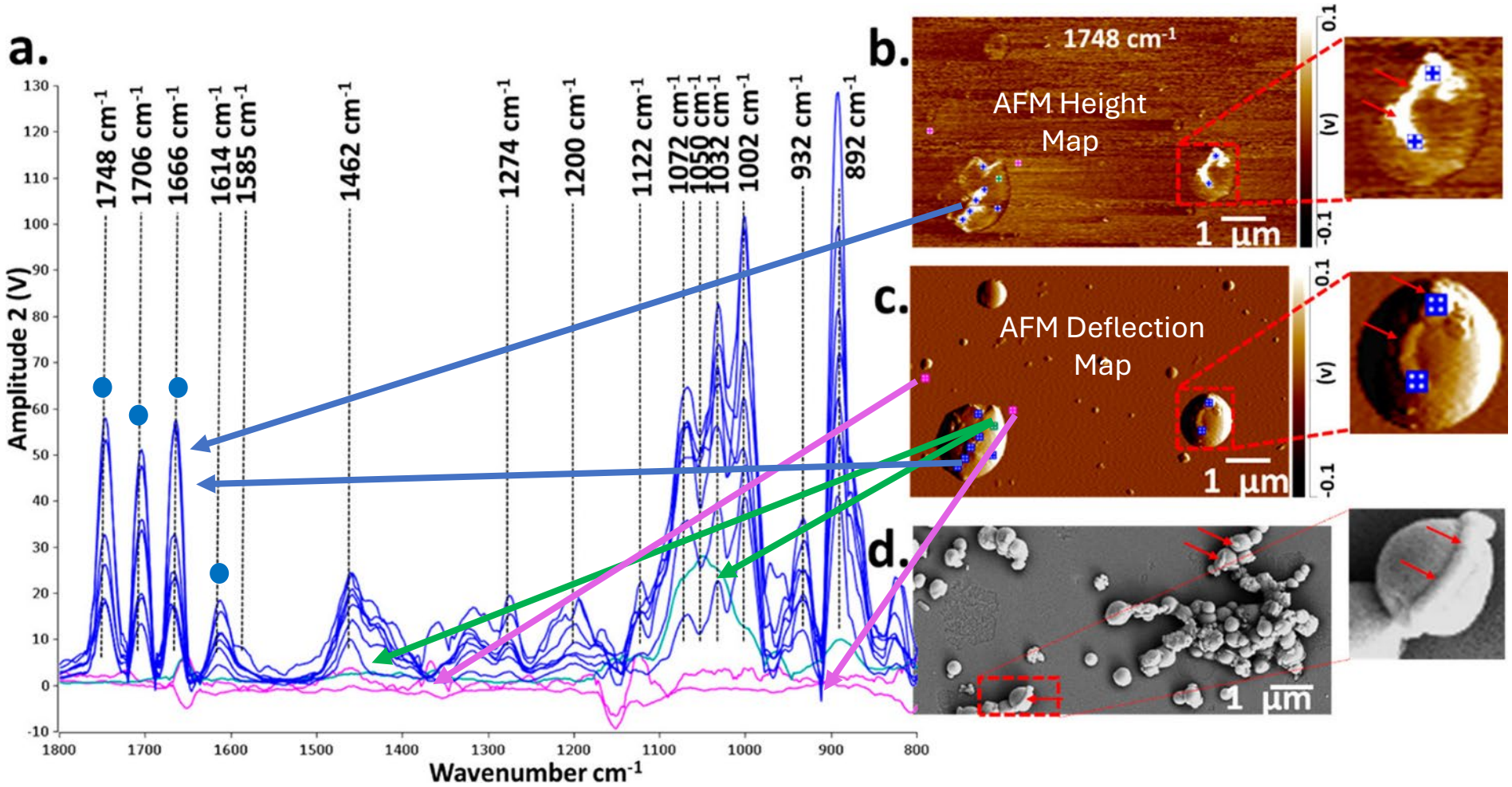
● Fluticasone Propionate ● Salmeterol Xinafoate

D. Khanal; J. Zhang, M. M.; Banaszak Holl; H.K. Kim, *Analytical Chemistry* 2020, *92*, 12, 8323-8332.

Localization of Fluticasone Propionate in 1 µm Lactose Particle

- Fluticasone Propionate
  Blue squares indicate location of spectra

aerosol particles of spray-dried powder



IR of Fluticasone Propionate

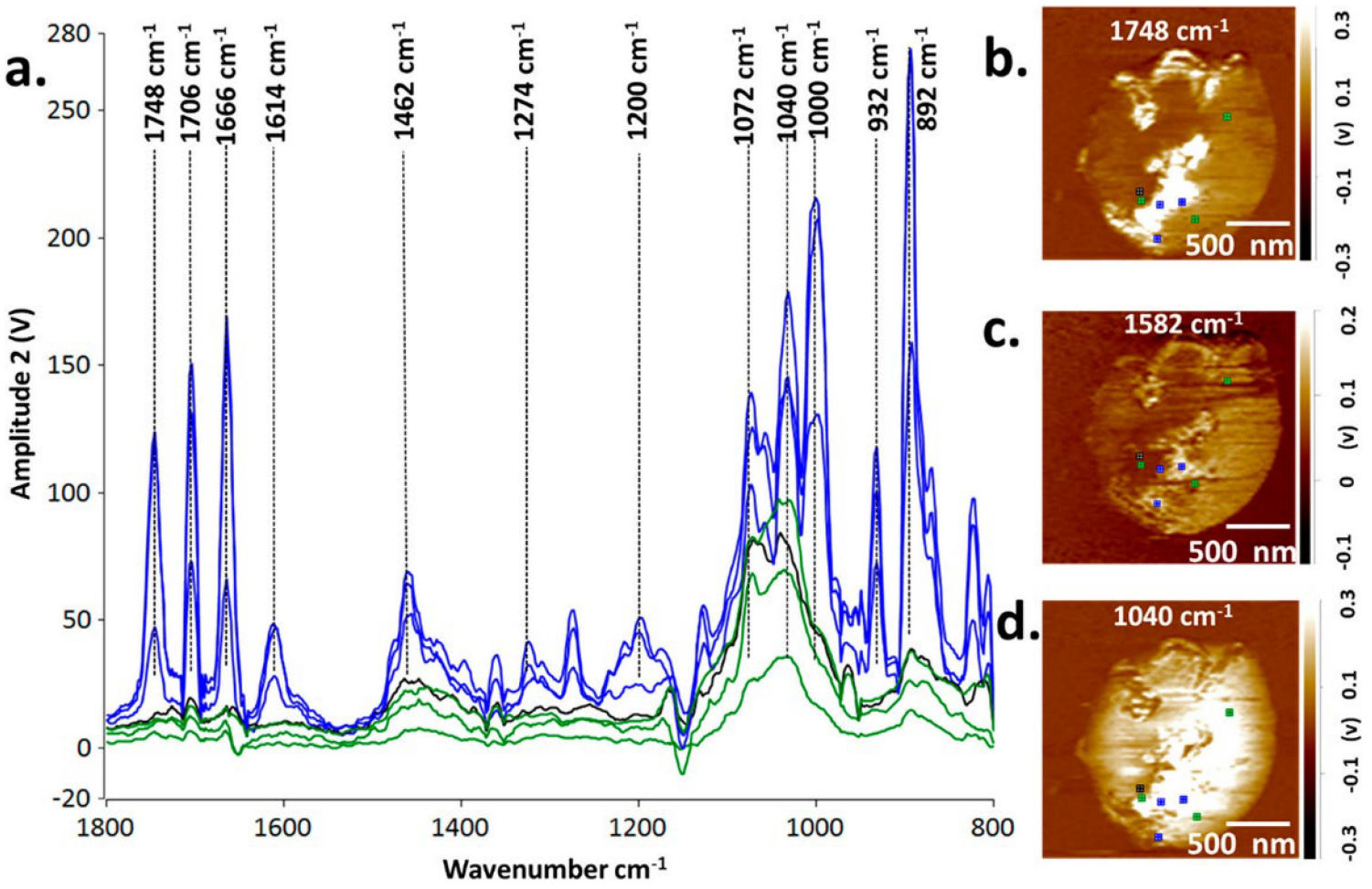
IR of Lactose Green Square

IR of Background Pink Square

SEM image
Drug Deposit Indicated

D. Khanal; J. Zhang, M. M.; Banaszak Holl; H.K. Kim, *Analytical Chemistry* 2020, *92*, 12, 8323-8332.

Single Particle Characterization of Drug Aerosol



Fluticasone Propionate
IR Map (1748 cm^{-1})

Highly Localized

Salmeterol Xinafoate
IR Map (1582 cm^{-1})

Smaller amount co-localized with Fluticasone

Lactose
IR Map (1040 cm^{-1})

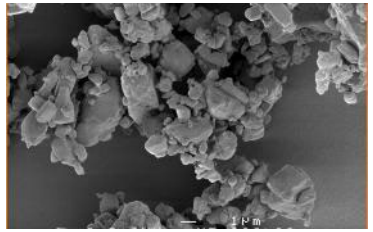
Bulk of particle

D. Khanal; J. Zhang, M. M.; Banaszak Holl; H.K. Kim, *Analytical Chemistry* 2020, *92*, 12, 8323-8332.

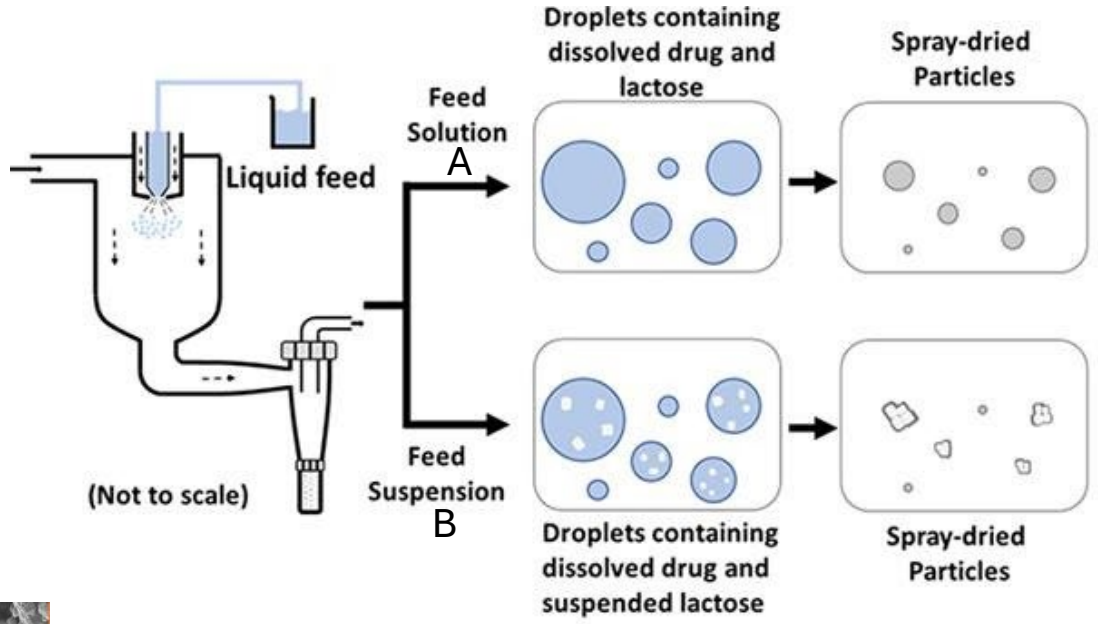
Formulation Effects on Drug Content with Aerosol Stage for Spray-Dried Alternatives

Khanal Ke

Zhang Kim

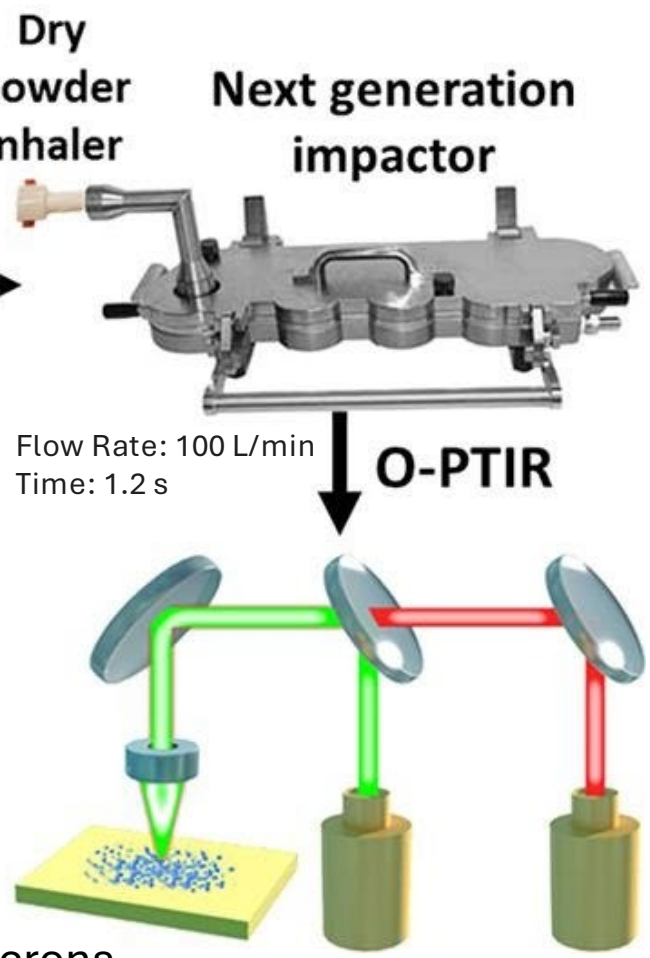


Lactohale® 300 is a very fine, micronised lactose with a D50 below 5 microns.



A: 8 FP to 1 SX w/ LH300 in 80/20 v/v isopropanol/water

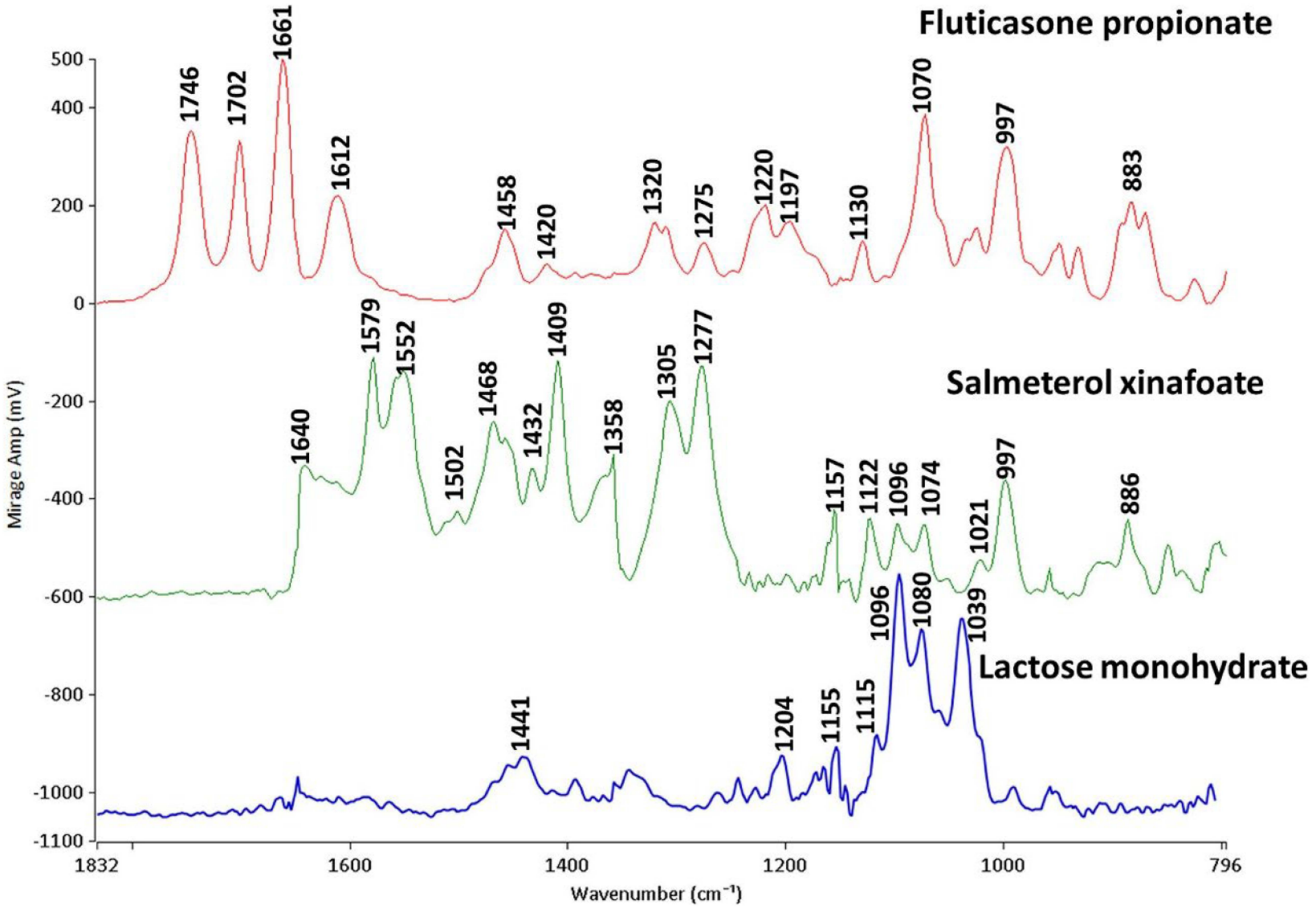
B: 8 FP to 1 SX w/ LH300 in isopropanol



D50 indicates *Median* particle size, which is 5 microns

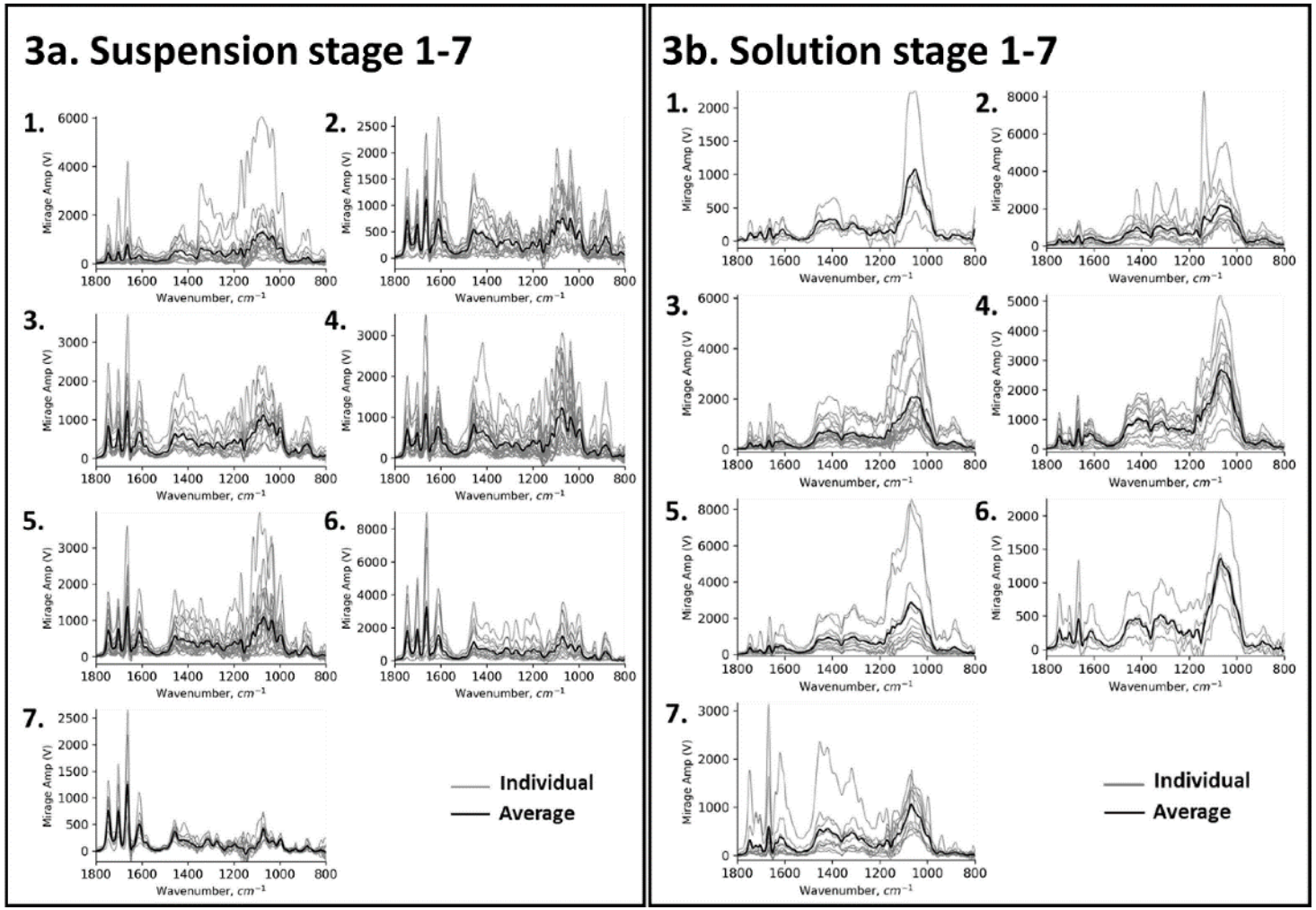
FP: fluticasone propionate; SX: salmeterol xinafoate; LH300: lactose monohydrate

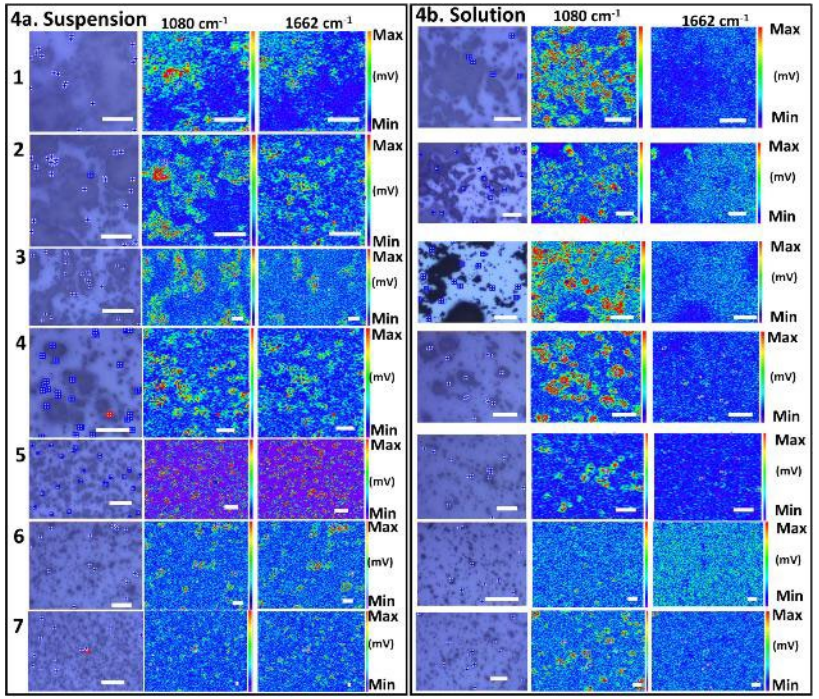
O-PTIR Spectra of Drugs and lactose Monohydrate (LH300)



O-PTIR spectra collected of particles collected from stages 1-7 of the NGI

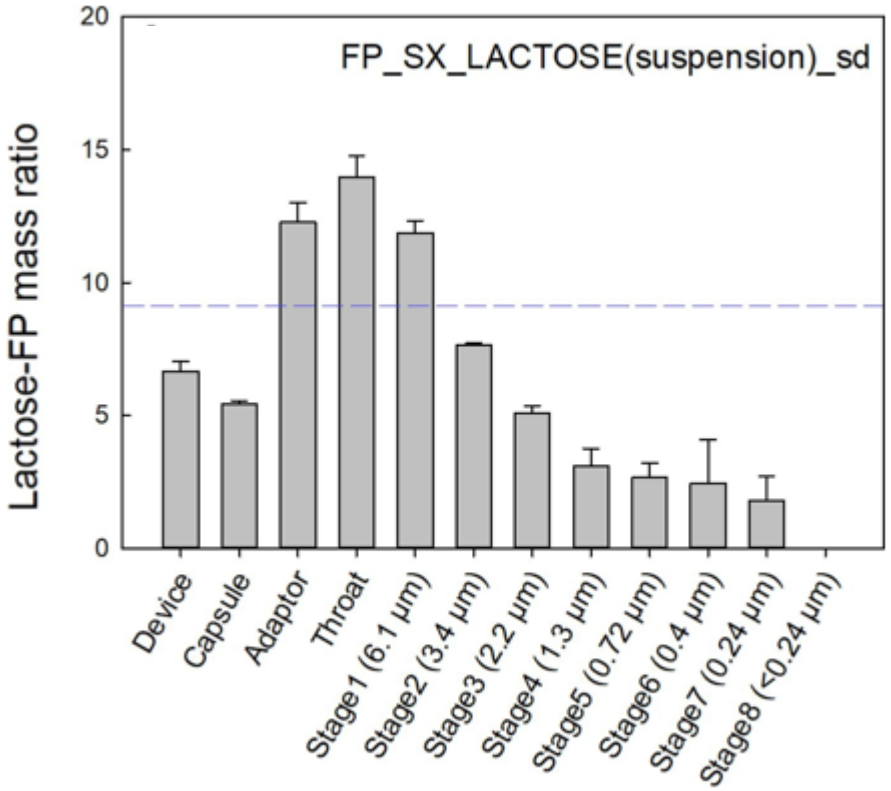
Particles collected on Si wafers or CaF_2 cover slips at different stages as a function of size



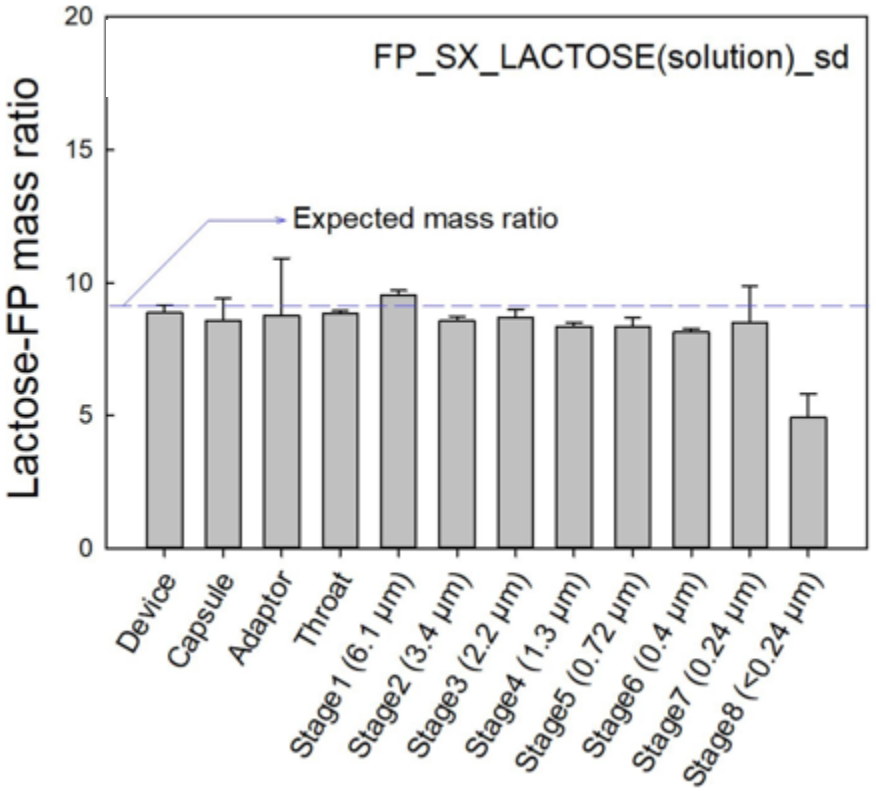


Stage	1	2	3	4	5	6	7	8
Cutoff Diameter in microns	6.1	3.4	2.2	1.3	0.72	0.4	0.24	< 0.24

Mass ratio of lactose to fluticasone propionate based on bulk HPLC chemical analysis of particles collected from stages 1–7 of NGI



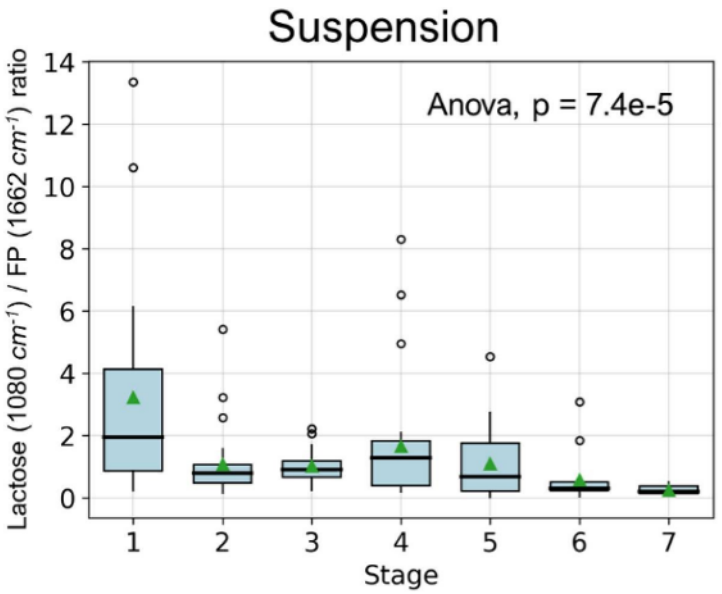
Solute composition in the suspension spray-dried particles varies with stage with less FP for smaller particles.

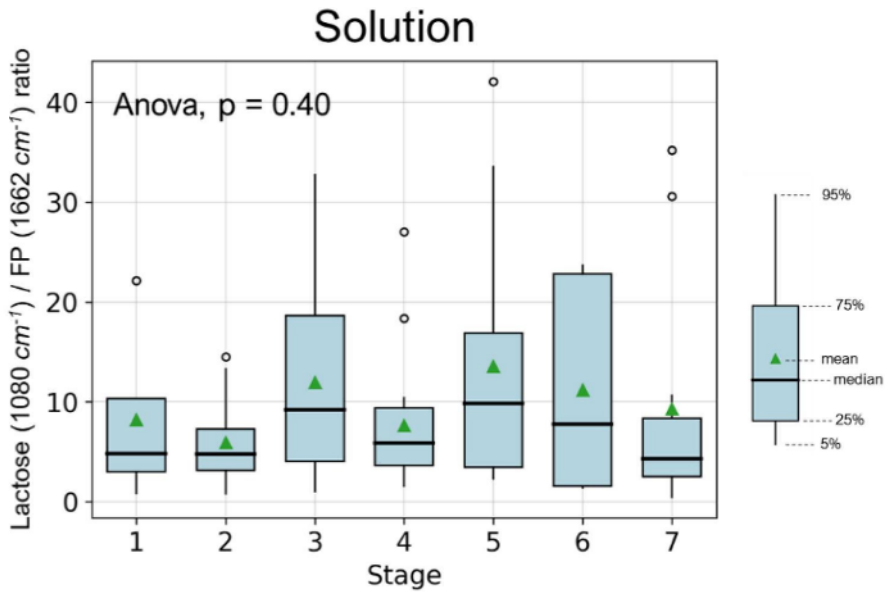


Solute composition in solution spray-dried particles is identical to that of the feed solution

FP: fluticasone propionate; SX: salmeterol xinafoate

Ratio of lactose to fluticasone propionate for particles collected from stages 1–7 of spray-dried powder for suspension





Stage	1	2	3	4	5	6	7
Mean	3.2	1.1	1.0	1.7	1.1	0.6	0.3
Median	1.9	0.8	0.9	1.3	0.7	0.3	0.2
n	18	24	17	26	24	16	12

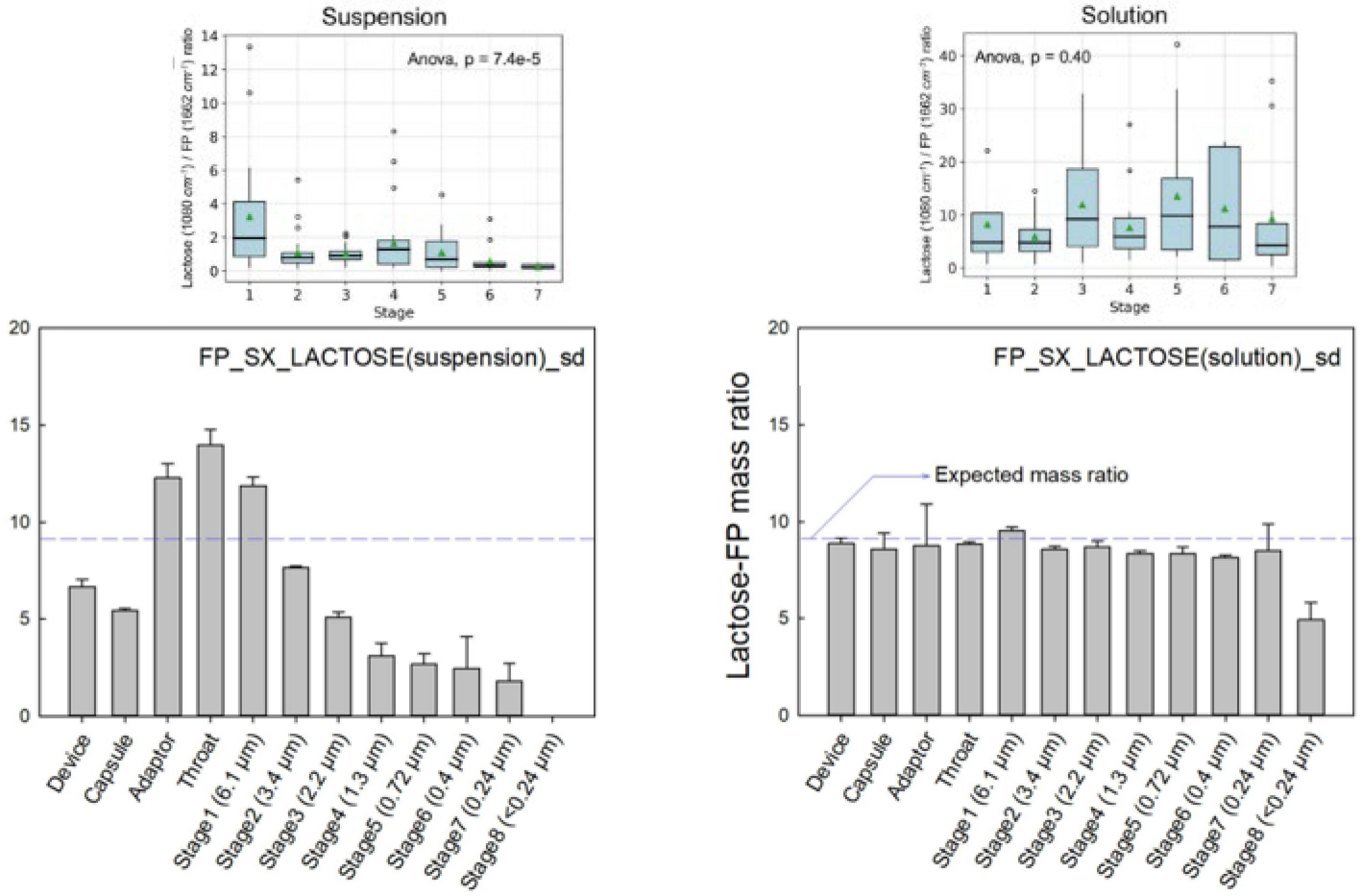
Stage	1	2	3	4	5	6	7
Mean	8.2	5.9	12.0	7.6	13.6	11.1	9.3
Median	4.8	4.8	9.2	5.9	9.8	7.8	4.3
n	5	12	20	16	12	8	11

Suspension formulation indicates a decrease in the average lactose-FP ratio by tenfold from 3.2 on stage 1 to 0.3 on stage 7

Solution formulation stages exhibit a relatively high lactose-FP ratio across all the stages with a minimum of 5.9 and maximum of 13.6 (9.9 ± 2.7)

FP: fluticasone propionate; SX: salmeterol xinafoate

IR peak ratio of lactose:FP (1080 cm^{-1}:1662 cm^{-1}) from each stage of the suspension and solution formulations closely follows the bulk lactose-FP mass ratio.

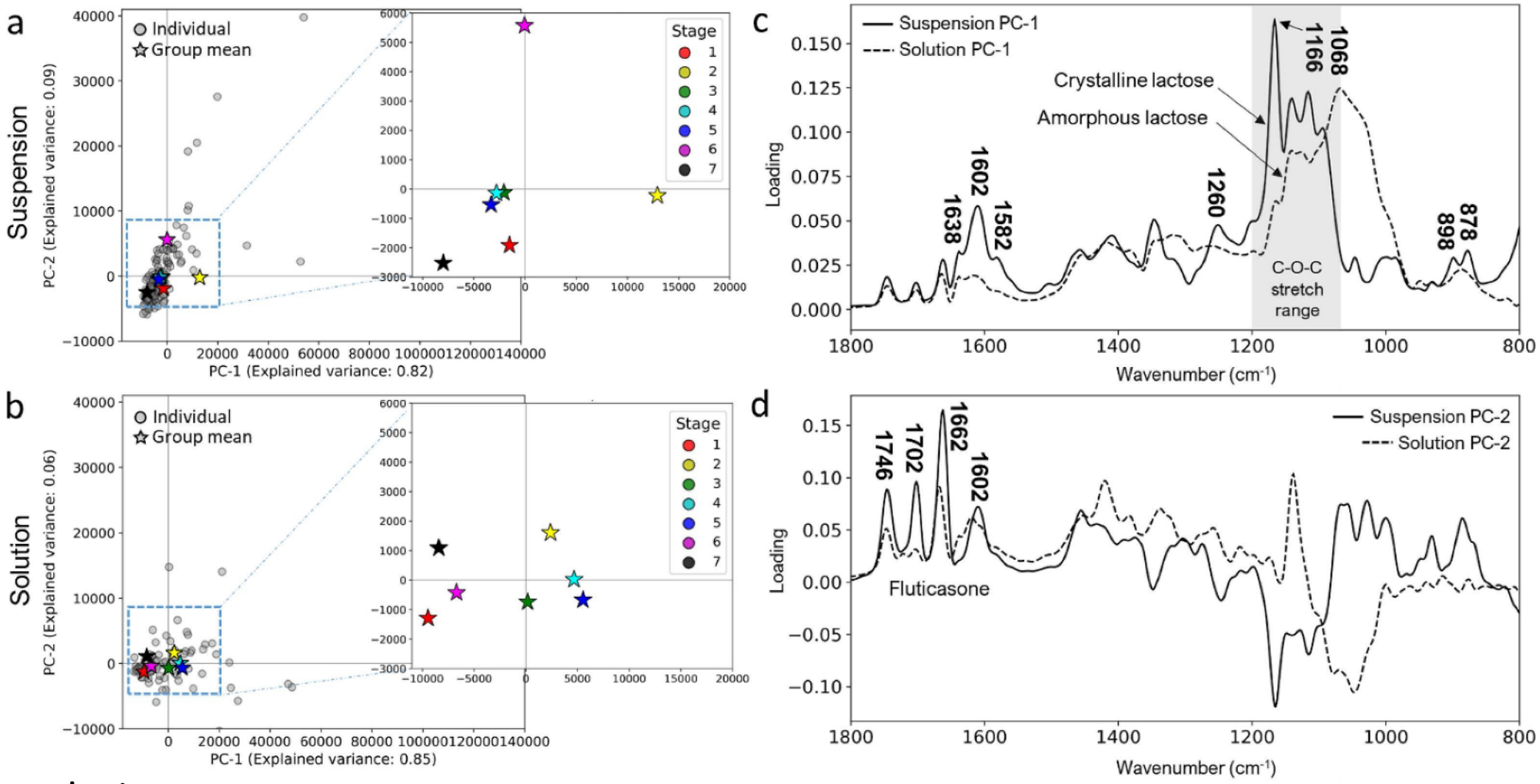


IR data ALSO indicates particle to particle dispersion in ratio that is not observed in bulk analysis

FP: fluticasone propionate; SX: salmeterol xinafoate

Principal component analysis indicates Lactose (LH300) Present in Crystalline form in Suspension Formulation and is Amorphous in Solution Formulation

In both formulations, variation largely results from changes in FP/lactose ratio



FP: fluticasone propionate

O-PTIR with Quantitative Morphology Analysis

A) Formulation Methods

Turbula Mixer

Buchi S-300 Spray dryer

B) Next Generation Impactor (NGI)

FP/SX/LH flow (60 L/min)

C) O-PTIR: mIRage-LS

Visible light

Detector

Tunable IR laser

D) Point Spectra

80 X 80 µm

FP Related Bands

SX Related Bands

LH Related Bands

E) Single Wavenumber Maps

1662 cm^{-1} FP

1559 cm^{-1} SX

1041 cm^{-1} LH

F) Summed Wavenumber Map

G) Colocalization Maps

20 µm

FP/SX/LH

FP/SX

FP/L

SX/L

H) Key Morphological Parameters

Area

O-PTIR Single Wavenumber Defined Region

Equivalent Area Diameter

Diameter defined by particle area

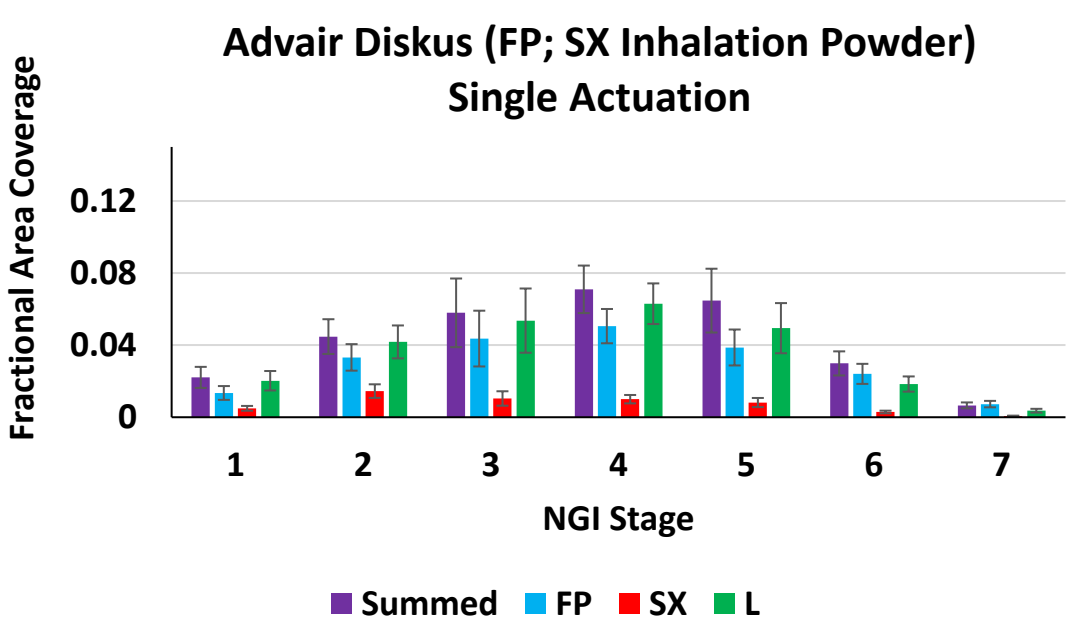
Integrated Intensity Volume

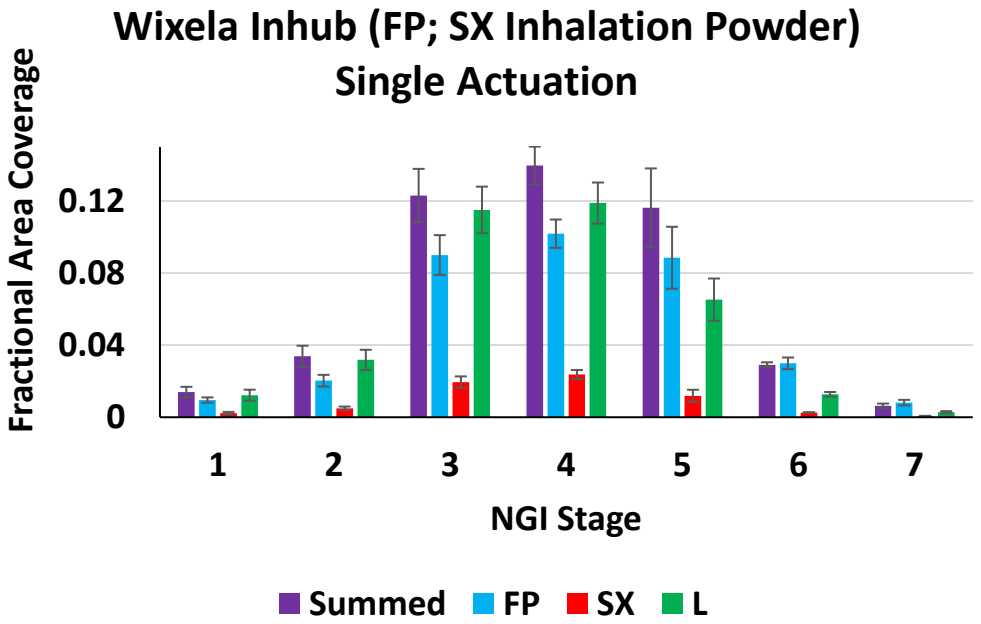
Volume sum of intensities at all particle pixels

FP: fluticasone propionate; SX: salmeterol xinafoate; LH or L: lactose monohydrate

Fractional Area Coverage of Particles on CaF_2 Surface

Strength: 0.1 mg/inh; EQ 0.05 mg base/inh (FP; SX)



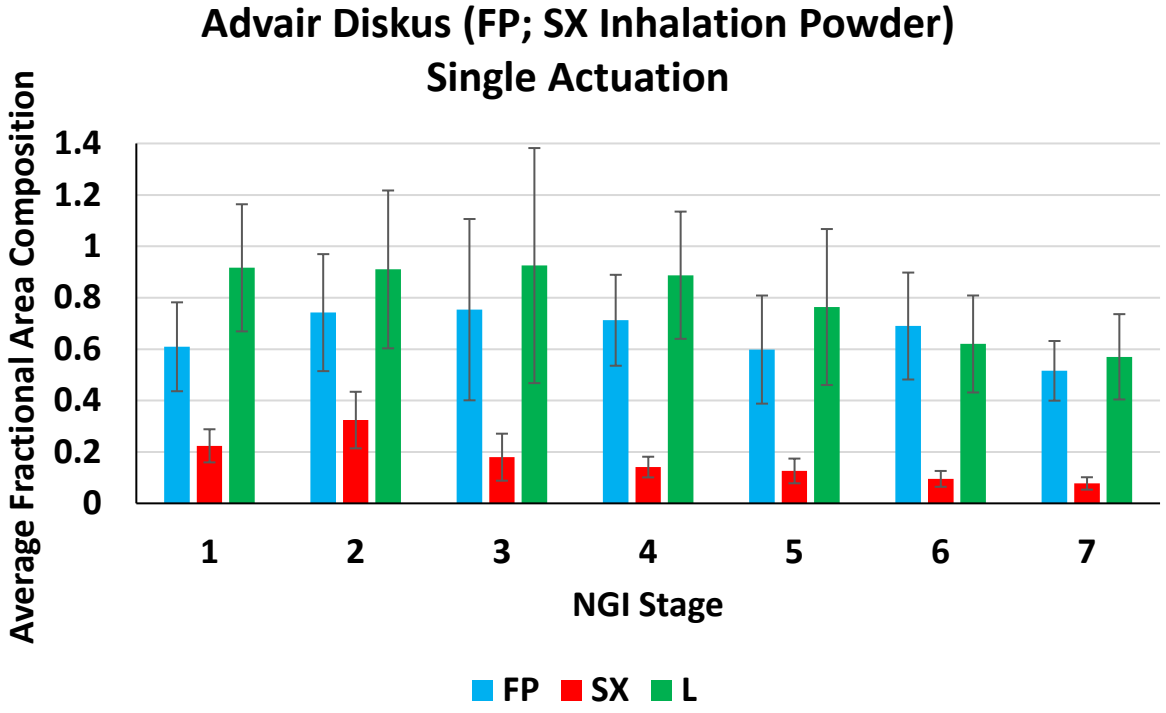


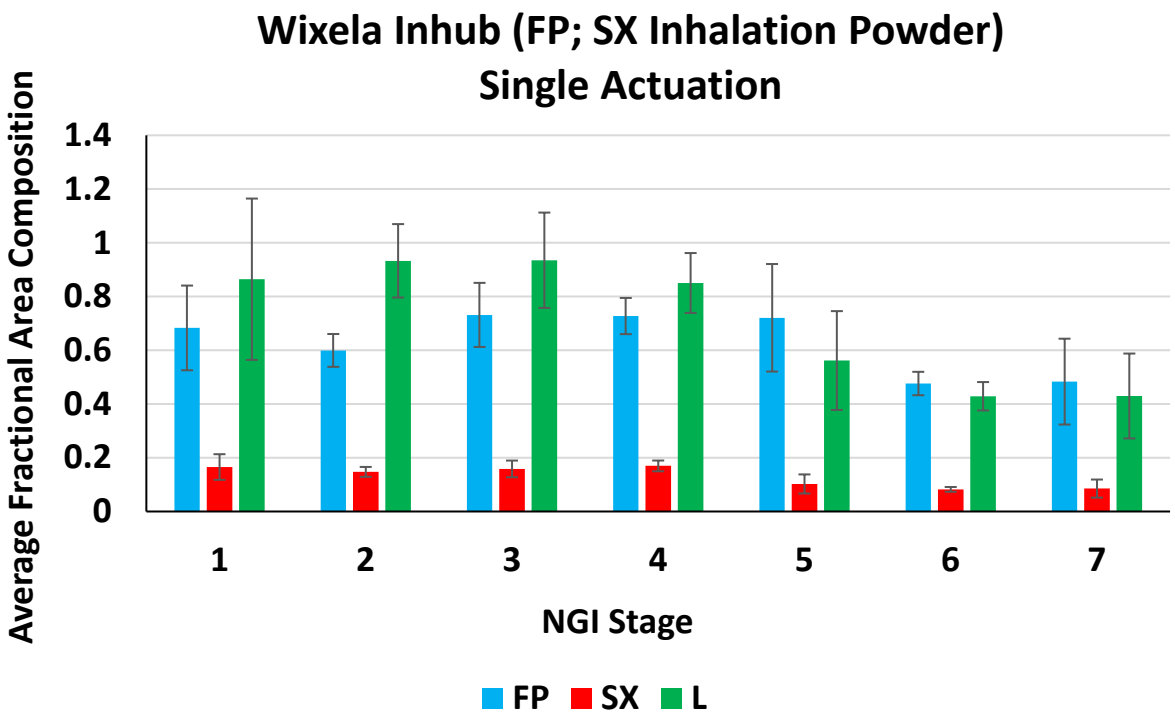
Advair Diskus (FP; SX inhalation powder) and Wixela Inhub (FP; SX inhalation powder) at 0.1 mg/inh; EQ 0.05 mg base/inh strength show similar total material deposited on the CaF_2 surface as measured by total surface area of material

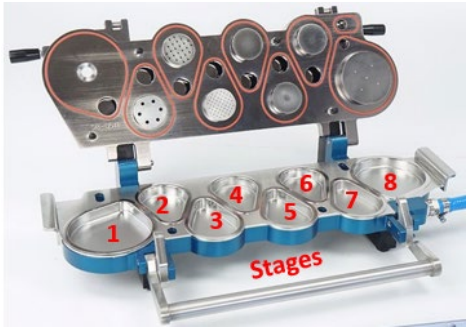
FP: fluticasone propionate; SX: salmeterol xinafoate; L: lactose monohydrate

Average Fractional Area Composition of Particles on CaF_2 Surface

Strength: 0.1 mg/inh; EQ 0.05 mg base/inh (FP; SX)





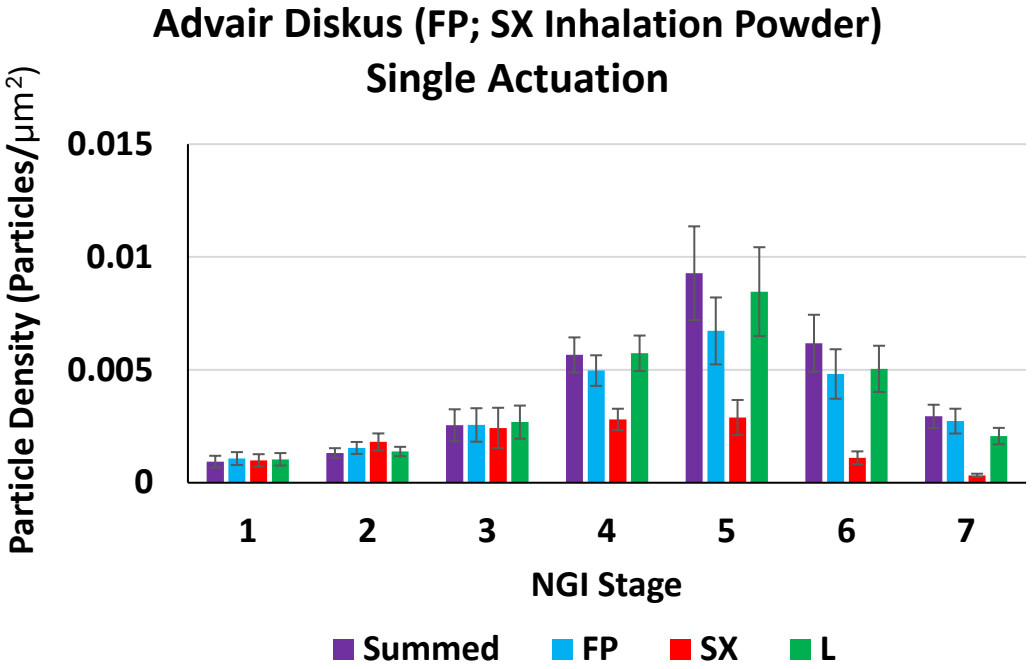


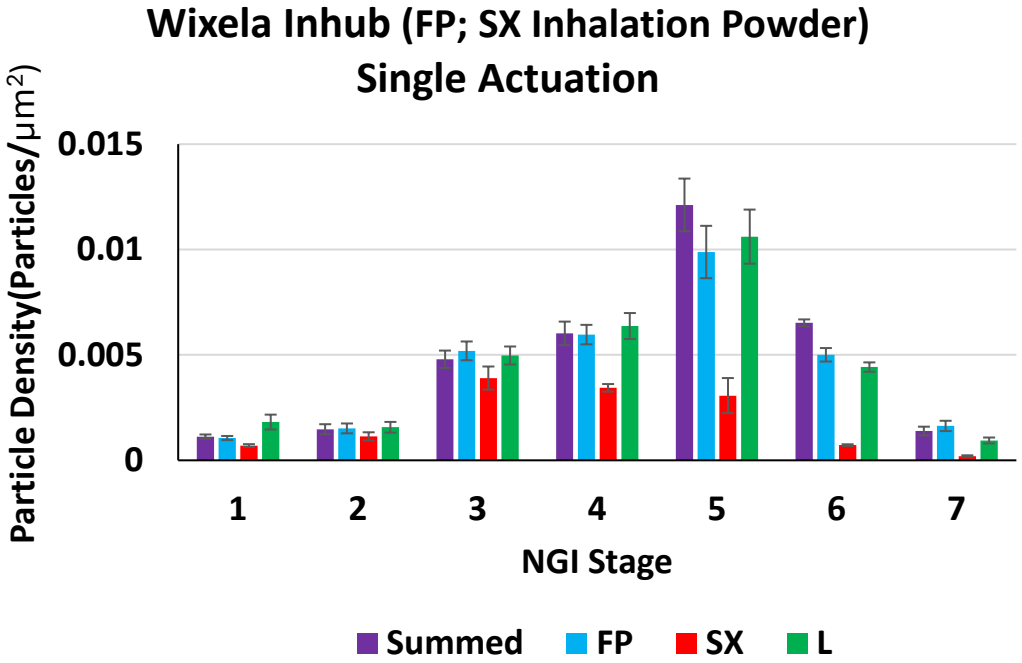
FP; SX inhalation powders, Advair Diskus and Wixela Inhub, at 0.1 mg/inh; EQ 0.05 mg base/inh strength are quite similar in fractional composition of the particles for both Fluticasone and Lactose

FP: fluticasone propionate; SX: salmeterol xinafoate; L: lactose monohydrate

Density of Particles on CaF_2 Surface

Strength: 0.1 mg/inh; EQ 0.05 mg base/inh (FP; SX)

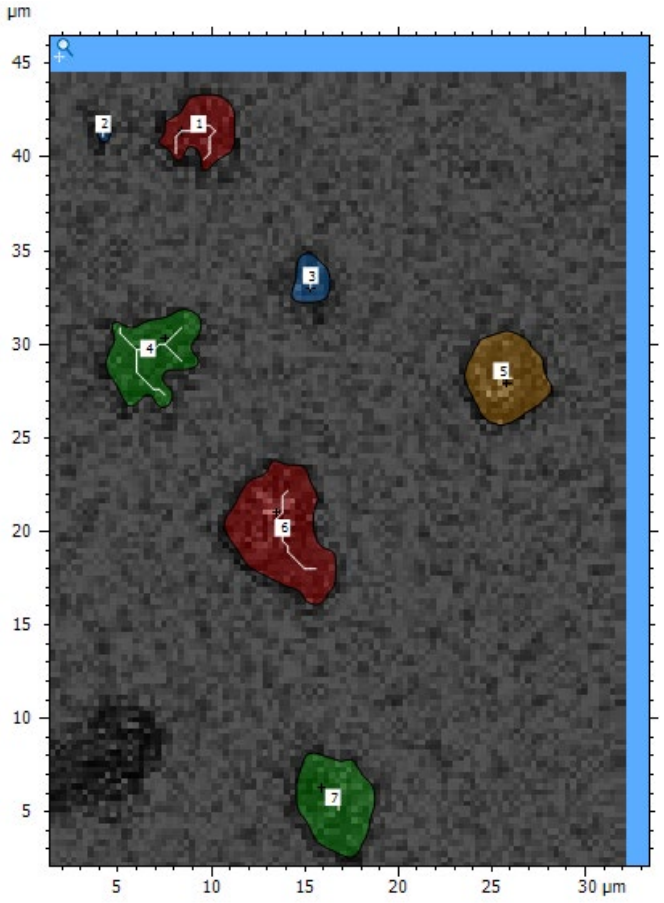




FP; SX inhalation powders, Advair Diskus and Wixela Inhub, at 0.1 mg/inh; EQ 0.05 mg base/inh strength show similar trend in particle density as a function of NGI stage

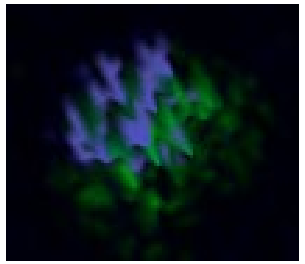
FP: fluticasone propionate; SX: salmeterol xinafoate; L: lactose monohydrate

Definitions of Particle Area and Diameter



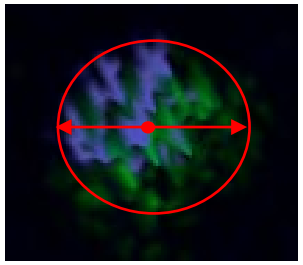
Area and Diameter Analysis for 7 Particle Case (1662 cm^{-1} image of Advair Diskus [FP; SX inhalation powder])

Spectroscopic Area



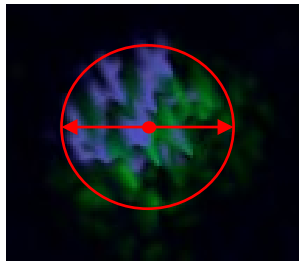
O-PTIR Single Wavenumber Defined Region

Spectroscopic Equivalent Area Diameter



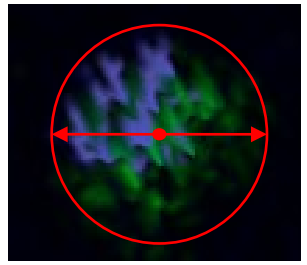
Diameter circle of equiv. area of particle

Spectroscopic Mean Diameter



Diameter defined by particle center of gravity

Spectroscopic Maximum Diameter



Diameter defined by longest rotation axis.

Individual results

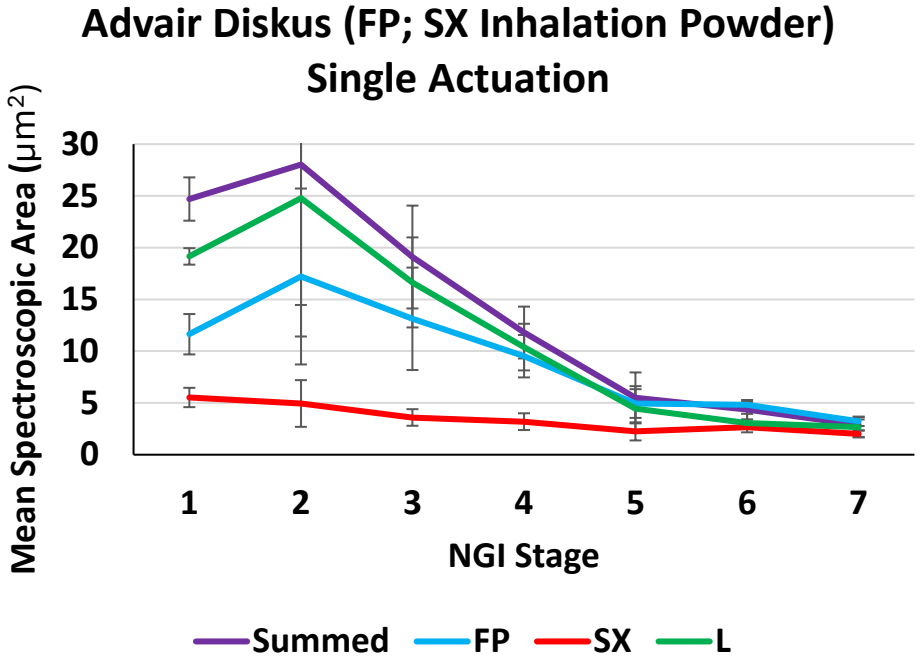
Parameters	Area	Equivalent diameter	Mean diameter	Maximum diameter
Unit	µm²	µm	µm	µm
Particle #1	10.78	3.705	3.525	4.216
Particle #2	0.8480	1.039	0.8963	1.315
Particle #3	4.150	2.299	2.157	2.645
Particle #4	17.30	4.694	4.502	5.787
Particle #5	16.41	4.571	4.404	5.023
Particle #6	27.92	5.962	5.739	7.568
Particle #7	17.13	4.670	4.503	5.859

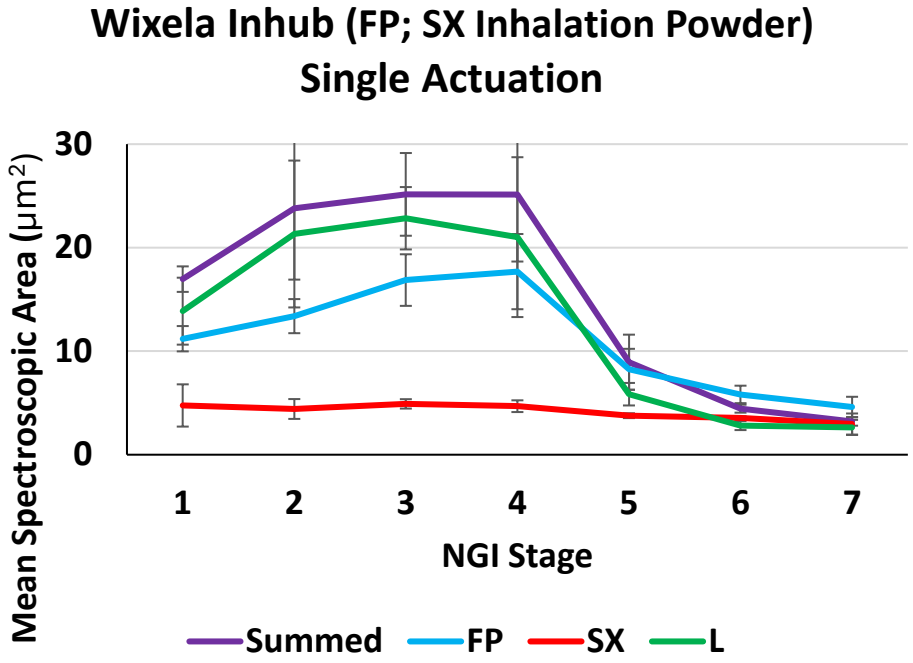
Global statistics

Mean	13.51	3.849	3.675	4.630

Mean Particle Area as Function of NGI Stage

Strength: 0.1 mg/inh; EQ 0.05 mg base/inh (FP; SX)

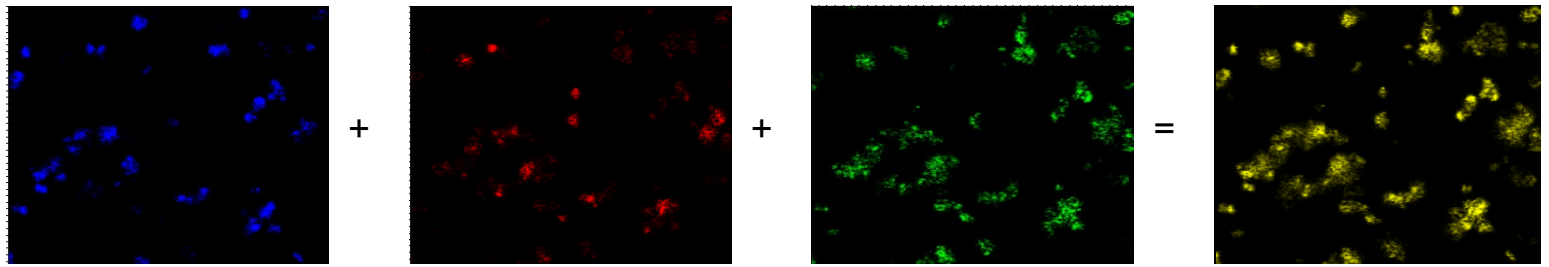




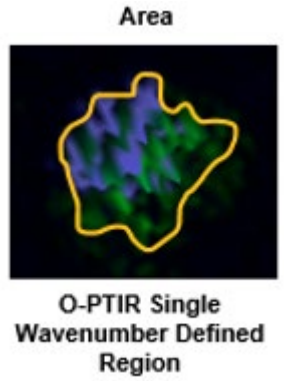
Aggregate particle size decreases with NGI Stage

Fluticasone and Lactose also decrease with NGI Stage

Salmeterol particle size constant!



FP SX L Summed

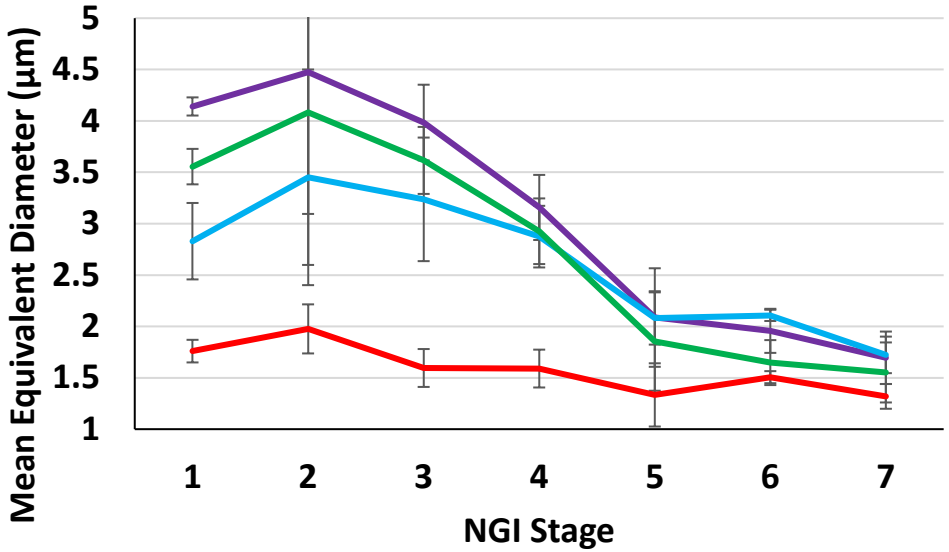


FP: fluticasone propionate; SX: salmeterol xinafoate; L: lactose monohydrate

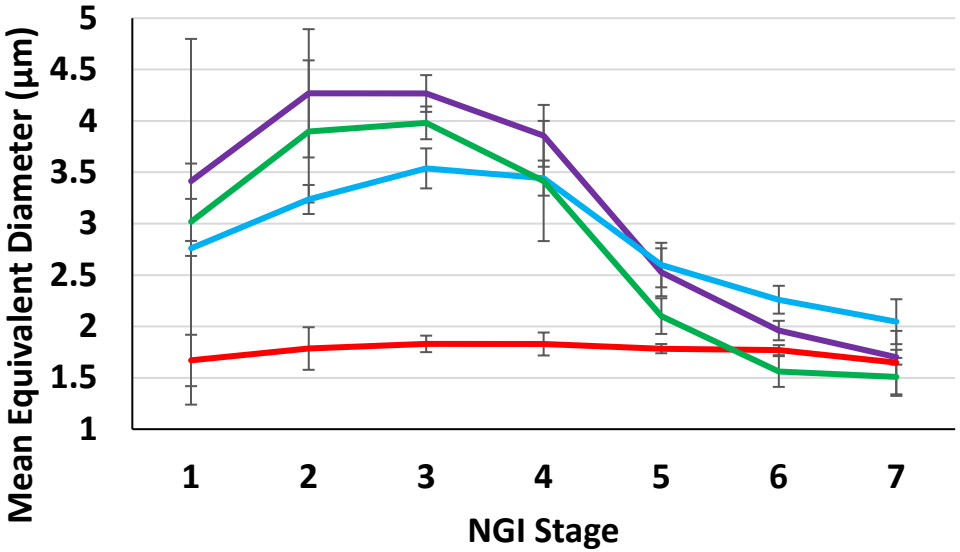
Mean Particle Equivalent Area Diameter as Function of NGI Stage

Strength: 0.1 mg/inh; EQ 0.05 mg base/inh (FP; SX)

Advair Diskus (FP; SX Inhalation Powder)
Single Actuation



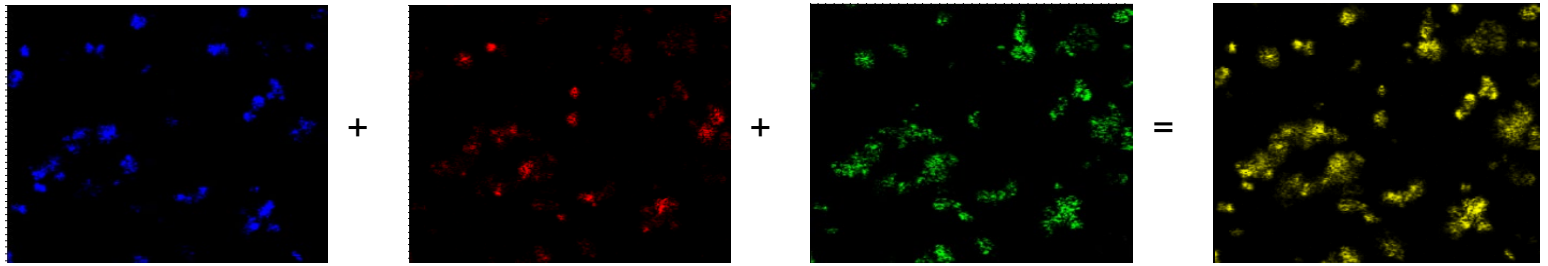
Wixela Inhub (FP; SX Inhalation Powder)
Single Actuation



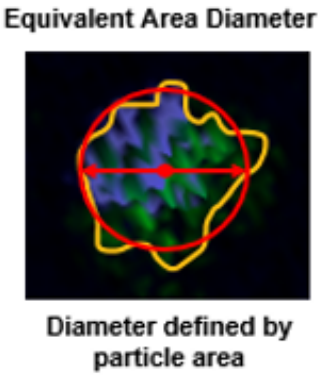
Aggregate particle size decreases with NGI Stage

Fluticasone and Lactose also decrease with NGI Stage

Salmeterol particle diameter is constant!



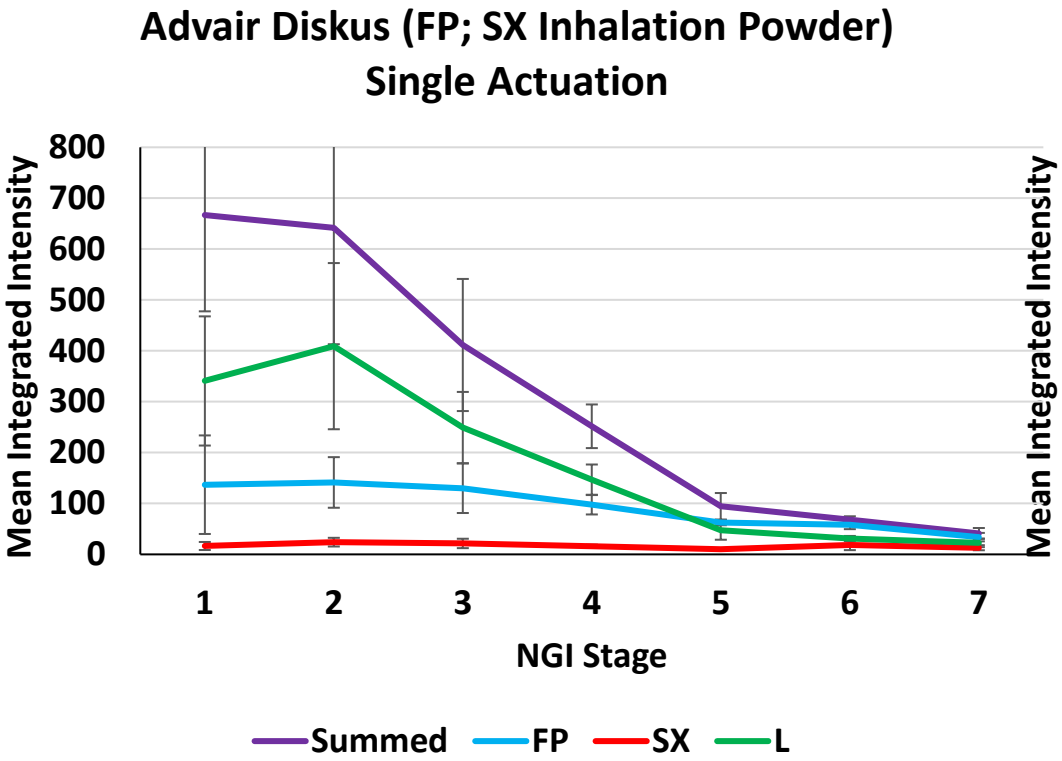
FP + SX + L = Summed

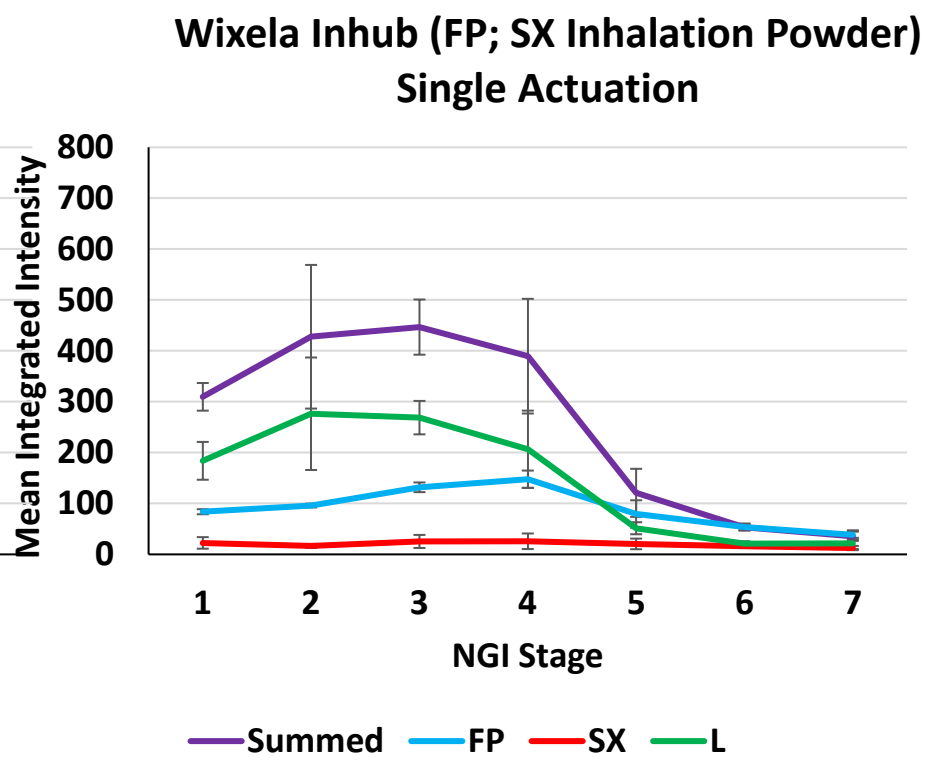


FP: fluticasone propionate; SX: salmeterol xinafoate; L: lactose monohydrate

Mean Integrated Intensity as Function of NGI Stage

Strength: 0.1 mg/inh; EQ 0.05 mg base/inh (FP; SX)

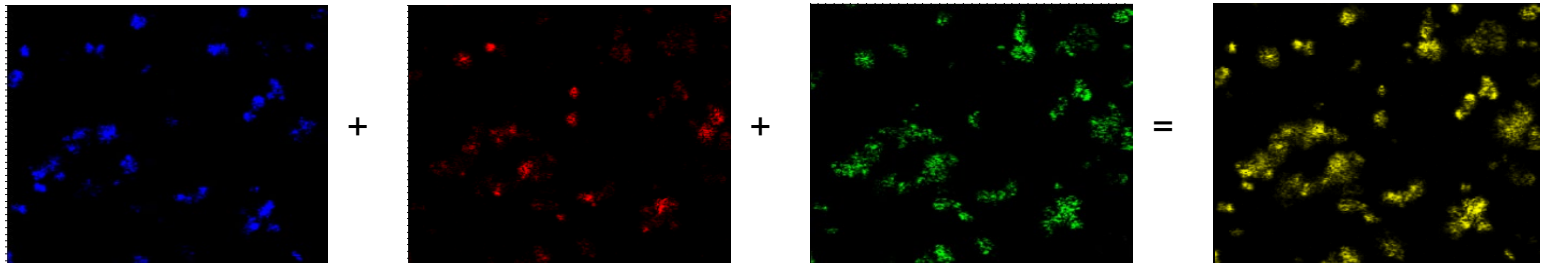




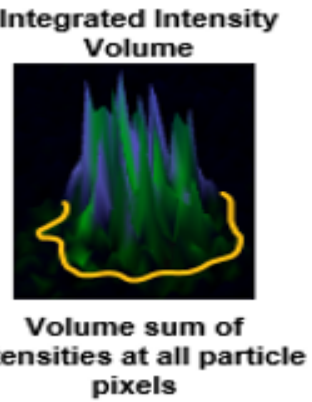
Aggregate particle volume decreases with NGI Stage. AD volume appears larger than WI for Stages 1-3.

Lactose and Fluticasone volume decrease with NGI stage

Salmeterol particle volume is constant!



FP SX L Summed

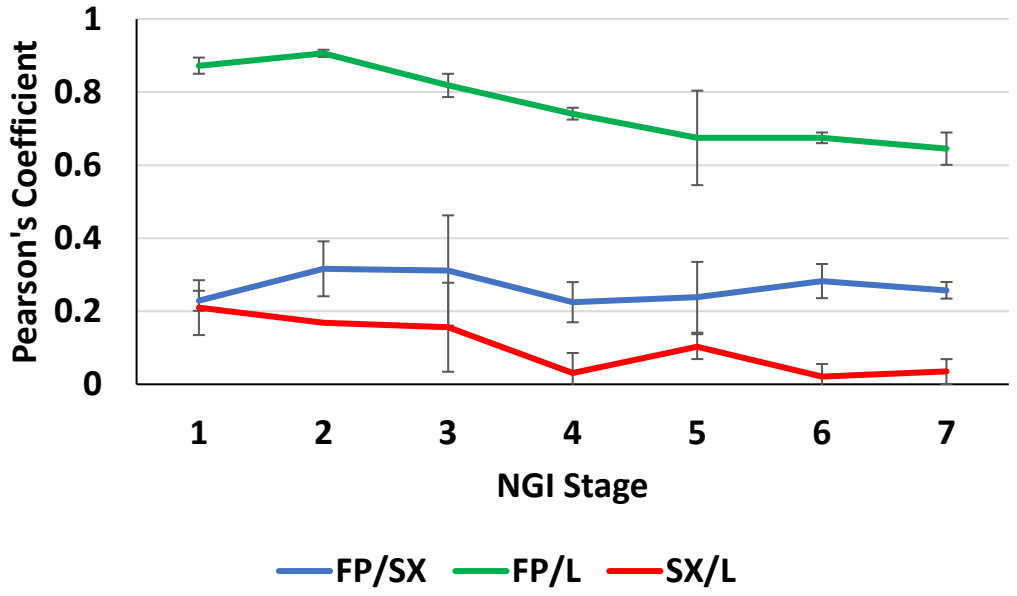


FP: fluticasone propionate; SX: salmeterol xinafoate; L: lactose monohydrate

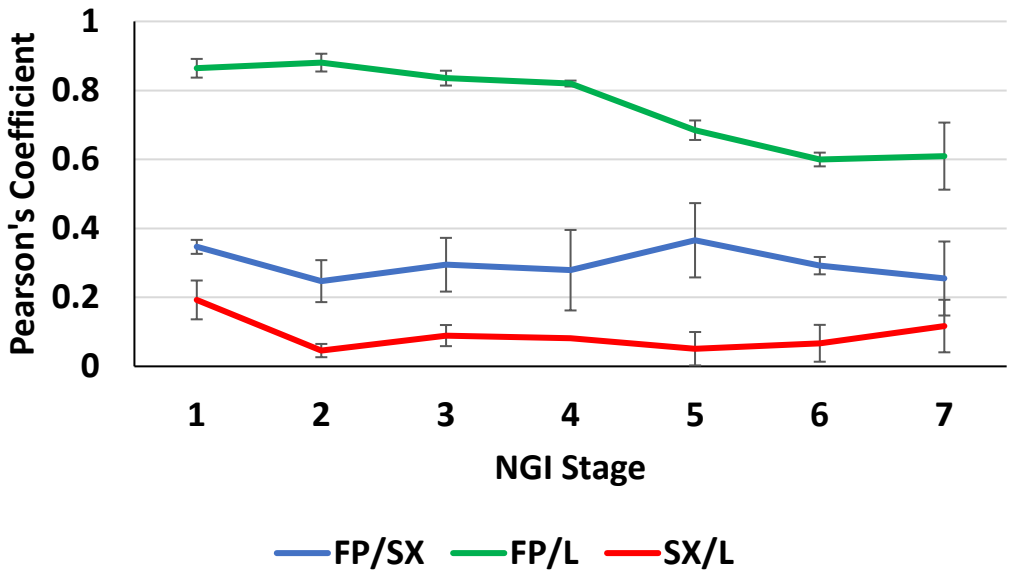
Pearson Coefficient Colocalization Analysis of FP; SX Inhalation Powders, Advair Diskus and Wixela Inhub, as function of NGI stages 1-7

Strength: 0.1 mg/inh; EQ 0.05 mg base/inh (FP; SX)

Advair Diskus (FP; SX Inhalation Powder) Single Actuation



Wixela Inhub (FP; SX Inhalation Powder) Single Actuation



The Pearson correlation coefficient indicates the strength and direction of a linear relationship between two continuous variables. +1 Positive Linear Correlation, -1 negative linear correlation, 0 no correlation

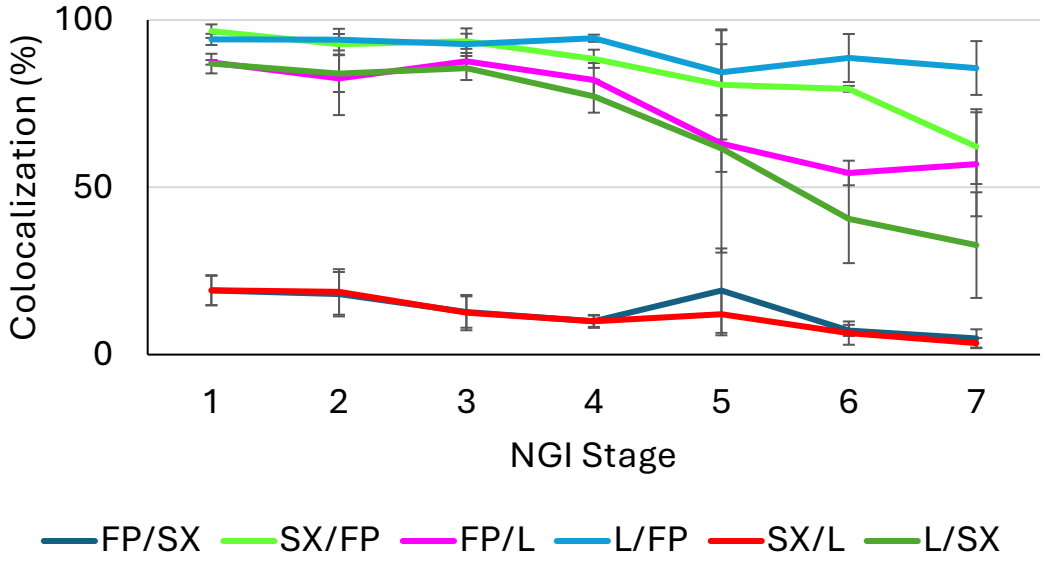
- FP and L exhibit a strong, positive correlation for both formulations
- FP and SX exhibit a weak, positive correlation for both formulations
- There is little correlation SX and L

FP; SX inhalation powders, Advair Diskus and Wixela Inhub (0.1 mg/inh; EQ 0.05 mg base/inh), show similar drug/drug and drug/excipient colocalization properties.

Manders Coefficient Colocalization Analysis of FP; SX Inhalation Powders, Advair Diskus and Wixela Inhub, as function of NGI stages 1-7

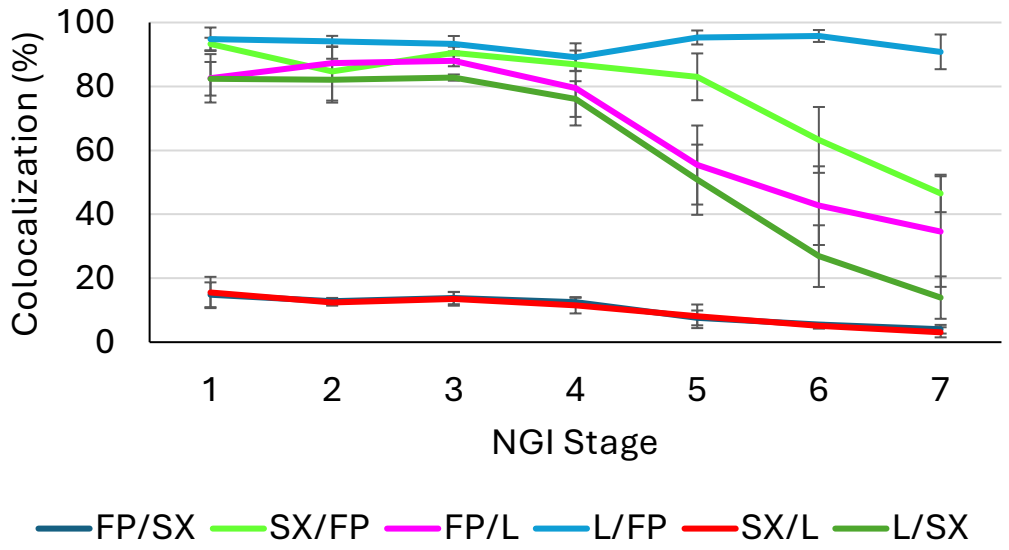
Strength:
0.1 mg/inh;
EQ 0.05 mg base/inh
(FP; SX)

Advair Diskus (FP; SX Inhalation Powder)
Single Actuation



FP/SX SX/FP FP/L L/FP SX/L L/SX

Wixela Inhub (FP; SX Inhalation Powder)
Single Actuation



FP/SX SX/FP FP/L L/FP SX/L L/SX

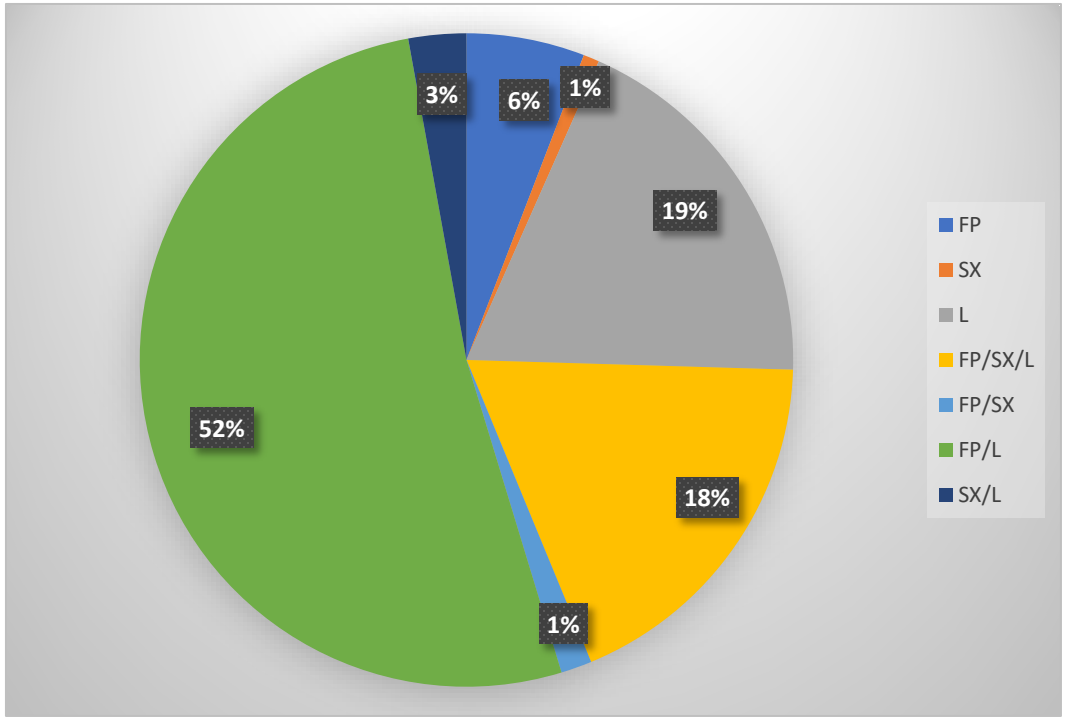
The Manders correlation coefficient indicates the degree of spatial colocalization between a pair of Infrared signals. A value of 1 indicates complete colocalization and a value of 0 indicates no colocalization.

- L is highly spatially localized with FP
- FP is highly spatially localized with L for larger particles and less so for smaller particles
- SX is highly spatially localized with FP
- FP is not spatially localized with SX (there is far more FP in formulation)
- SX is not spatially localized with L

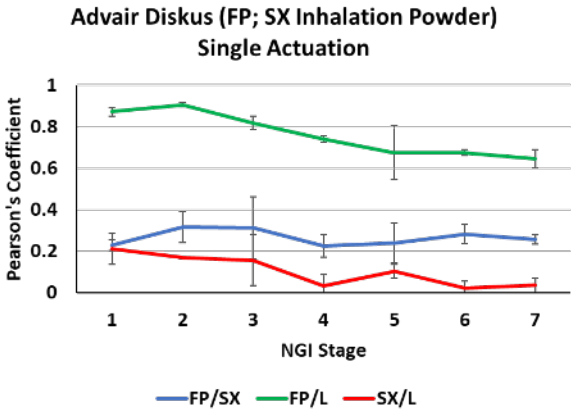
FP; SX inhalation powders, Advair Diskus and Wixela Inhub (0.1 mg/inh; EQ 0.05 mg base/inh) show similar drug/drug and drug/excipient spatial colocalization properties

MDRS and O-PTIR Comparison: NGI Stage 2

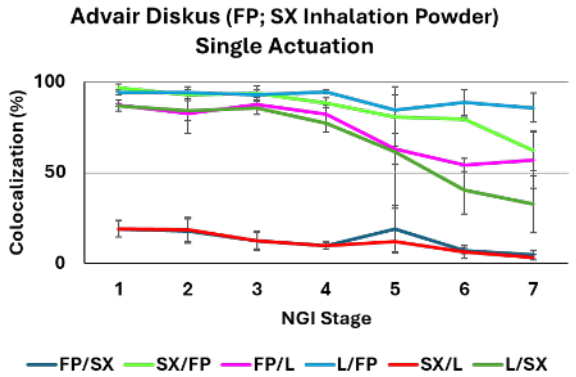
Single Actuation, NGI Stage 2



Pearson Coefficient



Manders Coefficient



Consistency:

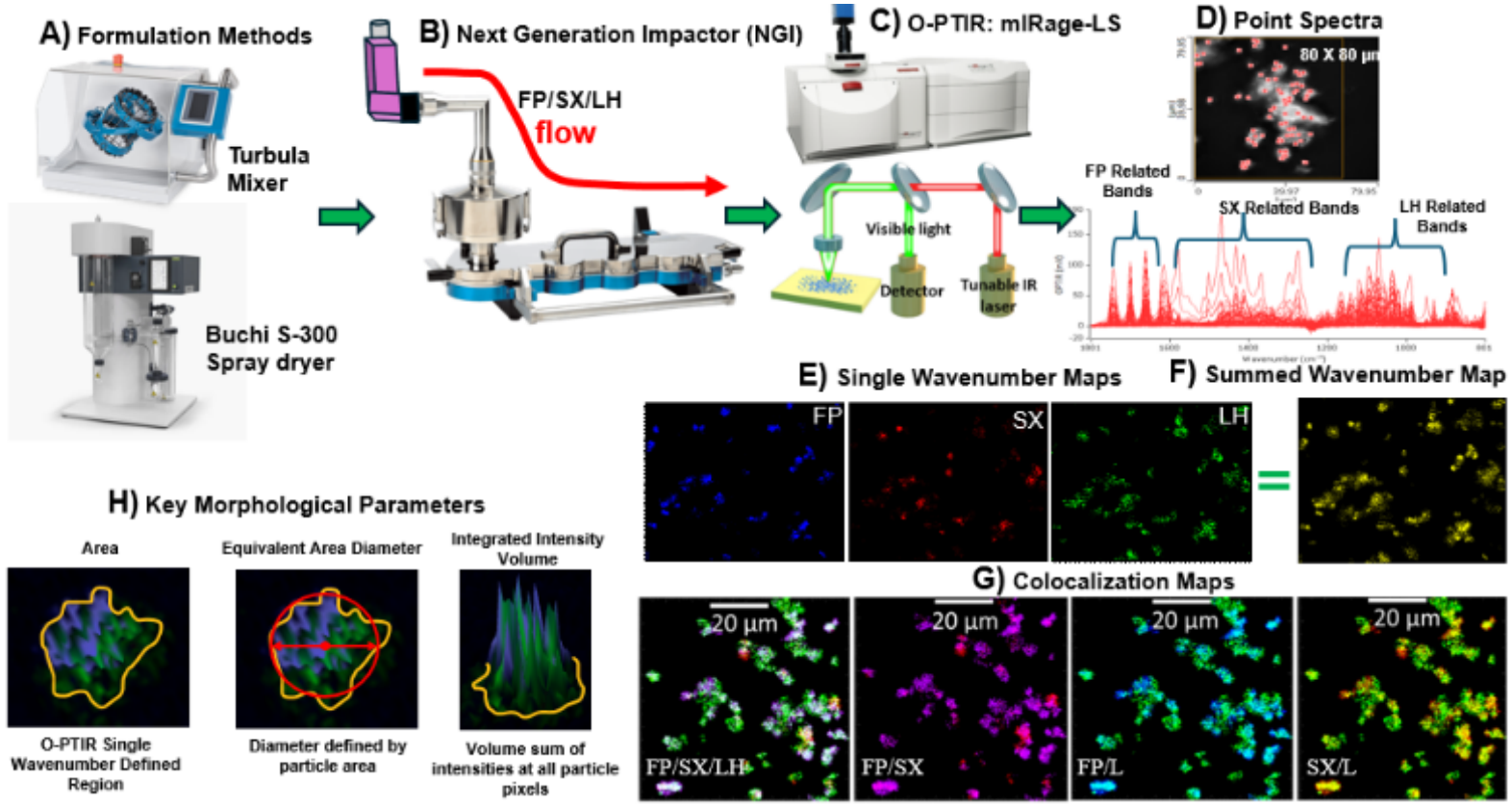
FP/L as major component of aggregates
SX/L as minor component of aggregates
FP/SX as minor component of aggregates

Note that O-PTIR also indicates that SX most often accompanied by FP consistent with Mass Spectroscopy reports

FP: fluticasone propionate; SX: salmeterol xinafoate; L: lactose monohydrate

Farias G, Ganley WJ, Price R, Conti DS, Mangal S, Bielski E, Newman B, Shur J., *Pharmaceutical Research*. **2024** Oct;41(10):2015-29.

O-PTIR with Quantitative Morphology Analysis



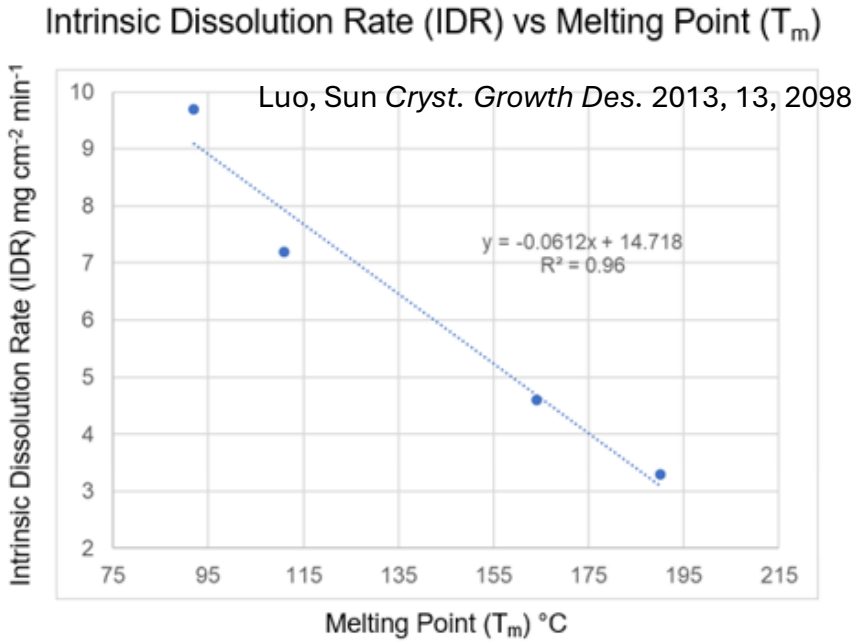
- Quantitative evaluation of area and diameter at the particle level for every component and the aggregate particles
- Quantitative evaluation of component colocalization
- Detailed morphological comparison of Brand Name and Generic Formulations

All for a Single Inhaler Actuation and Measured on Lung Relevant Emitted Dose

The Importance of Melting Points

- Criterion of purity
  - *Sensitive indicator of mixture for aerosol formulations*
- Strongly correlated with water solubility (thermodynamic)
  - *Sensitive indicator of dissolution rate? (kinetic)*
- Can we evaluate distribution of melting points for aerosol formulations?
  - *Role of distribution in formulation dissolution rate?*
  - *Particles and subparticle differences?*
- Importance of controlling melting points in aerosol formulation for tuning therapeutic index?

A powerful new tool in support of formulation design and product development of inhalation powders?



Intrinsic dissolution rate (IDR) versus melting point (T_m) for pyrazine carboxamide and co-crystalline mixtures with malonic, succinic, and glutaric acids

Kim, Grant *Int. J. Pharm*. 1989, 57, 117.
Teleki, Nylander, Bergstrom *Pharmaceutics* 2020, 12, 493.
Hanfeef, Chadha *J. Pharm. Sci.* 2019, 108, 3792.

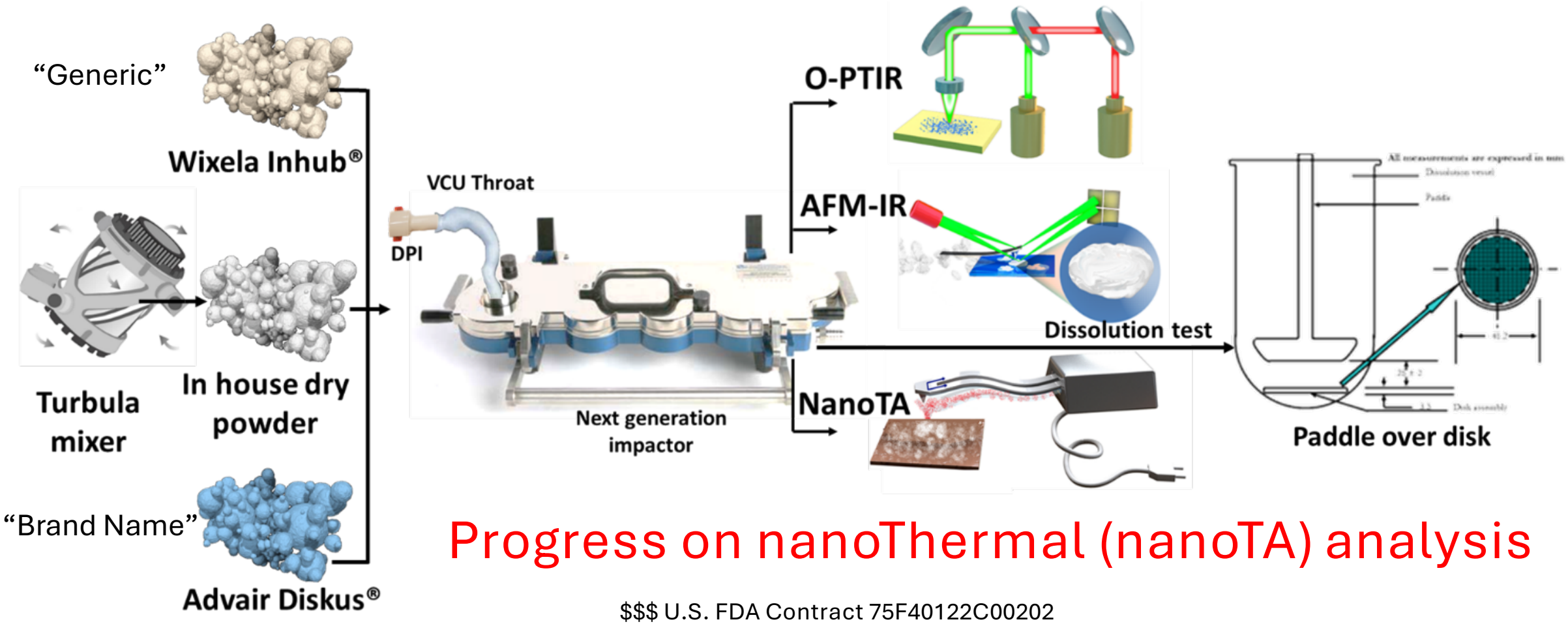
Katritzky, Jain, Lomaka, Petrukhin, Maran, Karelson
Crystal Growth & Design 2001, *1*, 4, 261–265

Assessing the Relationships of

- Particle compositions
- Melting points
- Particle mechanics

as a function of

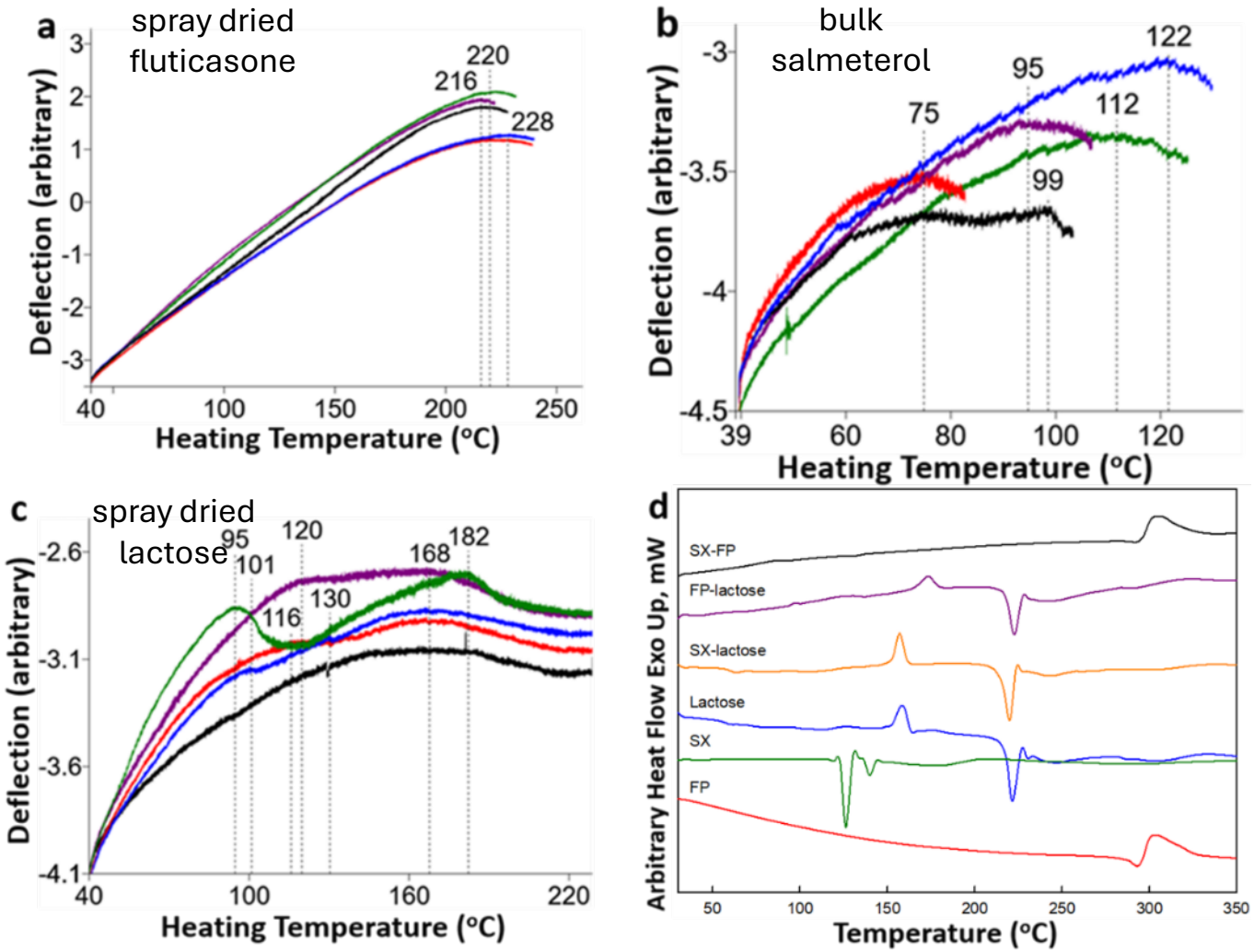
- Aerodynamics
- Dissolution Rate
- Bulk composition



Progress on nanoThermal (nanoTA) analysis

$$$ U.S. FDA Contract 75F40122C00202
Identification of Drug Distribution in Aerosols A Nanospectroscopy and NanoThermal Analysis

NanoTA thermal ramps and DSC thermograms of drugs and lactose excipient

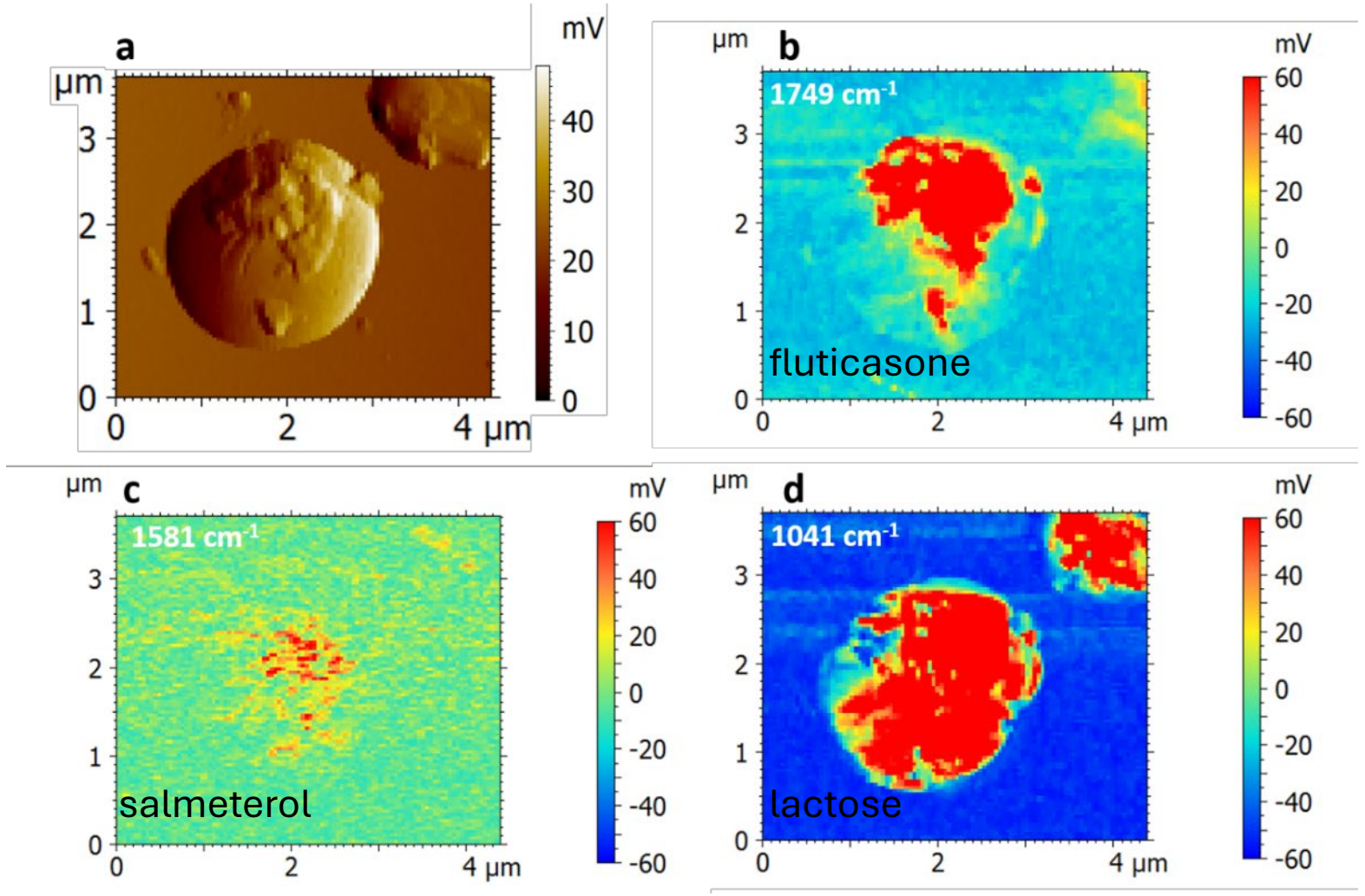


Differential Scanning Calorimetry (DSC)

- Thermal properties of spray dried fluticasone provide narrow melting point (MP) range
- Thermal properties of bulk, as-received salmeterol show substantial MP variation including presence of two known crystalline forms
- Thermal properties of lactose show evidence for glass transition (first peak 95-130 °C) and MP (168-182 °C)

Zhang, Khanal, Chan, Banaszak Holl Int. J. *Pharm. Res.* 2024, 656, 124116.

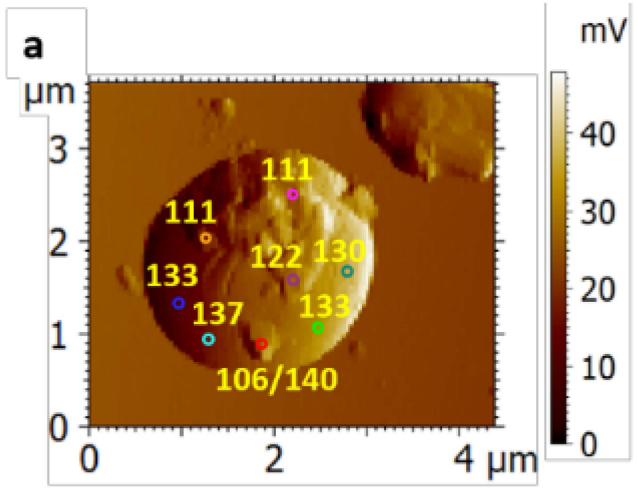
AFM-IR chemical maps acquired from an individual aerosol particle of a spray dried powder containing fluticasone propionate, salmeterol xinafoate and lactose collected on stage 5 of NGI.

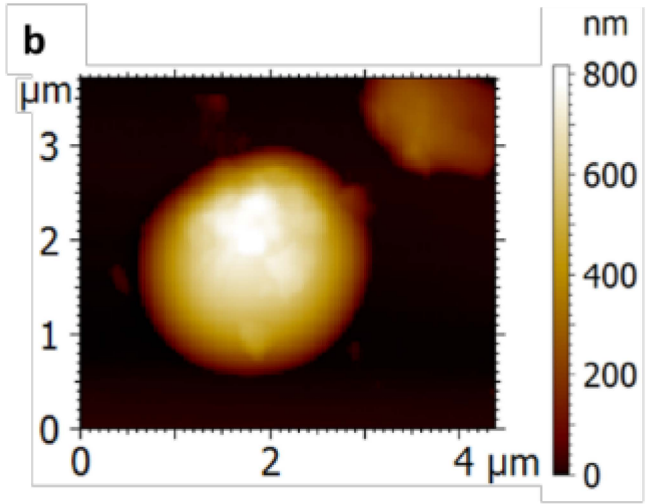


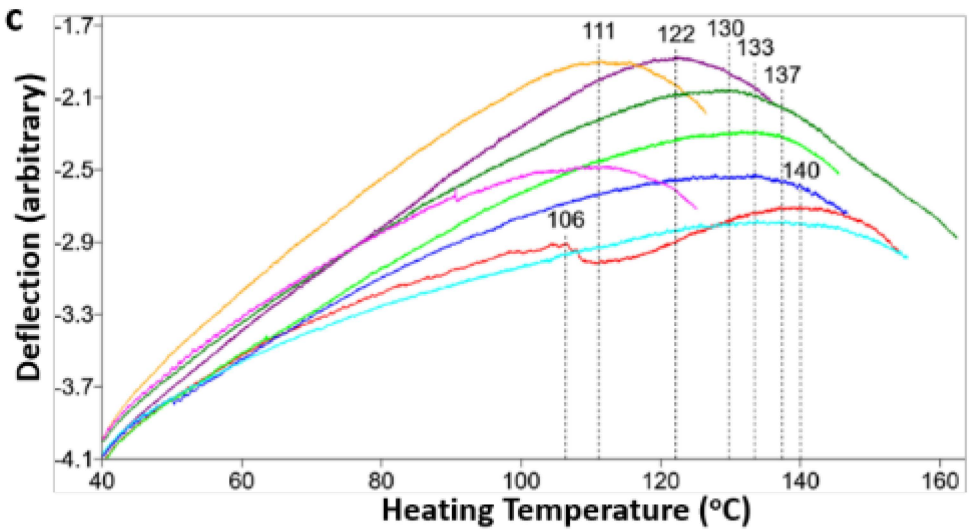
1:7:70 by weight salmeterol:fluticasone:lactose

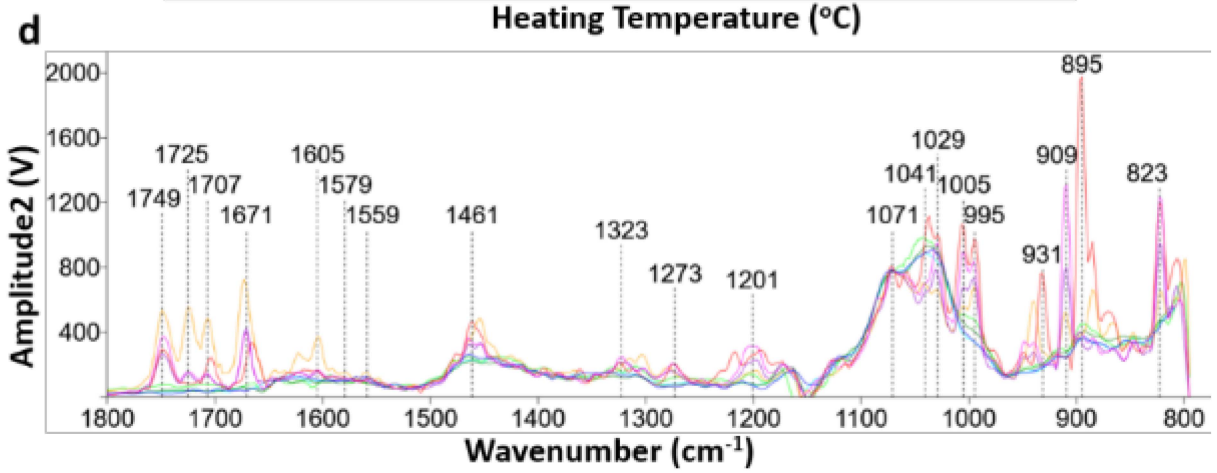
similar to Advair Diskus (fluticasone propionate; salmeterol xinafoate inhalation powder)

NanoTA thermal ramps and AFM-IR spectra acquired from different locations of an individual aerosol particle of a spray dried powder containing fluticasone propionate, salmeterol xinafoate and lactose collected on stage 5 of NGI.



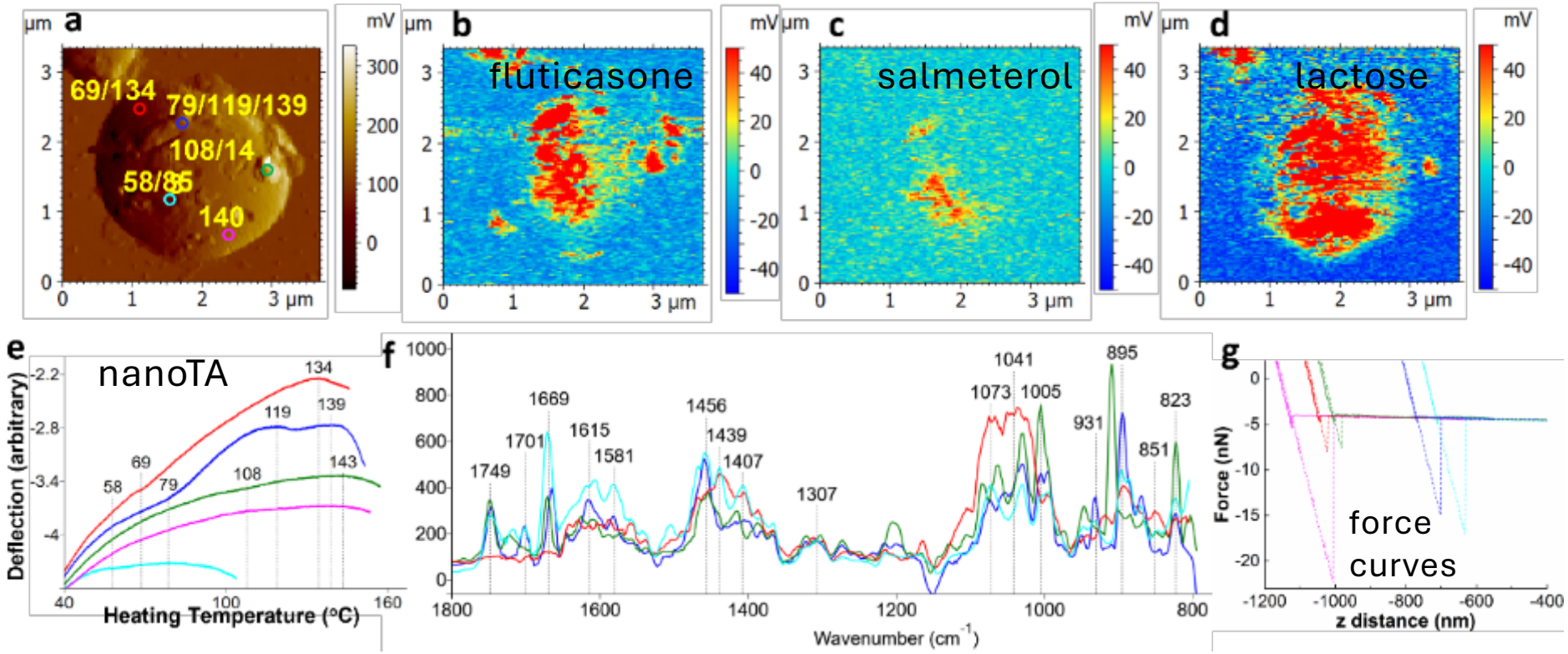






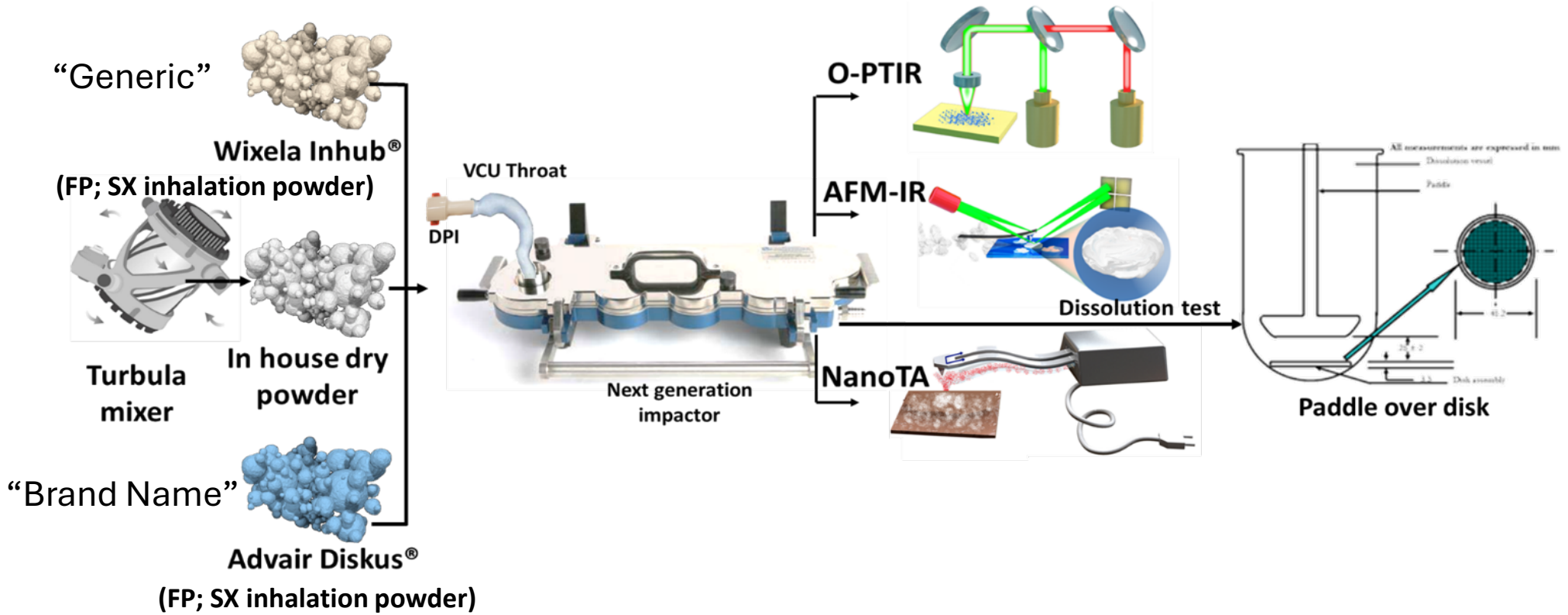
- T_m varies with local composition in particle
- Presence of drug lowers T_m as compared to lactose alone
- Smooth regions are lactose as indicated by spectra and T_m
- Rough regions are drugs as indicated by spectra and T_m

NanoTA thermal ramps, AFM-IR chemical maps, AFM-IR spectra and force curves acquired from an individual aerosol particle of a spray dried powder containing increased SX content collected on stage 5 of NGI (FP:SX:lactose = 1:2:23 by weight).



- Substantial decrease in MP for regions containing fluticasone, salmeterol, and lactose
- Lactose only regions of particle often exhibit MP for pure lactose
- Some “Lactose only” region do show decrease in MP – limited spectroscopic sensitivity?
- Greatest adhesion shown for lactose only regions; less adhesion shown for drug regions
  - Unmodified gold-coated tip

Future Directions



- Ongoing study for FDA using combination of Spectroscopic, Thermal, and Mechanical data to evaluate Generic vs Brand Name aerosol drug formulations
- Future Studies optimizing new formulations by evaluating therapeutic index as a function of Spectroscopic, Thermal, and Mechanical data