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Accurate aerosol dosimetry predictions using in silico and in vitro approaches for risk assessment methods

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PETA Science Consortium
Advancing 21st Century Toxicology

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Outline

- Introduction
  - Exposure and inhalation aerosol dosimetry
- Physicochemical aerosol characterization
  - Multispecies liquid aerosols
- In vitro exposure research
  - Bridging to in vivo/PBPK modeling
- In silico dosimetry models development
  - Aerosol inhalation and deposition predictions
- Summary

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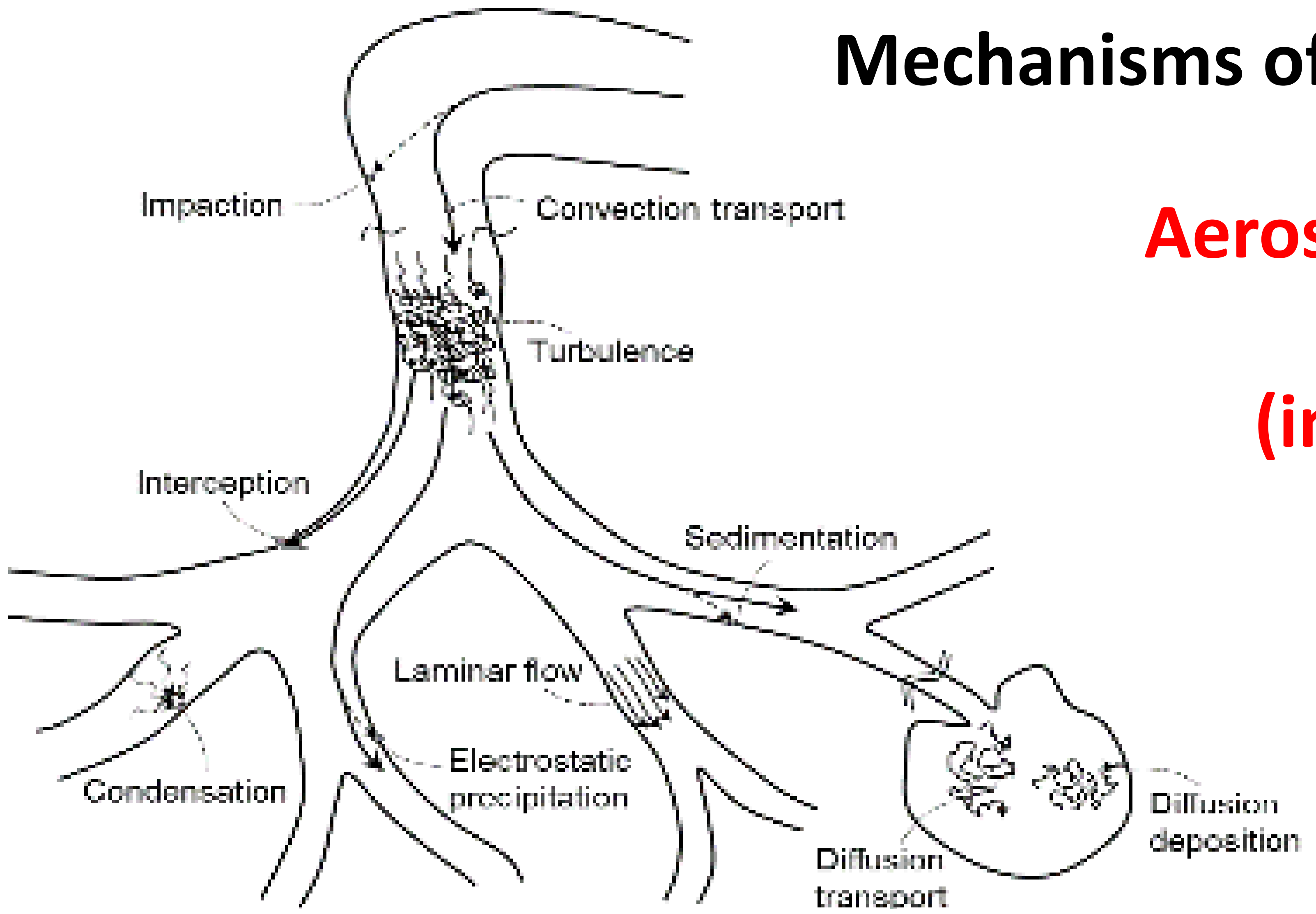
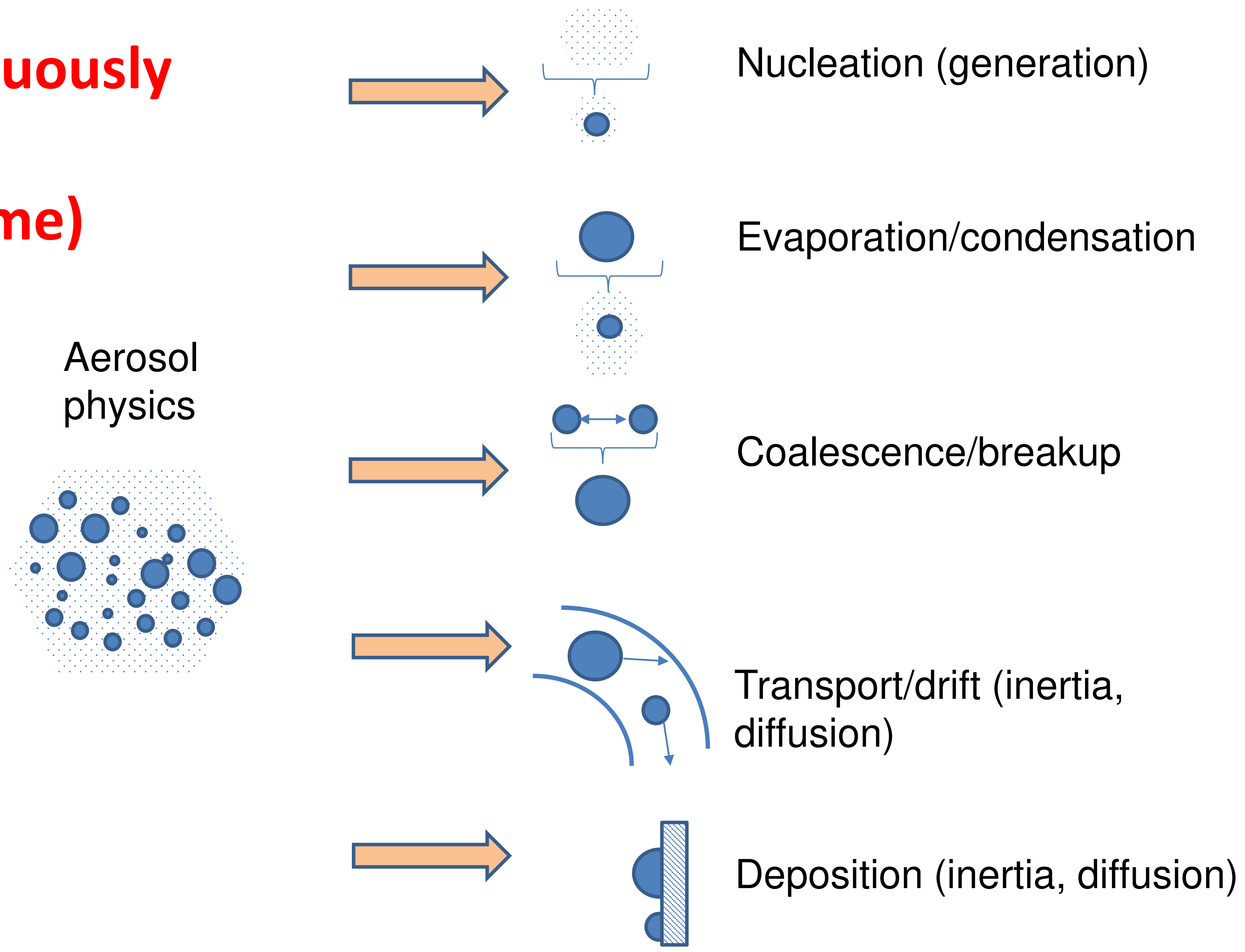
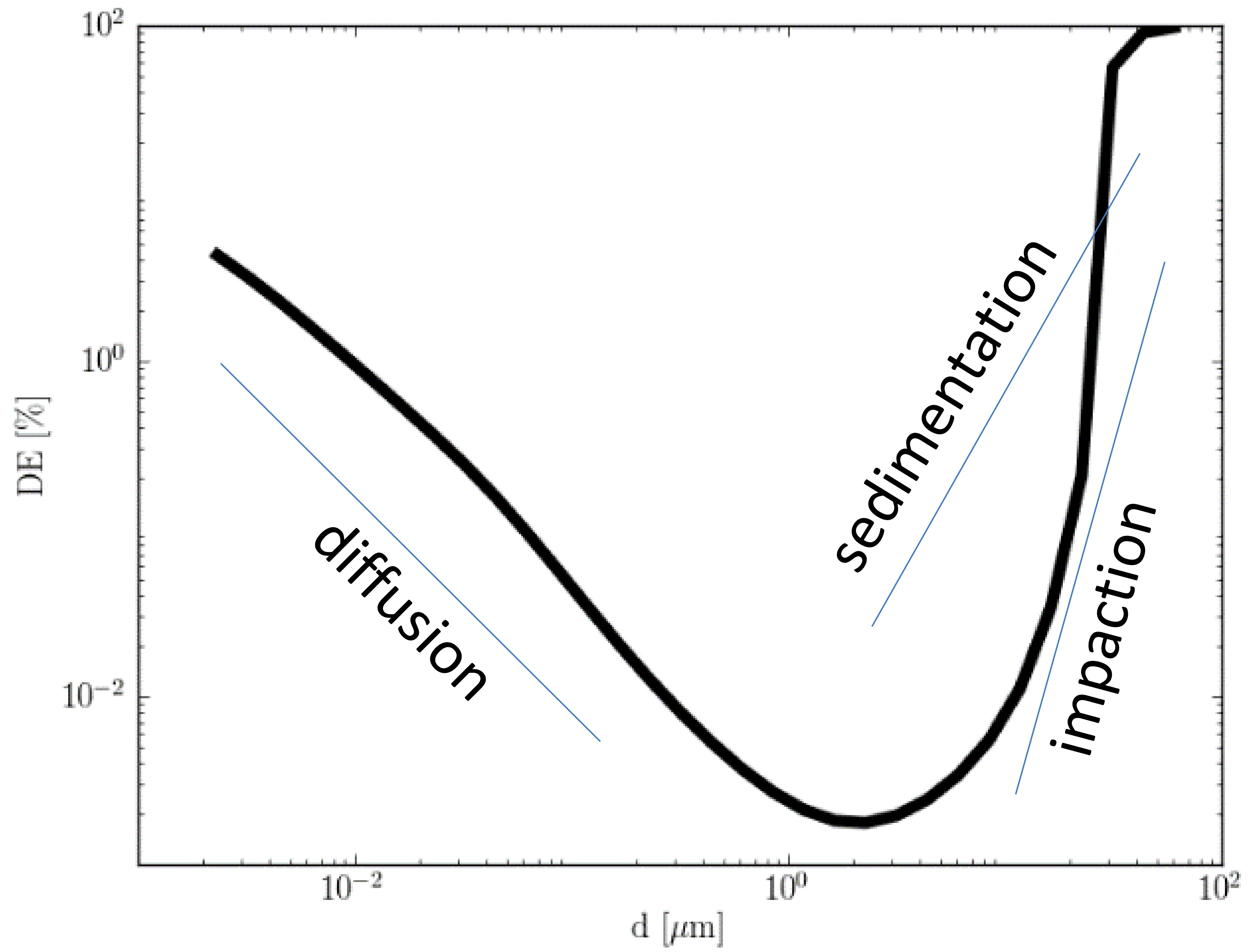
Aerosol lung delivery

Mechanisms of aerosol transport and evolution in the respiratory tract



Aerosols are continuously evolving (in space and time)





Deposition efficiency (DE) is driven by physical mechanisms:

$$DE = \frac{\text{Number of depositing particles}}{\text{Number of particles entering the geometry}}$$

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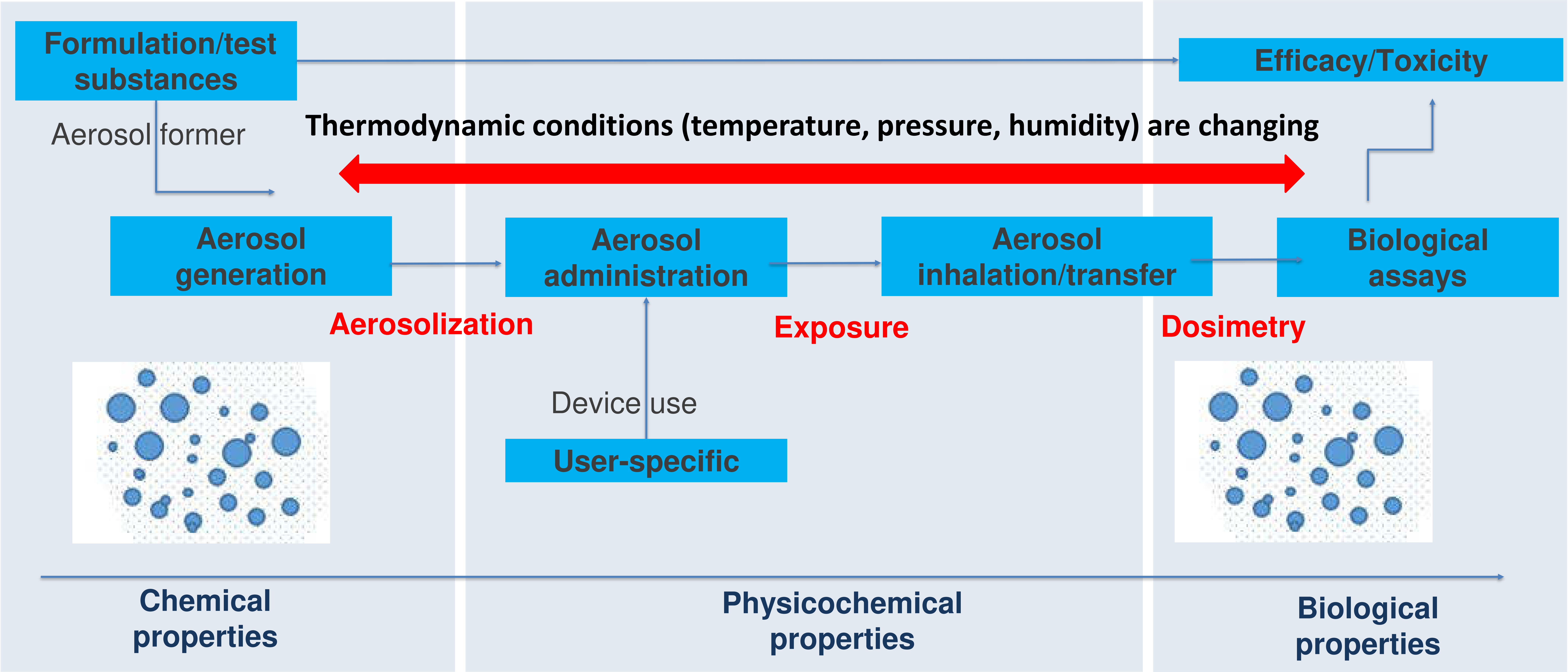
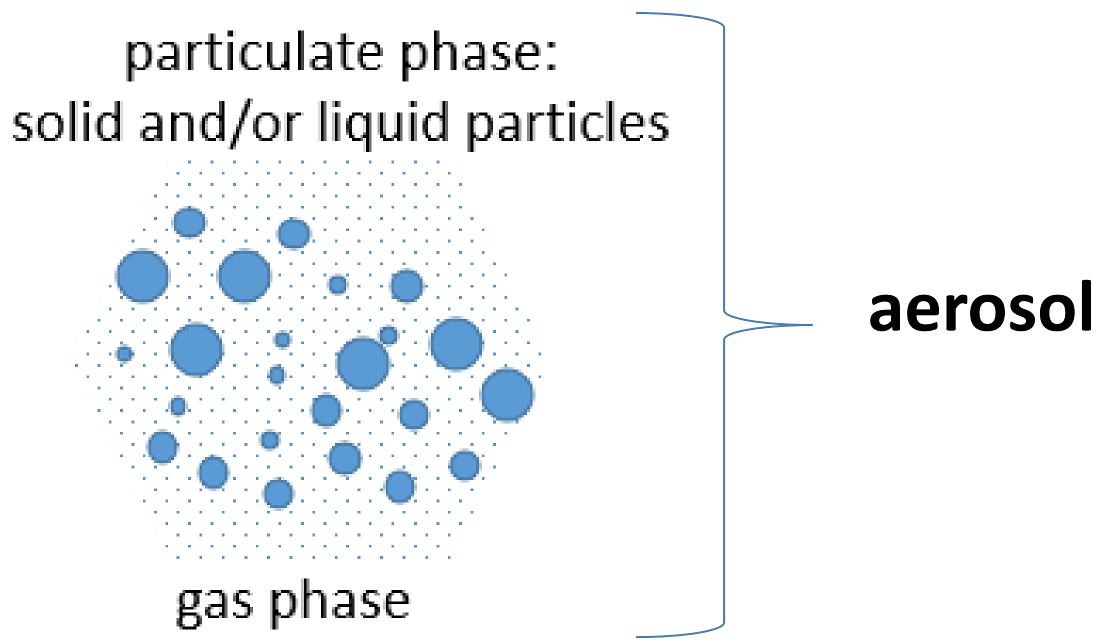
Aerosol delivery process



Importance of aerosol characterization at exposure site

Aerosol dosimetry



U.S. EPA, 1992

Exposure Assessment Tools by Routes - Inhalation

Potential dose is the amount of contaminant inhaled (i.e., amount that gets in the mouth or nose), not all of which is actually absorbed.

Applied dose is the amount of contaminant at the absorption barrier (e.g., respiratory tract) that can be absorbed by the body.

Internal dose is the amount of contaminant that gets past the exchange boundary (lung) and into the blood, or the amount of the contaminant that can interact with organs and tissues to cause biological effects.

Biologically effective dose is the amount of contaminant that interacts with the internal target tissue or organ.
Source: https://www.epa.gov/expobox/exposure-assessment-tools-routes-inhalation

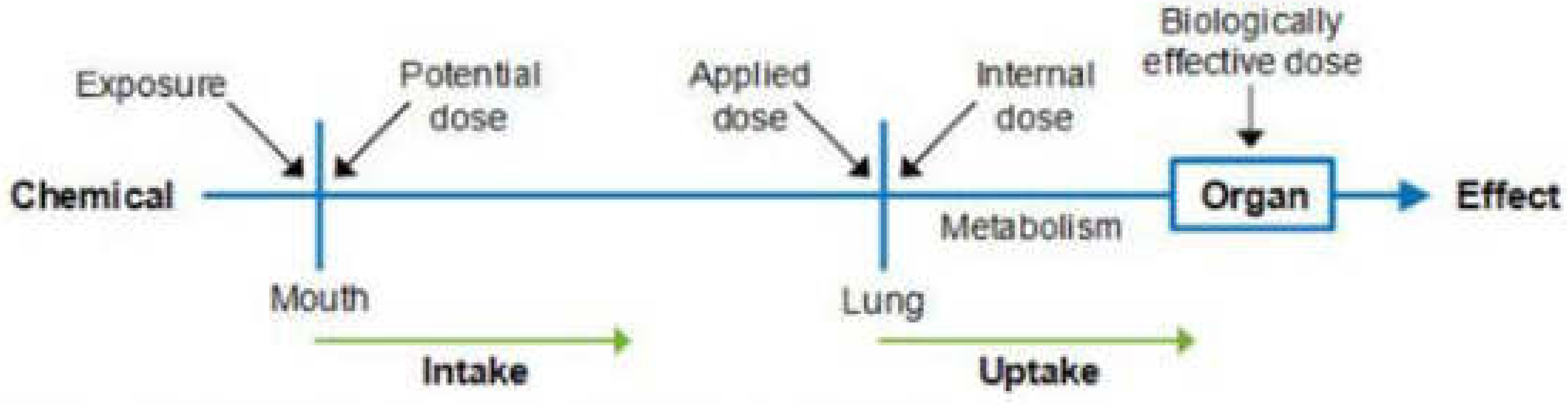


Illustration of Inhalation Route: Exposure and Dose (U.S. EPA, 1992)

Aerosols Handbook – Measurements, Dosimetry, and Health Effects

(Ed. L.S. Ruzer and N.H. Harley), 2013 (1st), 2019 (2nd) by CRC Press

O. Schmid, F.R. Cassee, On the pivotal role of dose for particle toxicology and risk assessment: exposure is a poor surrogate for delivered dose, Particle and Fibre Toxicology (2017) 14:52

R.A. Khanbeigi, A. Kumar, F. Sadouki, Ch. Lorenz, B. Forbes, L.A. Dailey, H. Collins, The delivered dose: Applying particokinetics to *in vitro* investigations of nanoparticle internalization by macrophages, Journal of Controlled Release (2012), 162(2), 259-266

Administered dose: particle mass, number or surface area administered per volume media at the onset of an experiment

Delivered dose: particle mass, number or surface area to reach the cell monolayer via diffusion and sedimentation over the duration of an experiment)

Cellular dose: particle mass, number or surface area internalized by the cells during the experiment

A Guide To Aerosol Delivery Devices for Respiratory Therapists

4th Edition American Association for Respiratory Care, 2017

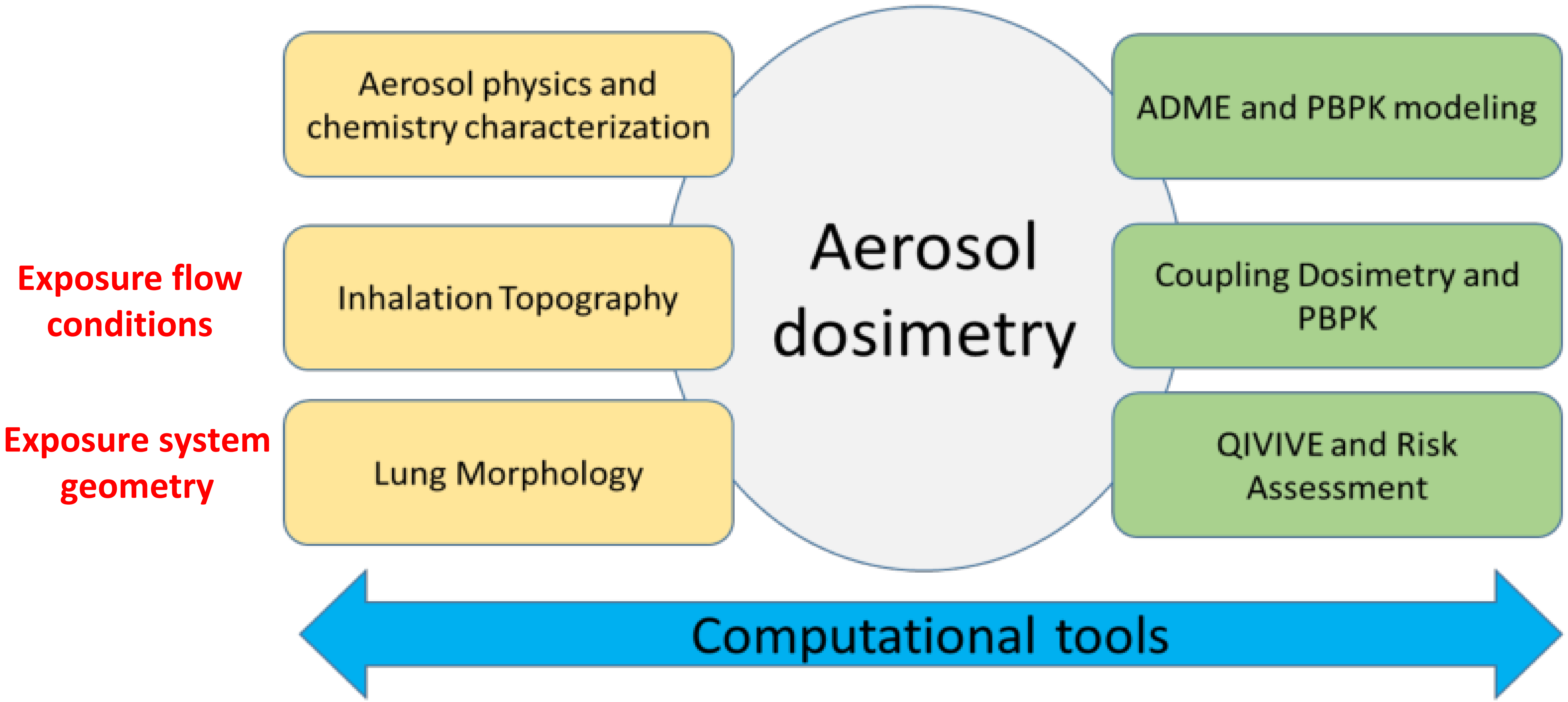
Nominal dose: the total drug dose placed in the nebulizer

Emitted dose: the mass of medication leaving an aerosol generated as aerosol

Inhaled dose: the proportion of nominal or emitted dose that is inhaled

Source: https://www.aarc.org/wp-content/uploads/2015/04/aerosol_guide_rt.pdf

Aerosol dosimetry challenges

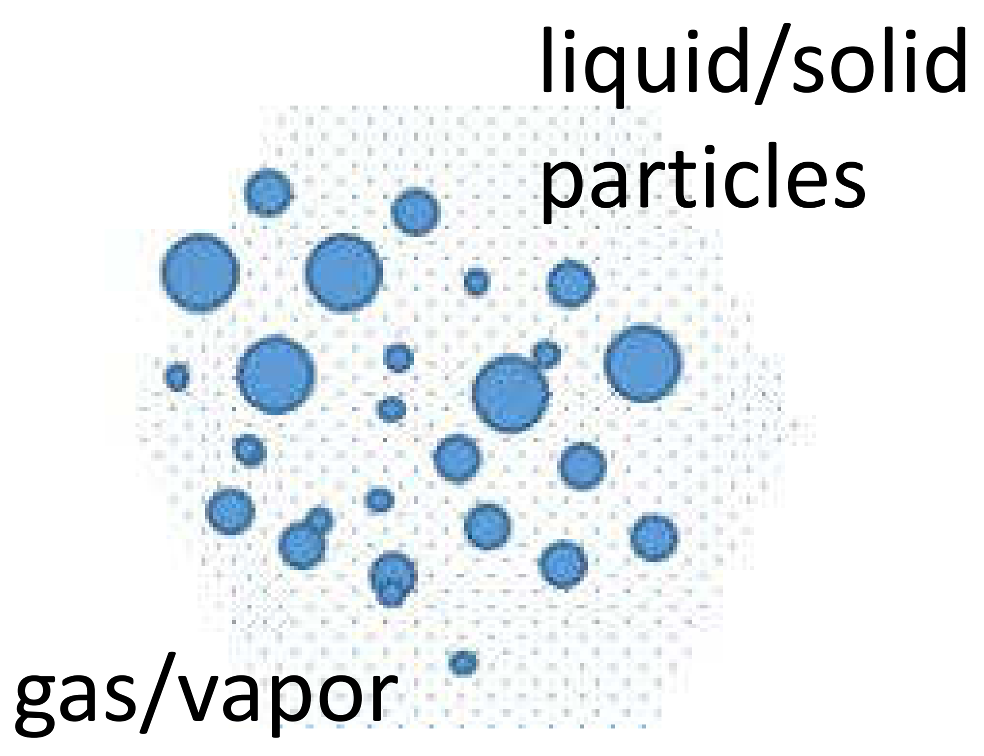


Alliance of Risk Assessment (ARA) workshop "Beyond Science and Decisions: From Problem Formulation to Dose-Response Assessment", 2019

ADME: absorption, distribution, metabolism, and excretion
PBPK: physiologically based pharmacokinetic modelling
QIVIVE: quantitative in vitro-to-in vivo extrapolation

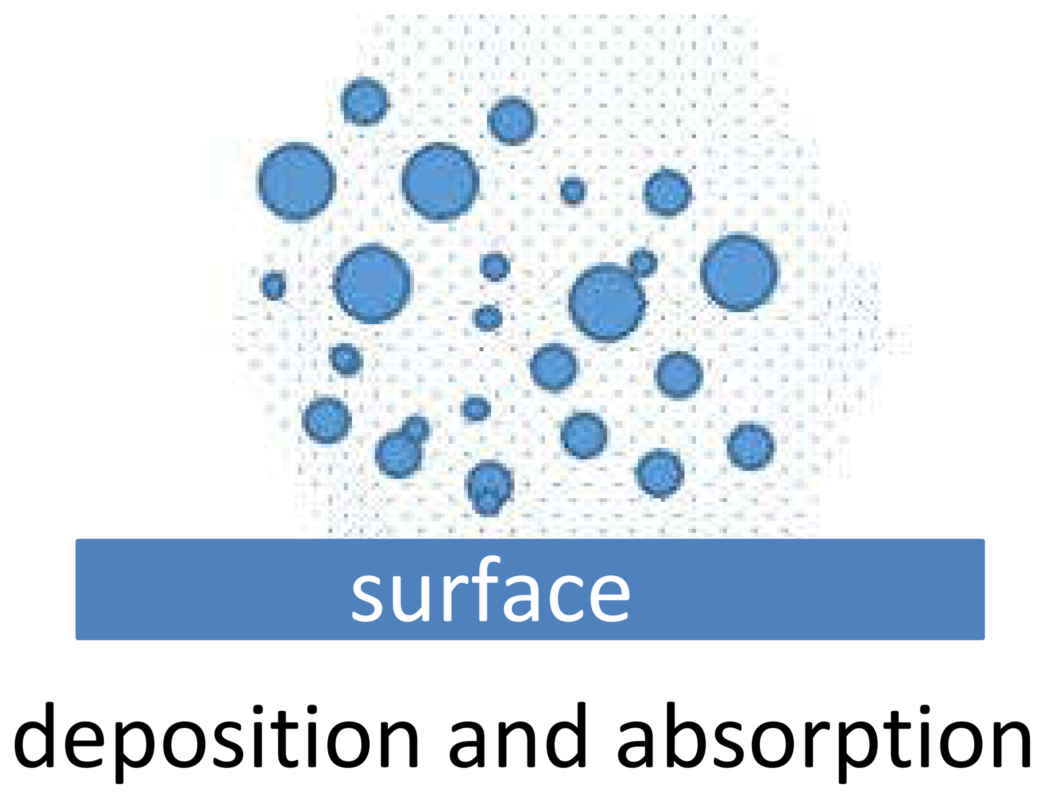
Kolli AR, Kuczaj AK, Martin F, Hayes AW, Peitsch MC, Hoeng J. Bridging inhaled aerosol dosimetry to physiologically based pharmacokinetic modeling for toxicological assessment: nicotine delivery systems and beyond. Crit Rev Toxicol. 2019 Oct;49(9):725-741. doi: 10.1080/10408444.2019.1692780. Epub 2020 Jan 6. PMID: 31903848.

Aerosol characterization



Individual particle properties
- Chemical composition
- Size, morphology, shape
- Optical, electrical, magnetic properties
- Aggregate properties

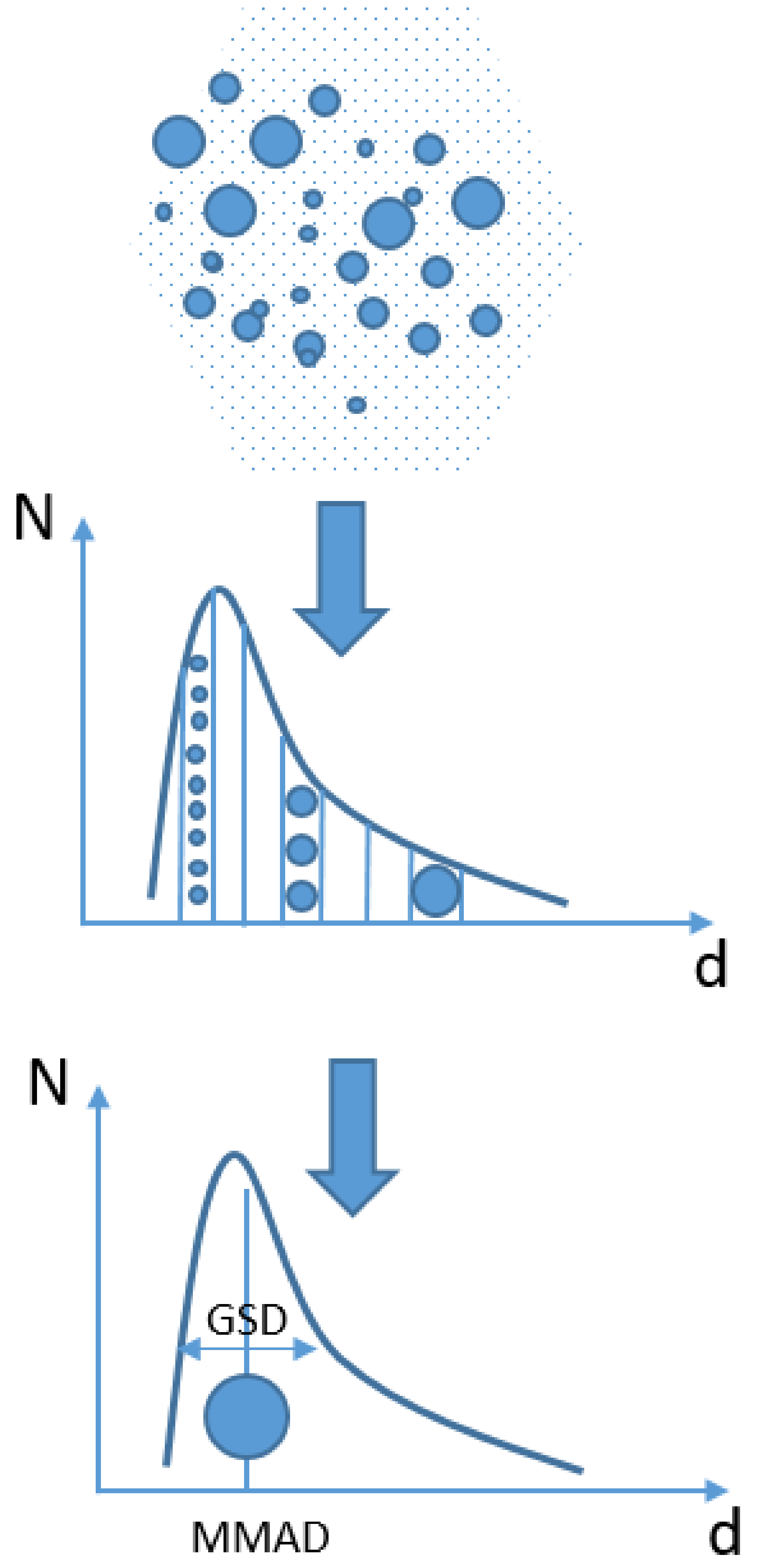
Aerosol properties
- Phase partitioning: gas, liquid, solid
- Particle size distribution (PSD): number, mass, surface area, ….
- Light absorption and scattering, charge distribution
- Hygroscopicity, volatility, dissolution



Particle size distribution (polydisperse / rarely monodisperse)

Parametrization: assumption of log-normal distribution

Measures:
- Particle number density (#/m^3)
- Average size (mass median aerodynamic diameter [MMAD])
- Distribution width (geometric standard deviation [GSD])



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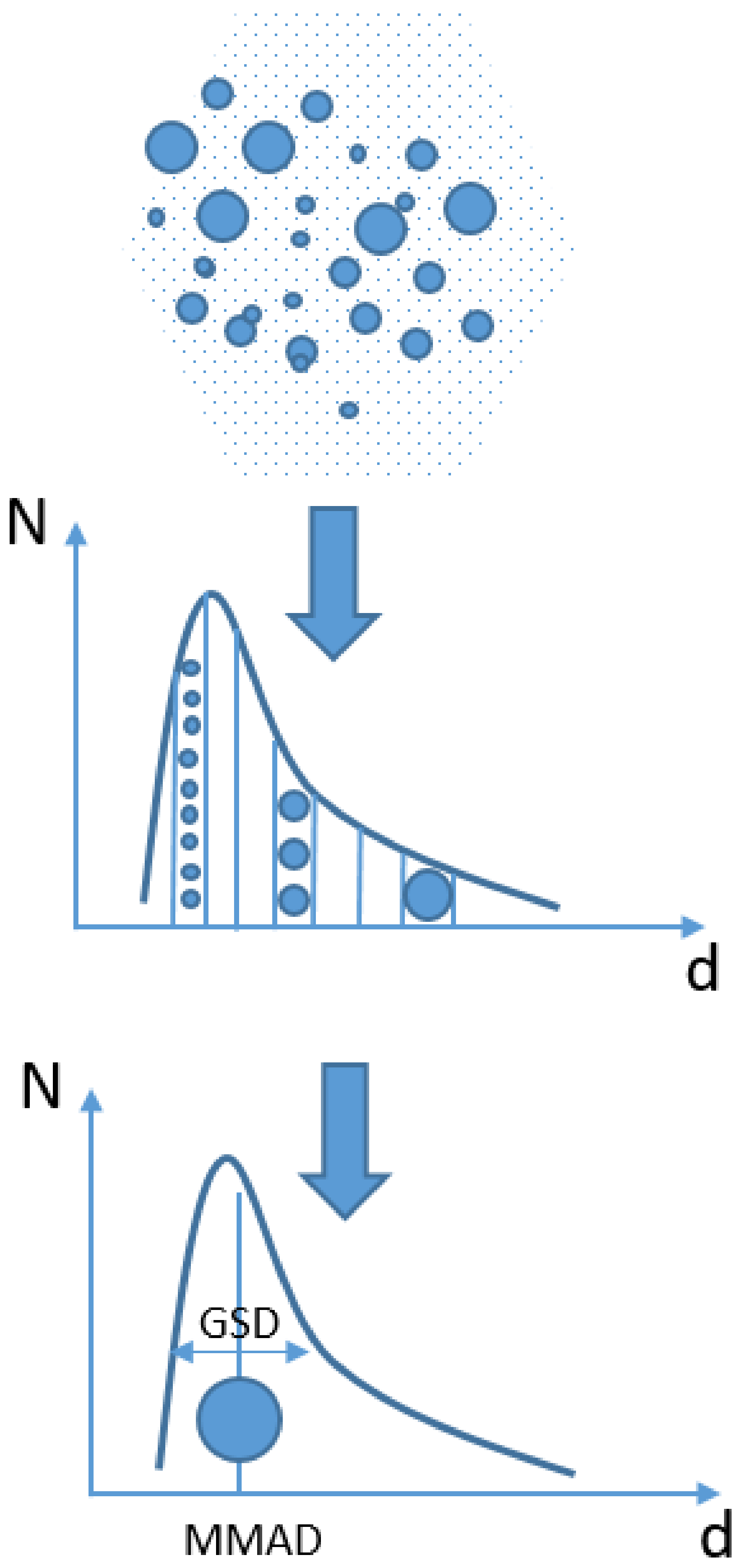
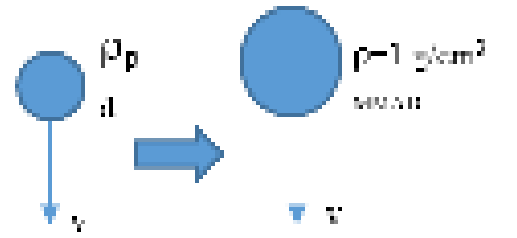
Physical characterization





Instruments based on various measuring principles:

- Inertia and aerodynamic drag
- Light scattering
- Electrical charging

Challenges:

- Invasive techniques
- Not applicable for large particle number densities
- Often needs dilution, thus leading to aerosol evolution

Andersen Cascade Impactor

source: www.copleyscientific.com

Next Generation Impactor

source: www.tsi.com

Spraytec

source: www.malvernpanalytical.com

Scanning Mobility Particle Sizer

source: www.tsi.com

Aerodynamic Particle Sizer

source: www.tsi.com

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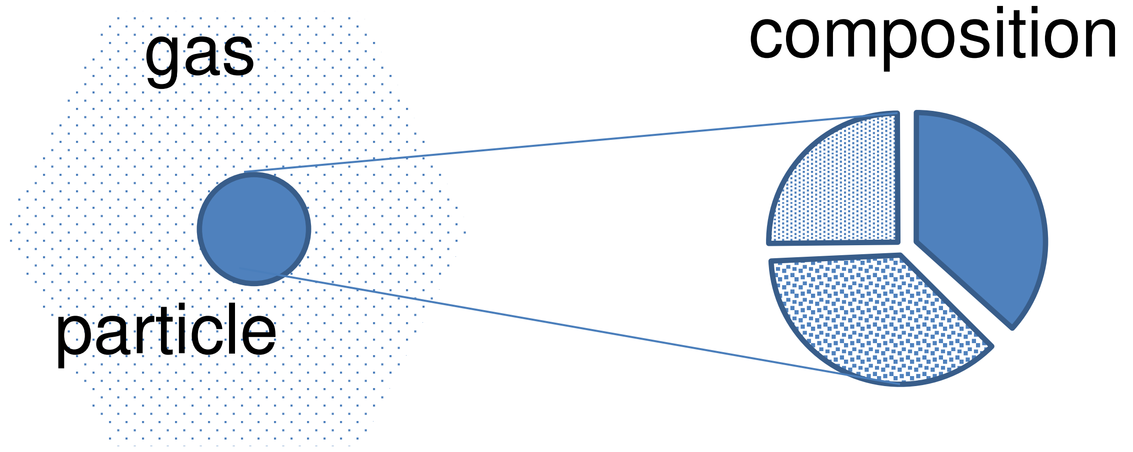
Chemical characterization



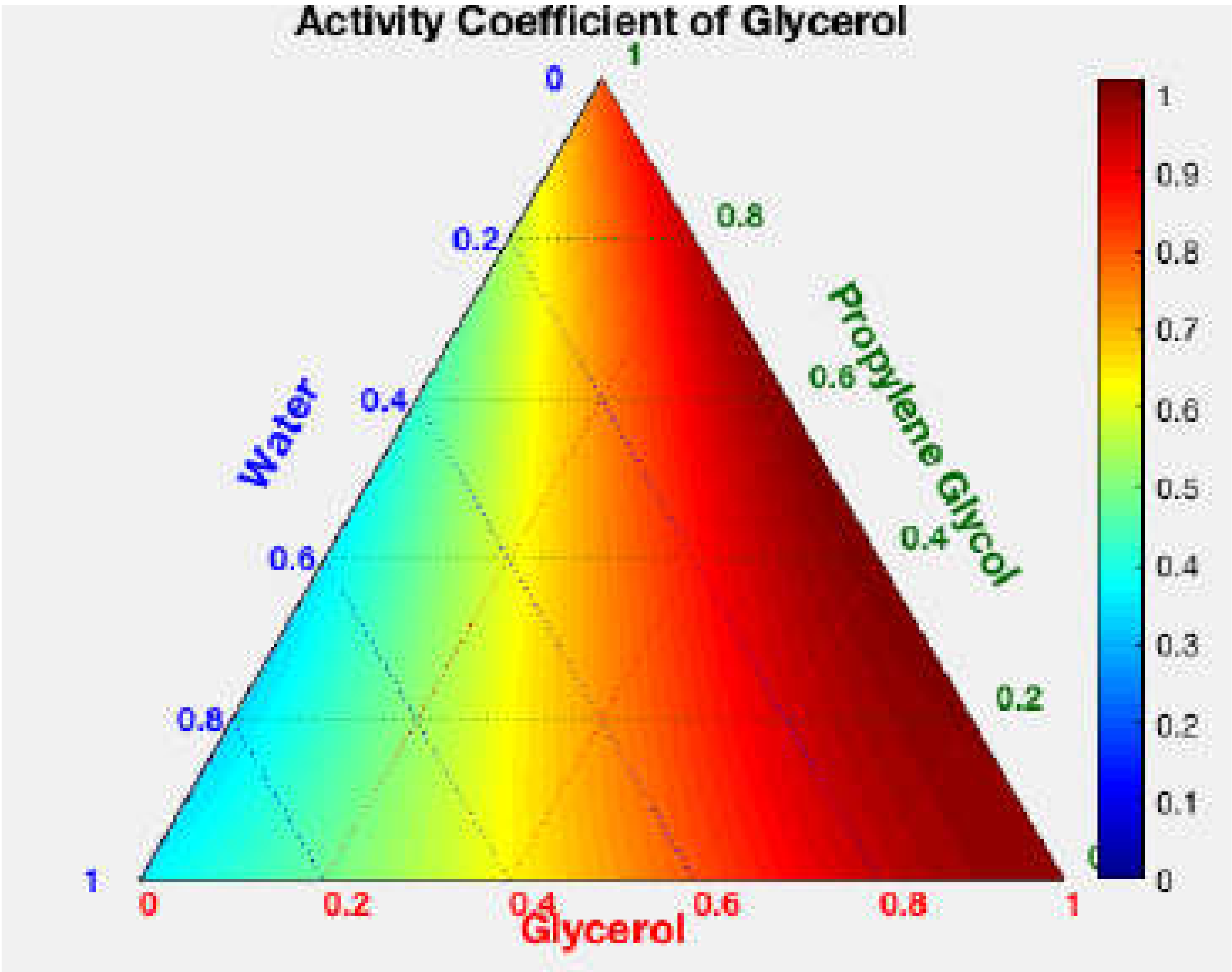
Chemical properties

- Individual substance properties
- Multispecies mixture properties
- Mixture stability

Molecular size, density, viscosity, surface tension, thermal conductivity, specific heat, boiling point, melting point, vapor pressure curve, hydrophobicity, solubility, miscibility, pH: temperature dependence

Raoult's law:
predicts gas composition (total pressure) from the partial vapor pressures of substances over an ideal mixture.

Mixture properties are often not a superposition of individual substances' properties



Vapor-liquid equilibrium (VLE) modeling

Challenges:

- **Phase partitioning:** Often composition in the gas phase is different from the one predicted from the liquid phase – mixtures have non-ideal behavior.

- Departure from the Raoult's law for non-ideal mixtures is captured by the **activity coefficient,** which is difficult to measure.

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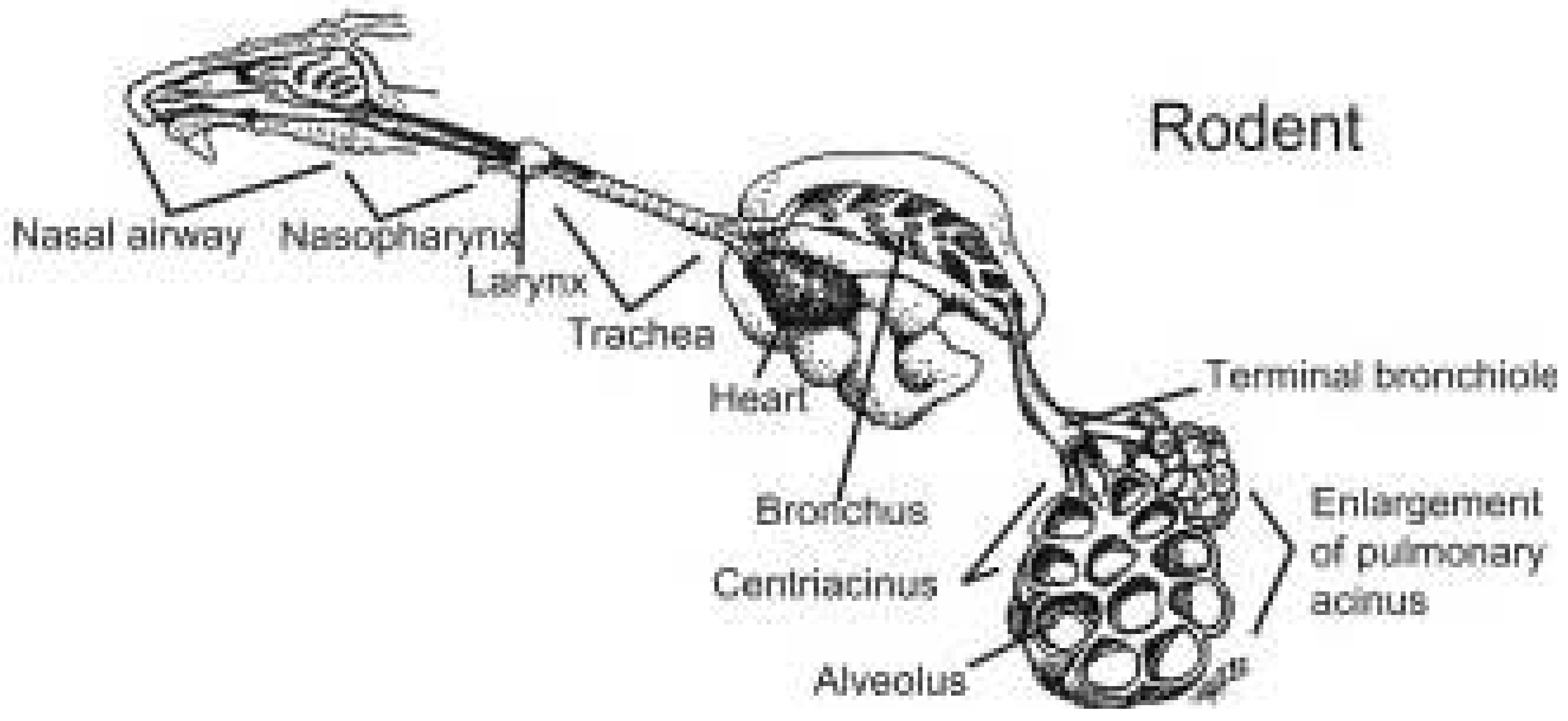
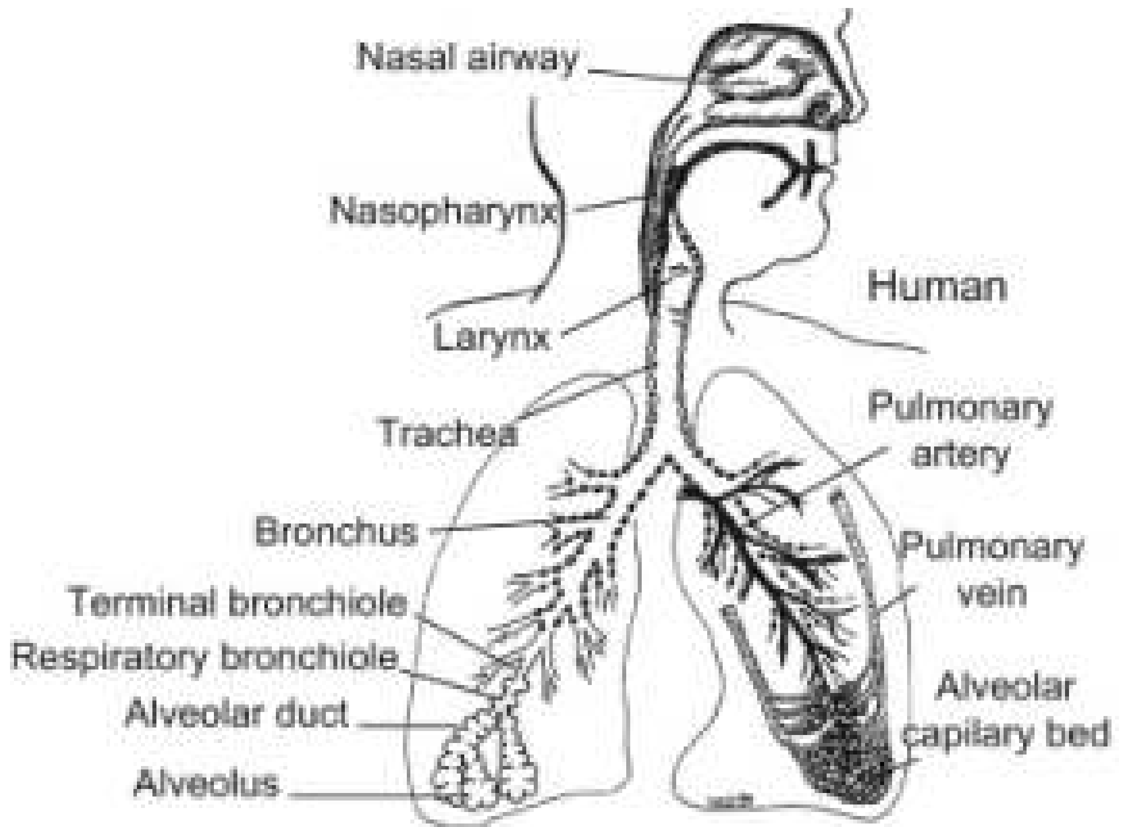
Lung morphology

Airway dimensions

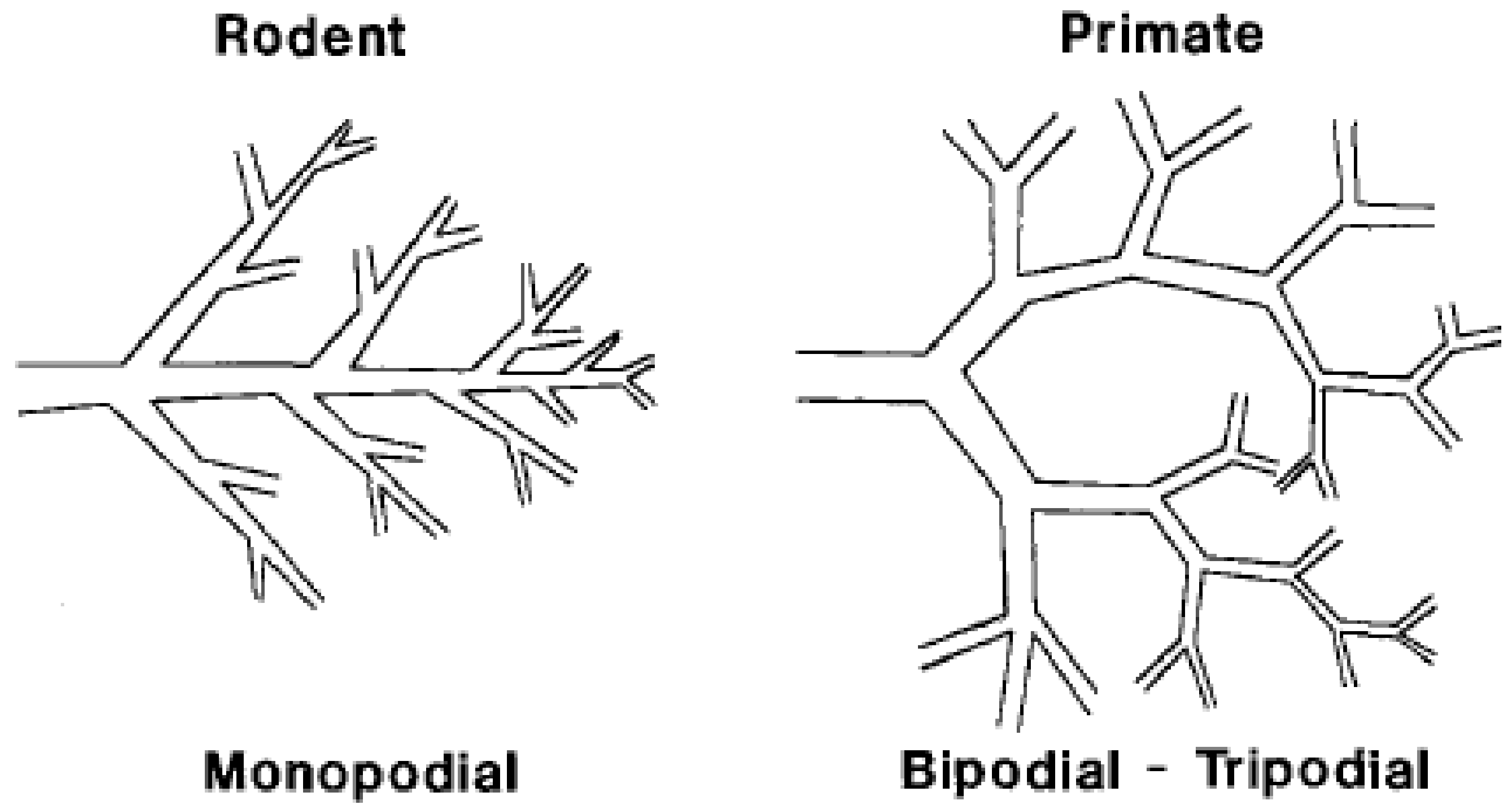


(Harkema 2013)

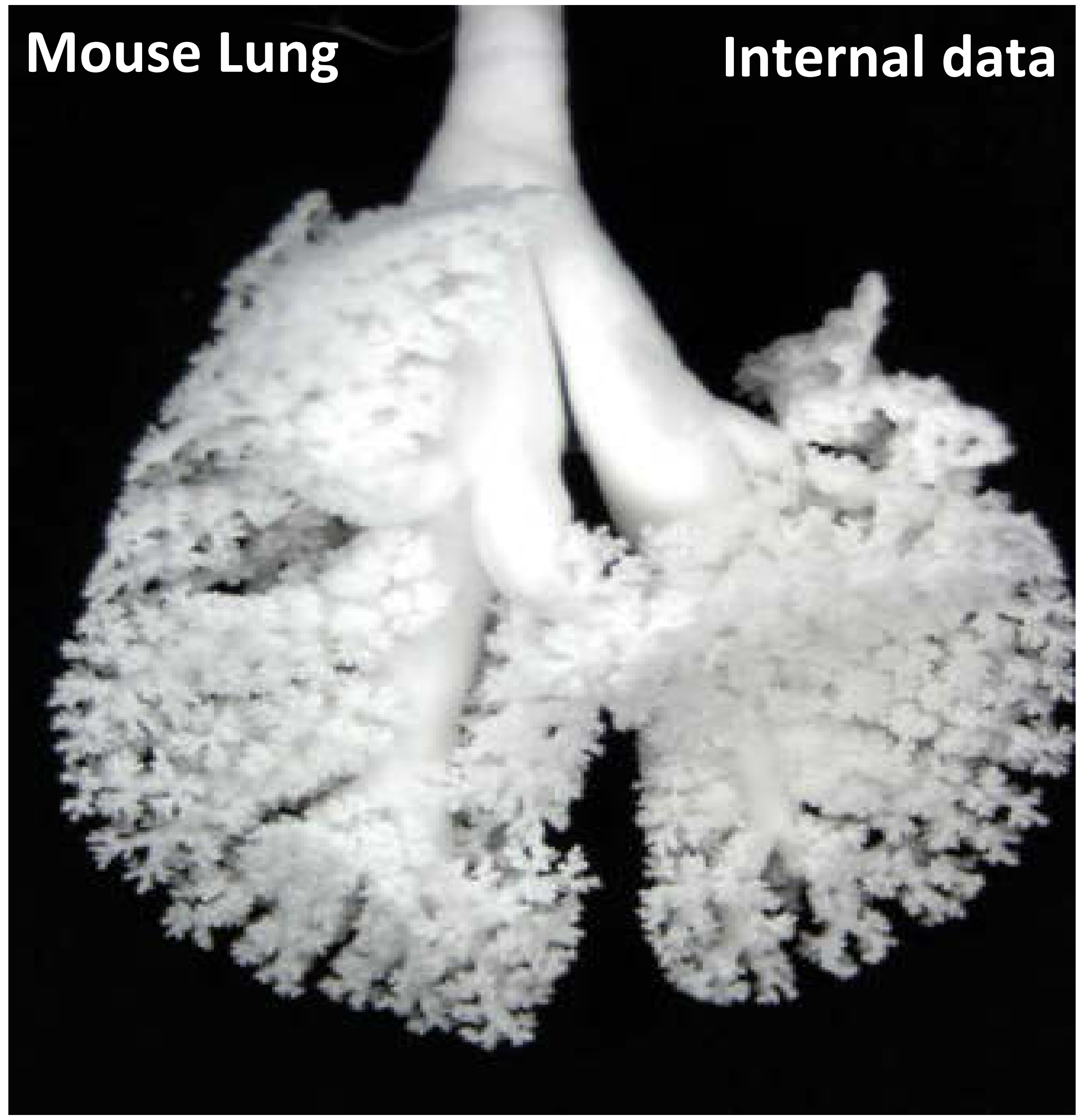


(Harkema 2013)

Branching pattern



(J. F. Miller, J.D. Crapo, 1993)



SOT Presentation, 2019

- Limited data on deposition
- Humans: Lovelace Morphometry Report (1976)
  - Measurements for lungs
- Rodents: limited information concerning strain differences
  - Cast-based measurements (Phalen, 1973)
- Individual-specific versus population-relevant

Harkema JR, Nikula KJ, Haschek WM. 2013. Chapter 51 - Respiratory System, p 1935-2003. *In* Haschek WM, Rousseaux CG, Wallig MA (ed), Haschek and Rousseaux's Handbook of Toxicologic Pathology (Third Edition) https://doi.org/10.1016/B978-0-12-415759-0.00051-0. Academic Press, Boston.

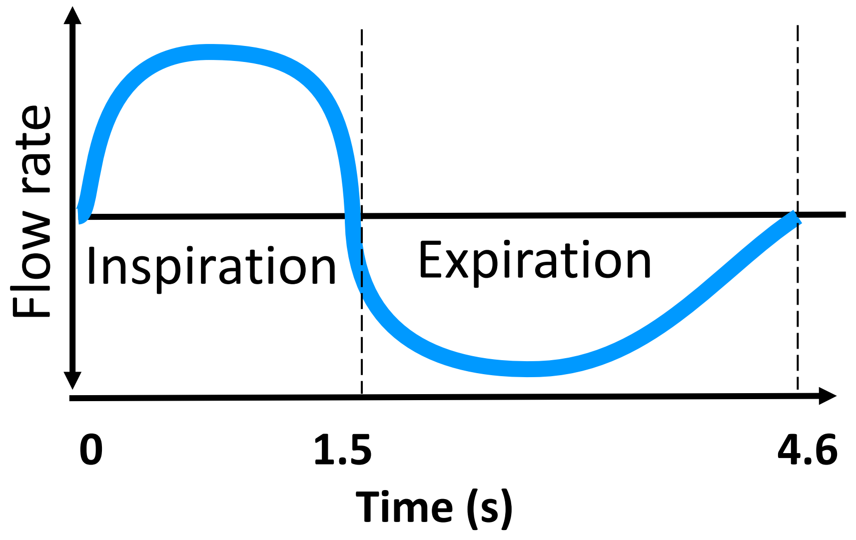
Miller FJ, Mercer RR, Crapo JD. 1993. Lower Respiratory Tract Structure of Laboratory Animals and Humans: Dosimetry Implications. Aerosol Science and Technology 18:257-271.

Phalen RF, Yeh HC, Raabe OG, Velasquez DJ. Casting the lungs In-situ. Anat Rec. 1973 Oct;177(2):255-63. doi: 10.1002/ar.1091770207. PMID: 4756760.

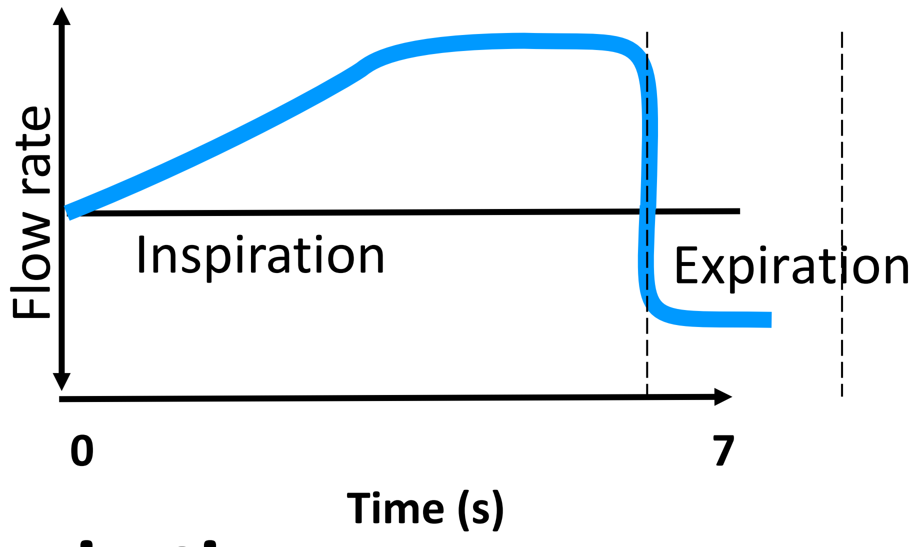
Inhalation topography

Tidal breathing patterns

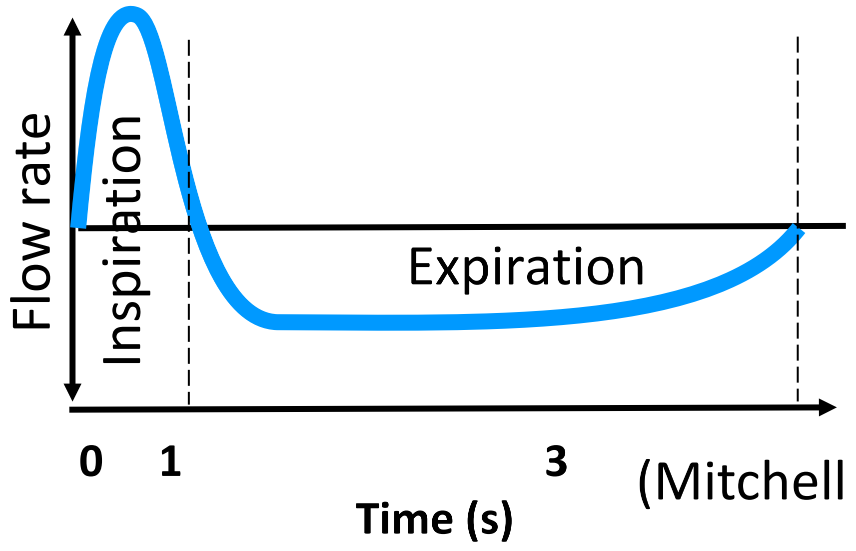
Standard



Slow inspiration and breath hold



Forced inspiration

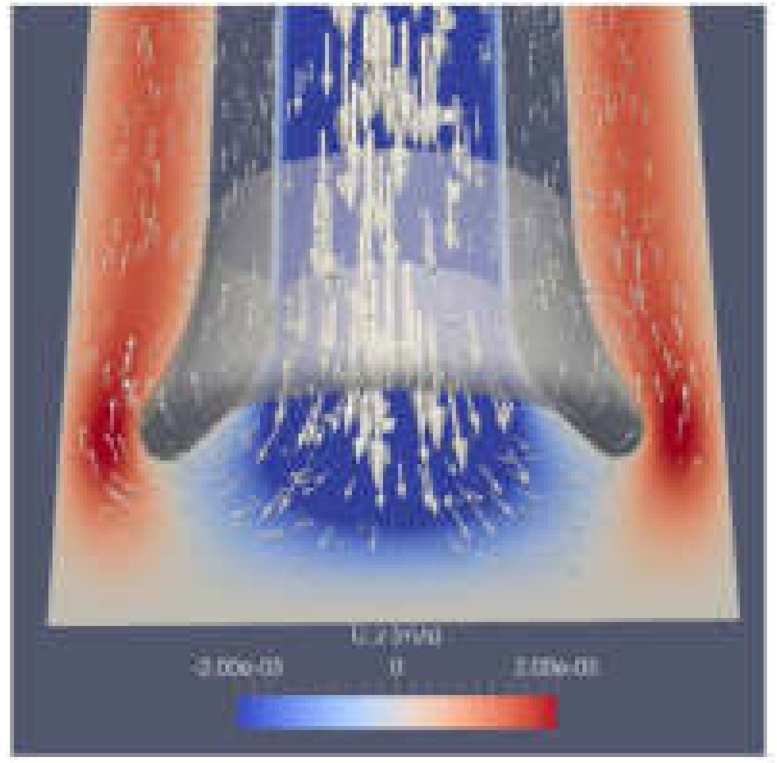


(Mitchell 2016)

- Single individual
- Measured flow rate during inhalation cycle
- Each step can vary across population

Inhalation topography and lung morphology alter deposited dose

In vitro:



Exposure system geometry and flow conditions alter deposited dose

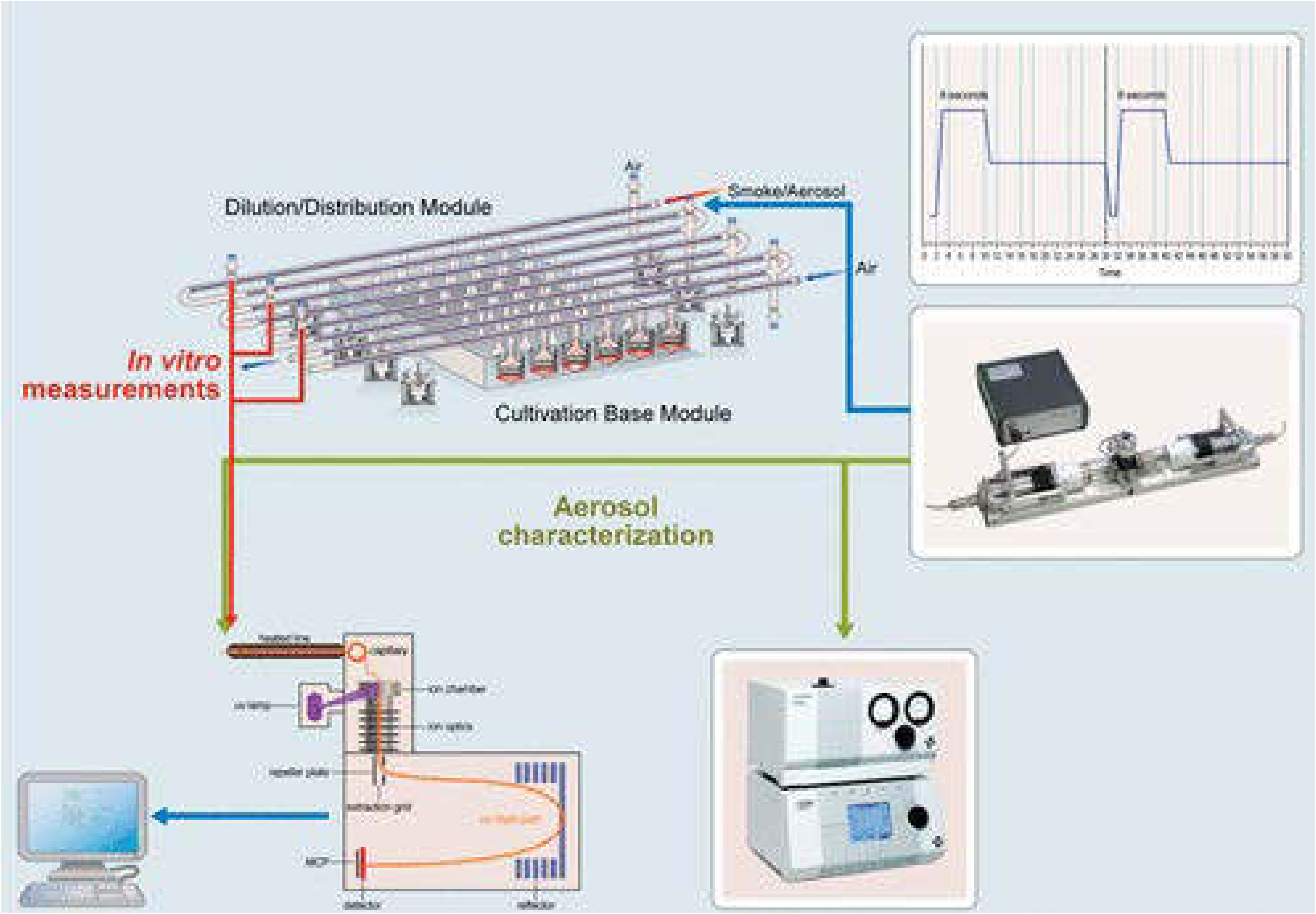
Lucci F, Castro ND, Rostami AA, Oldham MJ, Hoeng J, Pithawalla YB, Kuczaj AKK, Characterization and modeling of aerosol deposition in Vitrocell® exposure systems - exposure well chamber deposition efficiency, Journal of Aerosol Science, 123, 2018, 141-160, https://doi.org/10.1016/j.jaerosci.2018.06.015.

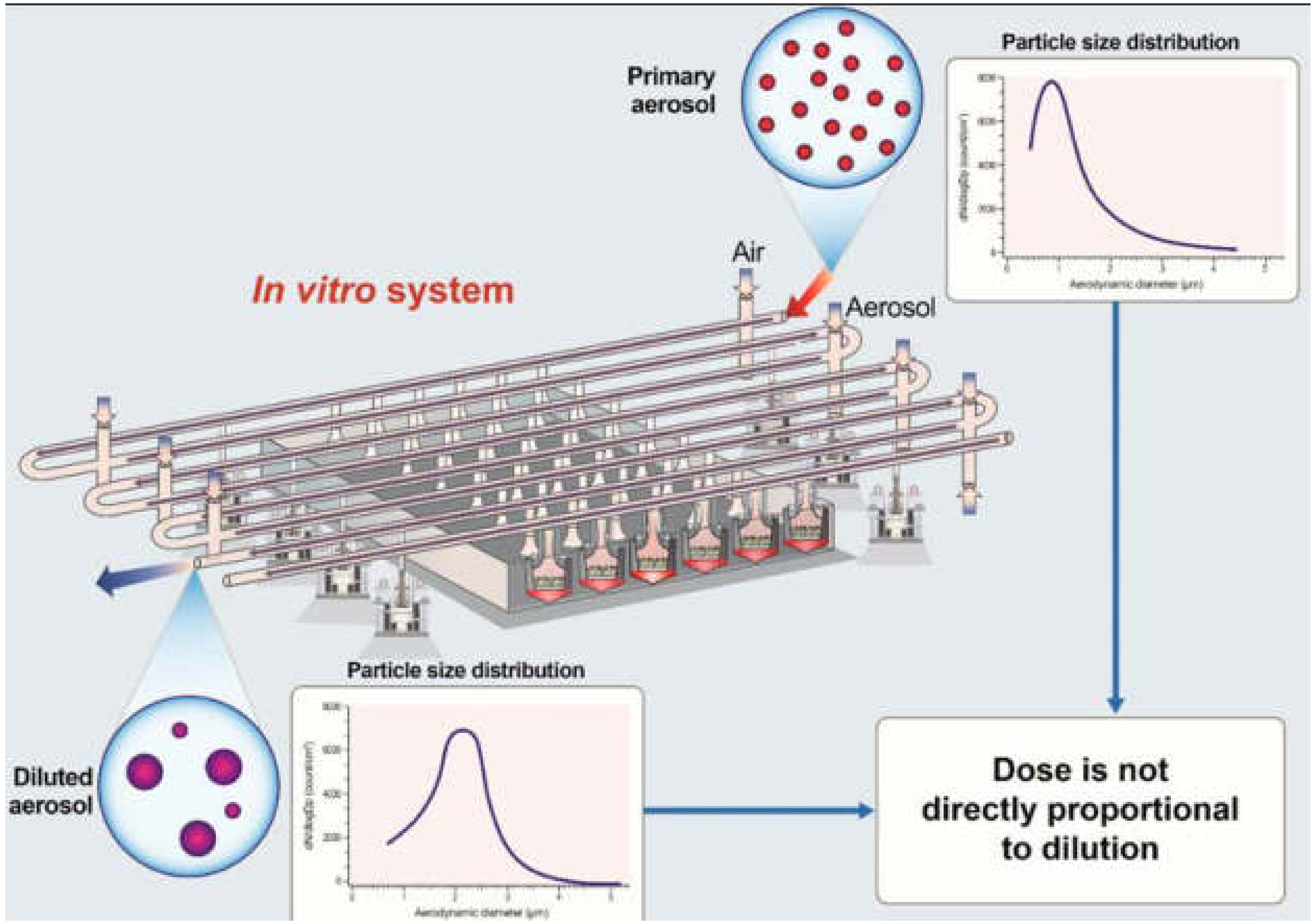
Mitchell JP, Suggett J, Nagel M. 2016. Clinically Relevant In Vitro Testing of Orally Inhaled Products—Bridging the Gap Between the Lab and the Patient. AAPS PharmSciTech 17:787-804.

Vas CA, Yurteri CÜ , Dickens CJ, Prasad K, Development and Characterisation of a Smoking Behaviour Measurement System, Beiträge zur Tabakforschung International 26 (5), 2015, DOI: 10.1515/cttr-2015-0010

Influence of exposure flow conditions and system geometry

Assessment of single-photon ionization mass spectrometry (SPI-MS) technique to measure on-line the main substances of a test aerosol during *in vitro* exposure





source: Frege C, Asgari M, Steiner S, Ferreira S, Majeed S, Lucci F, Frentzel S, Hoeng J, Kuczaj AK. Assessment of Single-Photon Ionization Mass Spectrometry for Online Monitoring of *in Vitro* Aerosol Exposure Experiments. Chem Res Toxicol. 2020 Feb 17;33(2):505-514. doi: 10.1021/acs.chemrestox.9b00381. Epub 2020 Jan 7. PMID: 31909610.

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Inhaled dose calculation

Association of Inhalation Toxicologists recommend standard delivered dose calculation in aerosol inhalation studies

$$DD = \frac{C \times RMV \times D(\times IF)}{BW}$$

(Alexander 2008)

DD = delivered dose
C = concentration of substance in air
RMV = respiratory minute volume
D = duration of exposure
IF = inhalable fraction
BW = body weight

In vitro:

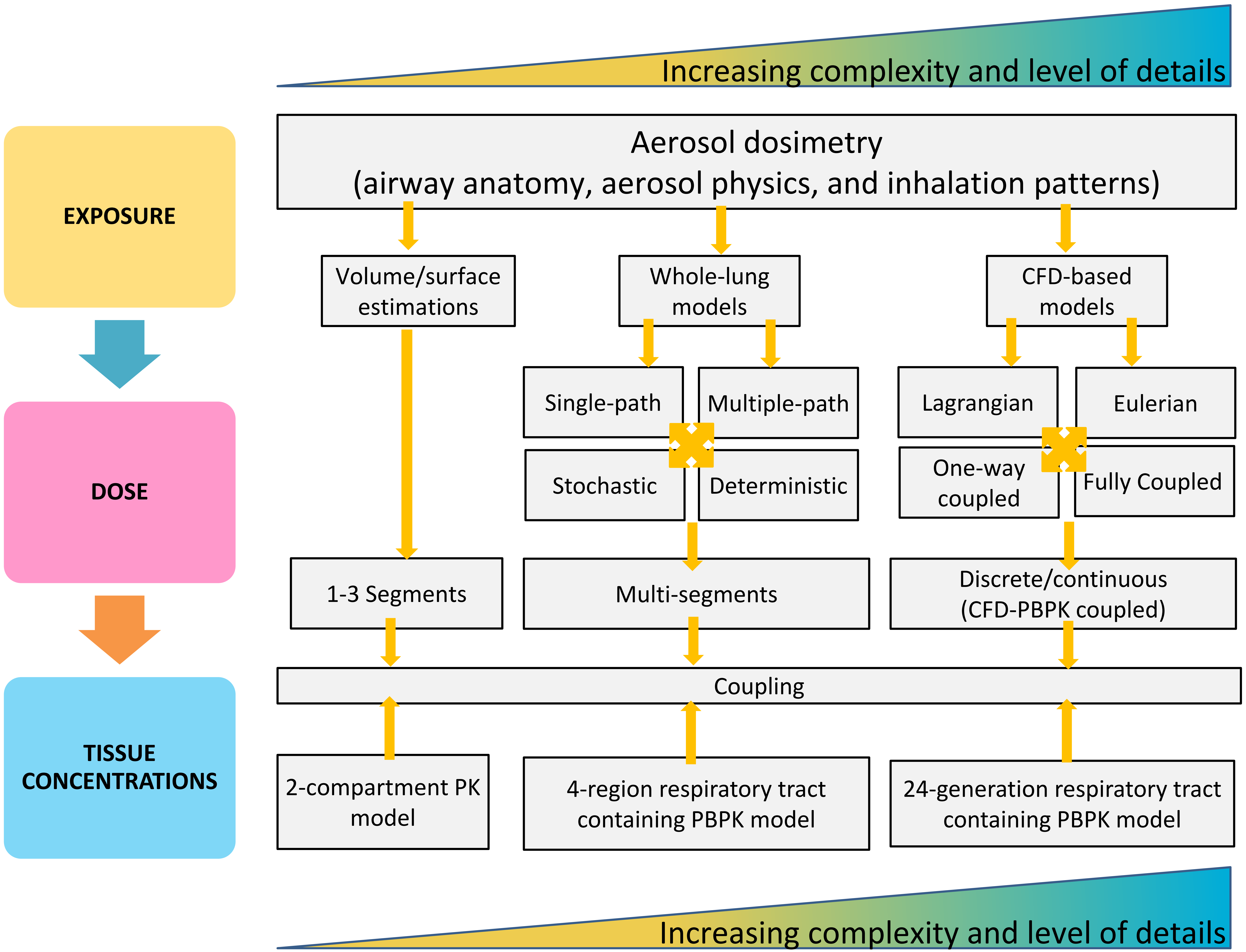
Similar estimations based on the substance concentration, available surface area and exposure duration are performed.

More holistic determination of dose calculation must account for at least the following:

- Aerosol physics (aerosol evolution)
- Inhalation topography (flow conditions)
- Lung morphology (system geometry)

Alexander DJ, Collins CJ, Coombs DW, Gilkison IS, Hardy CJ, Healey G, Karantabias G, Johnson N, Karlsson A, Kilgour JD, McDonald P. Association of Inhalation Toxicologists (AIT) working party recommendation for standard delivered dose calculation and expression in non-clinical aerosol inhalation toxicology studies with pharmaceuticals. Inhal Toxicol. 2008 Oct;20(13):1179-89. doi: 10.1080/08958370802207318. PMID: 18802802.

Modeling of inhaled aerosol – dosimetry/PBPK view



A.R. Kolli et al., Bridging inhaled aerosol dosimetry to physiologically based pharmacokinetic modeling for toxicological assessment: nicotine delivery systems and beyond, Critical Reviews in Toxicology, 49 (9), 2019

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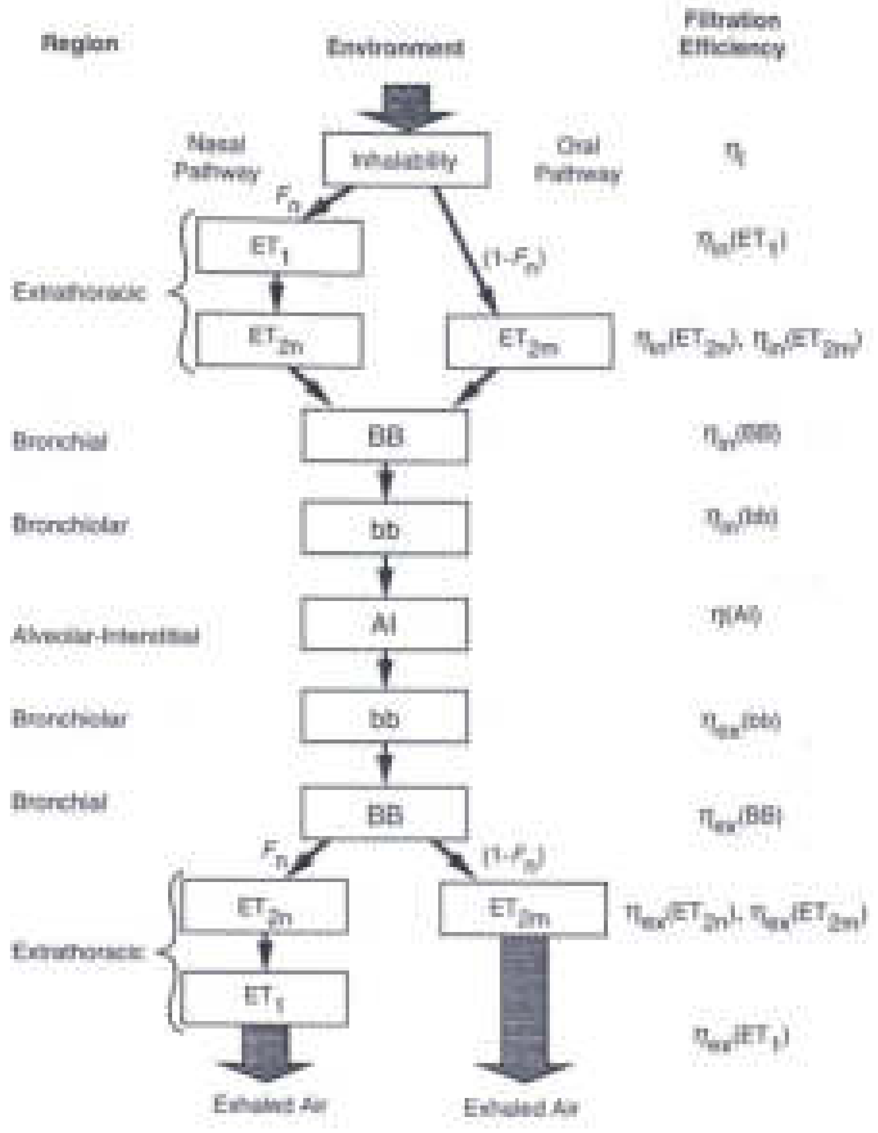
1D (whole lung) models development

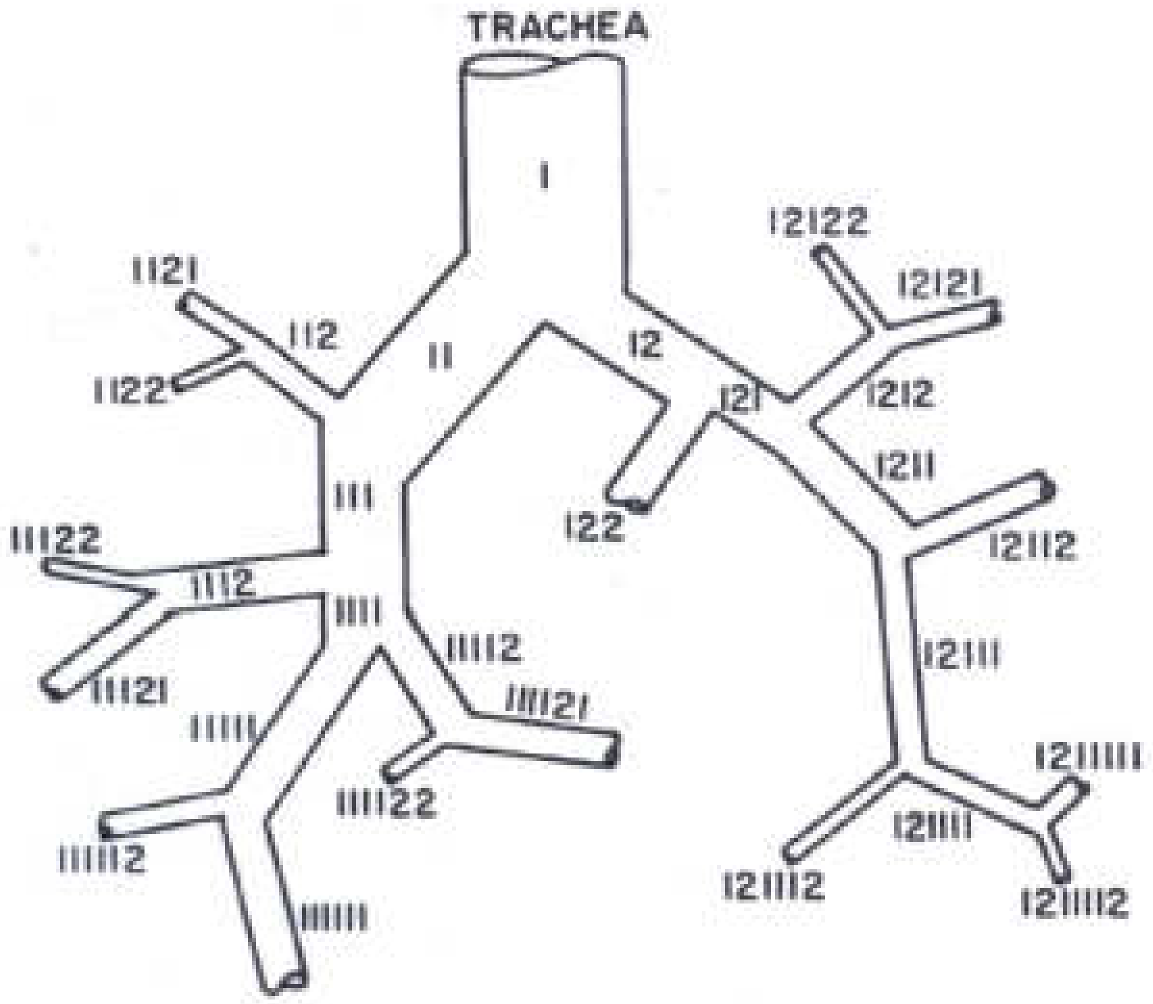
ICRP (1994): Human respiratory tract model for radiological protection

- Regional deposition efficiencies for inhalation and exhalation
- Described by semi-empirical equations as functions of particle size and flow rate
- Derived from mathematical fits through the available experimental data

ICRP. Human respiratory tract model for radiological protection. ICRP Publication 66. 1994. Ann. ICRP 24 (1-3).



Raabe et al. (1976):
Morphometric measurements of the tracheobronchial tree:
from the trachea to the terminal bronchioles



- Stahlhofen et al, J. Aerosol Sci., 1983
- Yeh, Bull. Math. Biol., 1980
- Anjilvel & Asgharian, Fundam. Appl. Toxicol., 1995
- Hofmann, Journal of Aerosol Science, 2011

MPPD
MULTIPLE-PATH PARTICLE DOSIMETRY MODEL

Multiple-Path Particle Dosimetry Model
Applied Research Associates, Inc. (ARA)

- Based on the Raabe et al. (1976) measurements for the first 10 airway generations (asymmetric branching)
- Supplemented by 50 structurally different multi-path models of the bronchial tree derived from the stochastic lung model
- Typical path (i.e., symmetric alveolar geometry) based on the Yeh and Schum (1980) model
- Asymmetric flow splitting at airway bifurcations proportional to distal volume
- Solution of the mass balance equation with different loss terms for the various deposition mechanisms

- **Limited airway geometries**
- **Limited available correlation data**
- **Developed for solid (non-evolving) particles**
- **Constrained to application in selected geometries (not directly useful for the in vitro)**

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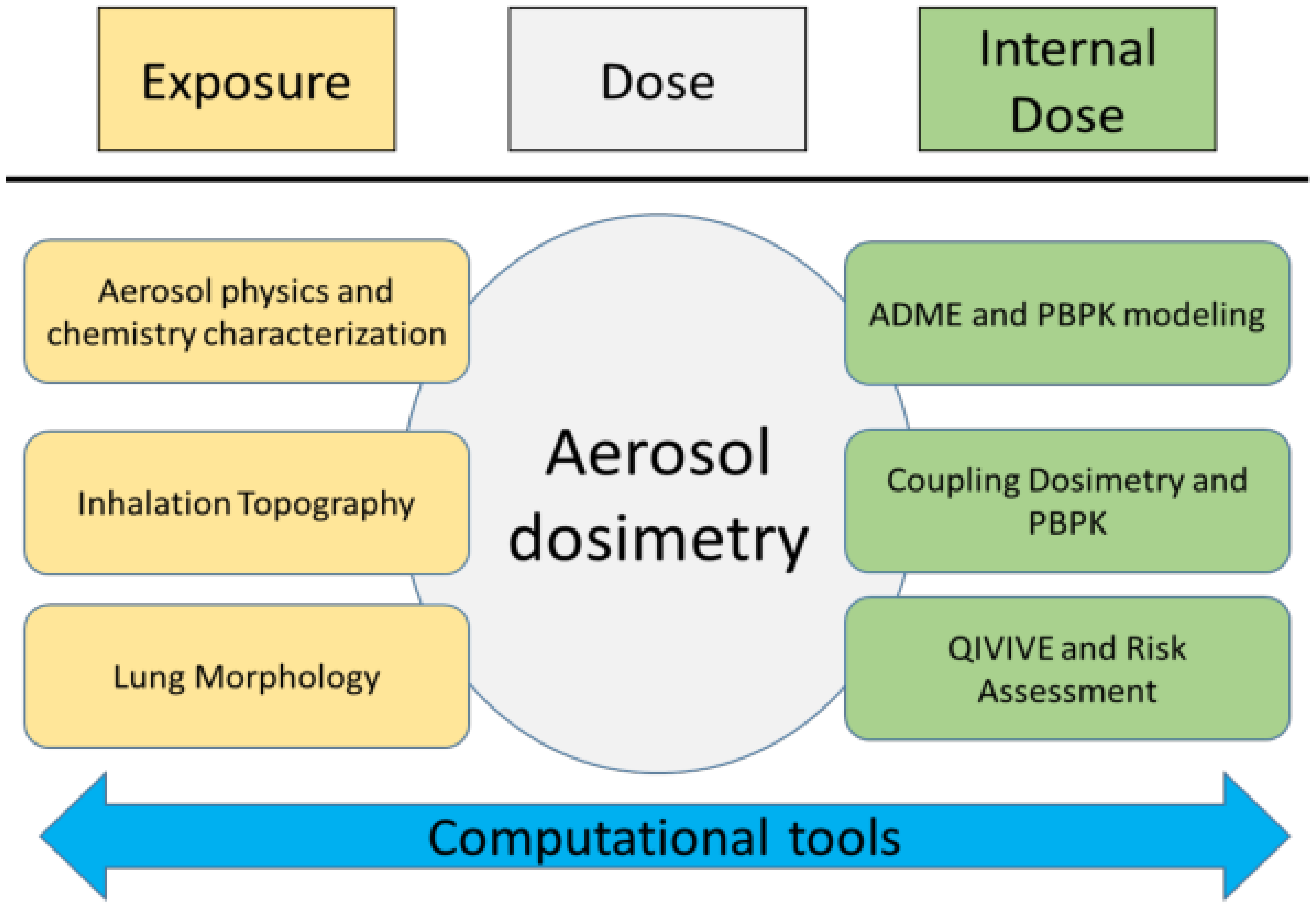
Dosimetry requires multidisciplinary and synergistic efforts



- **Dosimetry predictions: mouse lung morphometry data**
- **Development of in vitro exposure systems that are based on physiologically driven processes (InHALES)**
- **Development of cast models for understanding aerosol evolution in the respiratory tract**
- **Development of computational tools for prediction of aerosol evolution and delivery (AeroSolved)**

Rodent dosimetry predictions

Aerosol generation

Exposure characterization

SysTox: biological responses

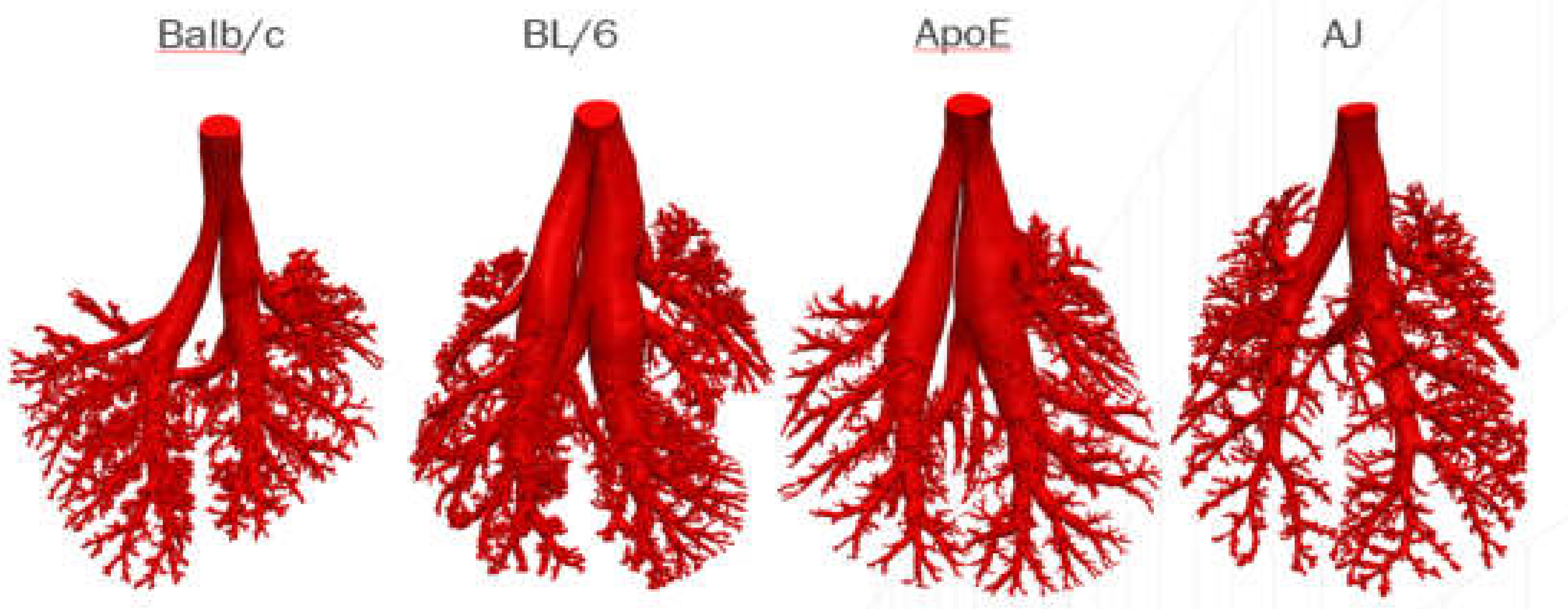
Animal exposure:
- Animal lung morphometry
- Animal respiratory physiology

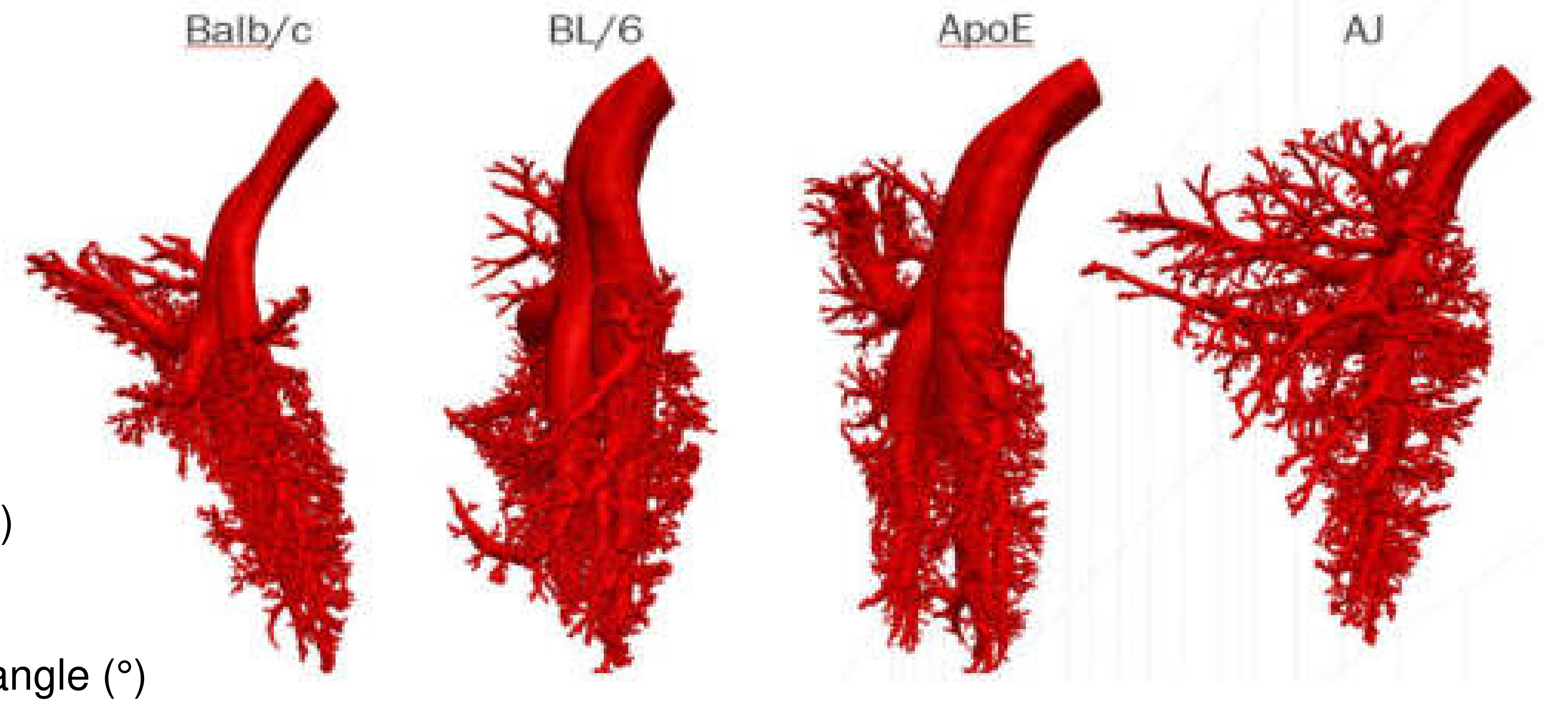
Aerosol:
- Aerosol characterization
- Exposure system characterization

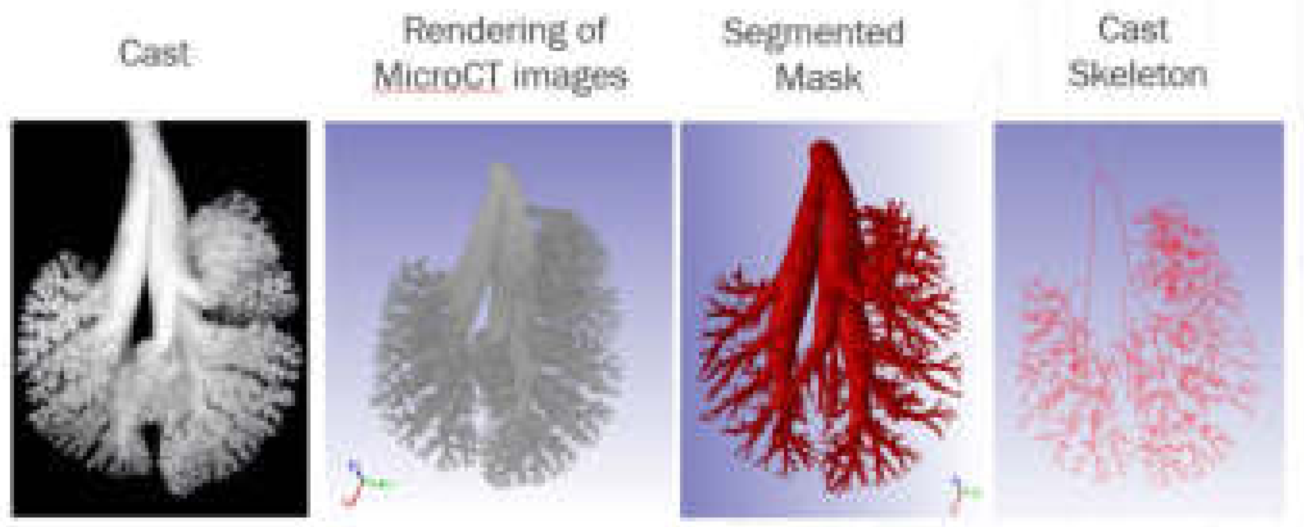
Dosimetry model (whole-lung)

Dosimetry characterization

Aerosol dose estimation



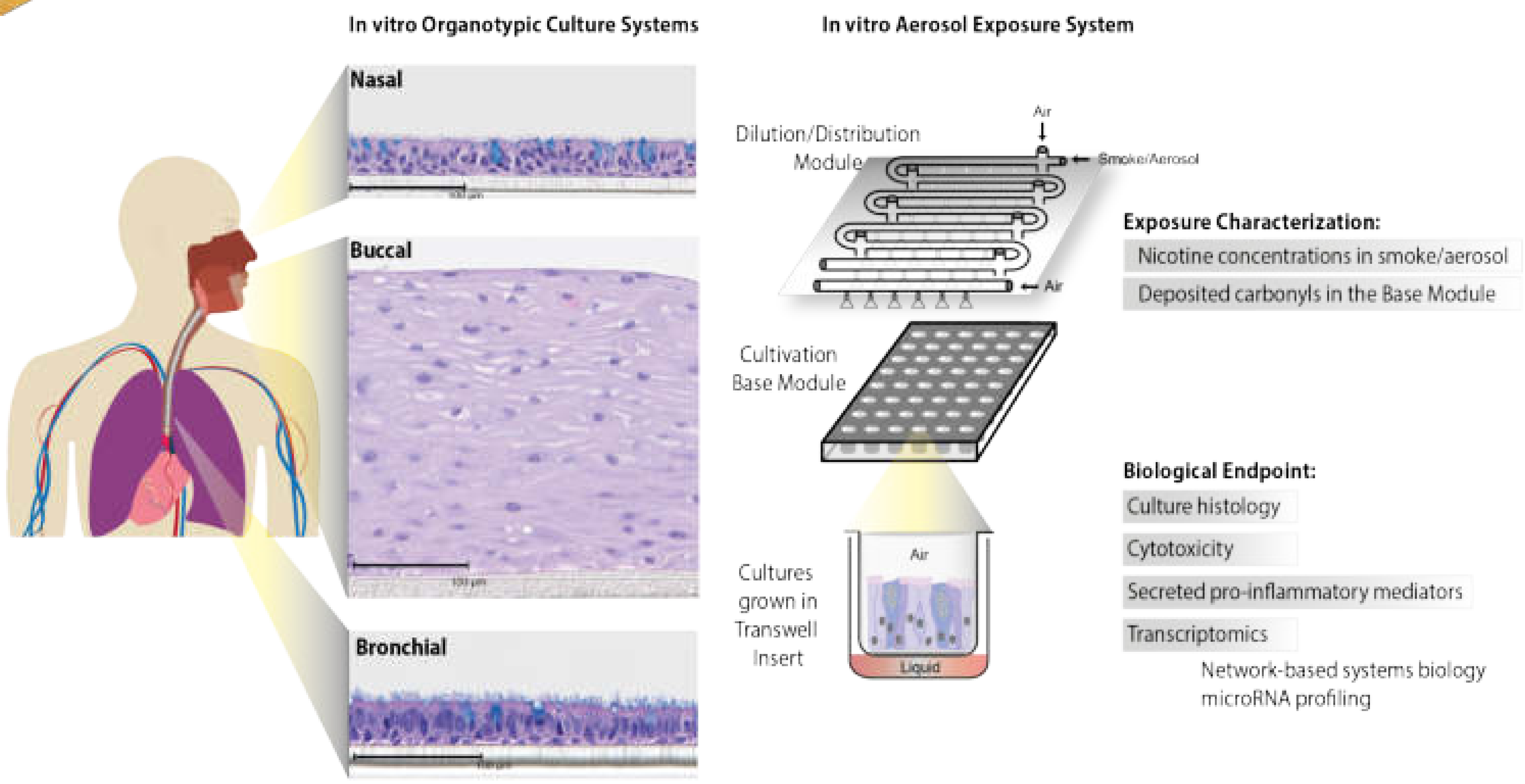




- Number of airways
- Airway length (mm)
- Airway diameter (mm)
- Branching angle (°)
- Inclination to gravity angle (°)

Oldham MJ, Lucci F, Foong C, Yeo D, Asgharian B, Cockram S, Luke S, Chua J, Hoeng J, Peitsch MC, Kuczaj AK. Use of micro-CT to determine tracheobronchial airway geometries in three strains of mice used in inhalation toxicology as disease models. Anat Rec (Hoboken). 2021 Feb 7. doi: 10.1002/ar.24596.

In vitro exposure studies



Existing *in vitro* aerosol exposure systems:

- often depend on aerosol generators
- provide continuous, uniform aerosol dilution
- based on constant unidirectional aerosol flow in exposure chambers
- may suffer from unanticipated and uncontrolled aerosol evolution and losses during aerosol transport and internal aerosol sampling

Iskandar AR, Titz B, Sewer A, Leroy P, Schneider T, Zanetti F, Mathis C, Elamin A, Frentzel S, Schlage WK, Martin F, Ivanov NV, Peitsch MC, Hoeng J. Systems toxicology meta-analysis of *in vitro* assessment studies: biological impact of a candidate modified-risk tobacco product aerosol compared with cigarette smoke on human organotypic cultures of the aerodigestive tract. Toxicol Res (Camb). 2017 May 29;6(5):631-653. doi: 10.1039/c7tx00047b. PMID: 30090531; PMCID: PMC6062142.

InHALES: the **In**dependent **H**olistic **A**ir-**L**iquid **E**xposure **S**ystem

- *Independent*: no aerosol generator required
- *Holistic*: the whole human respiratory tract is simulated
- *Air-Liquid Exposure System*: enables aerosol exposures to be conducted at the air-liquid interface

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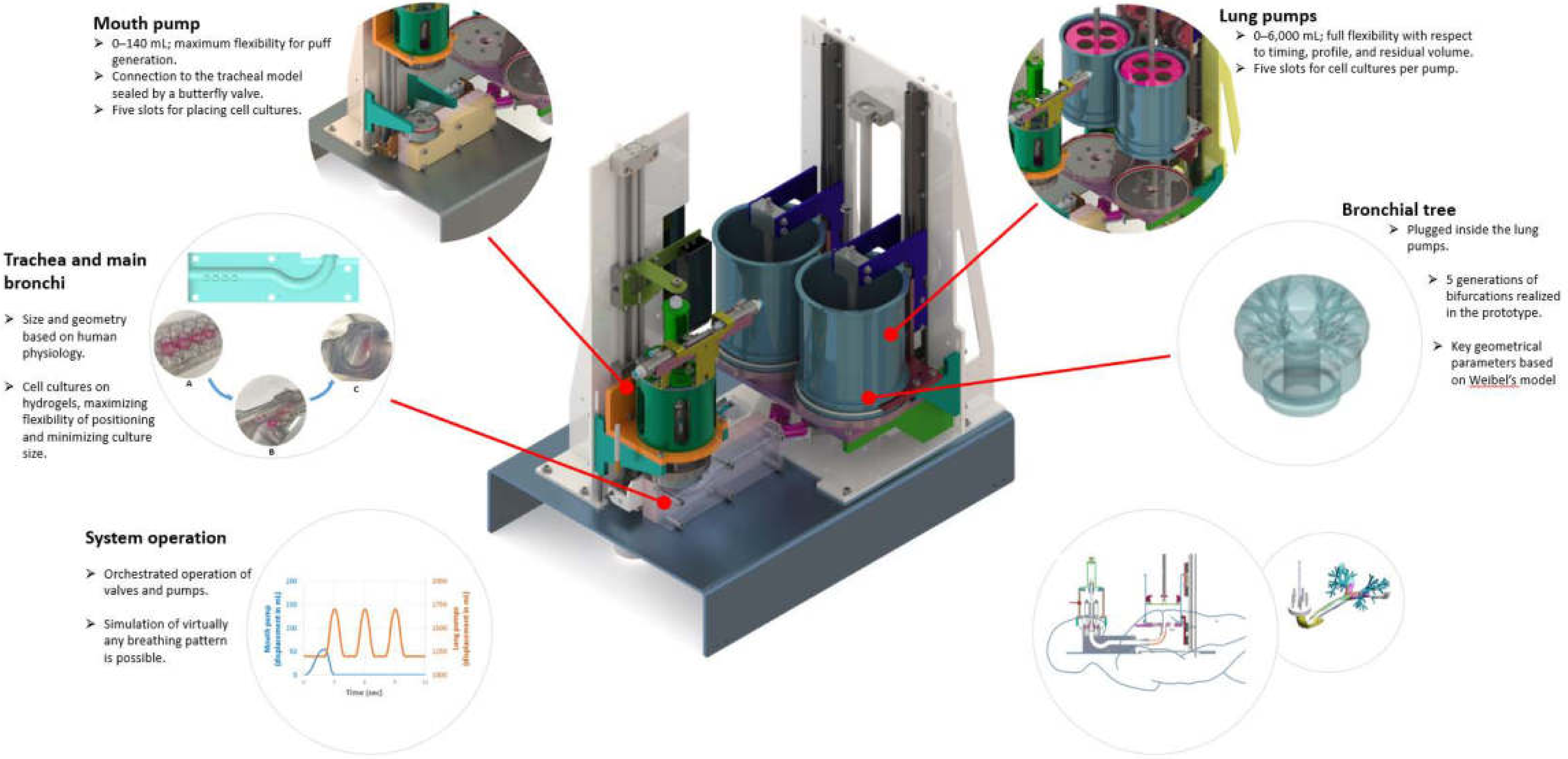
InHALES



Steiner S, Herve P, Pak C, Majeed S, Sandoz A, Kuczaj A, Hoeng J. Development and testing of a new-generation aerosol exposure system: The independent holistic air-liquid exposure system (InHALES). Toxicol In Vitro. 2020 Sep;67:104909. doi: 10.1016/j.tiv.2020.104909. Epub 2020 Jun 5. PMID: 32512146.

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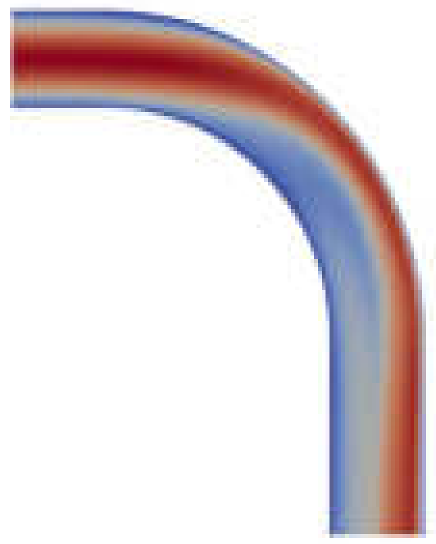
Computational Fluid Dynamics: AeroSolved

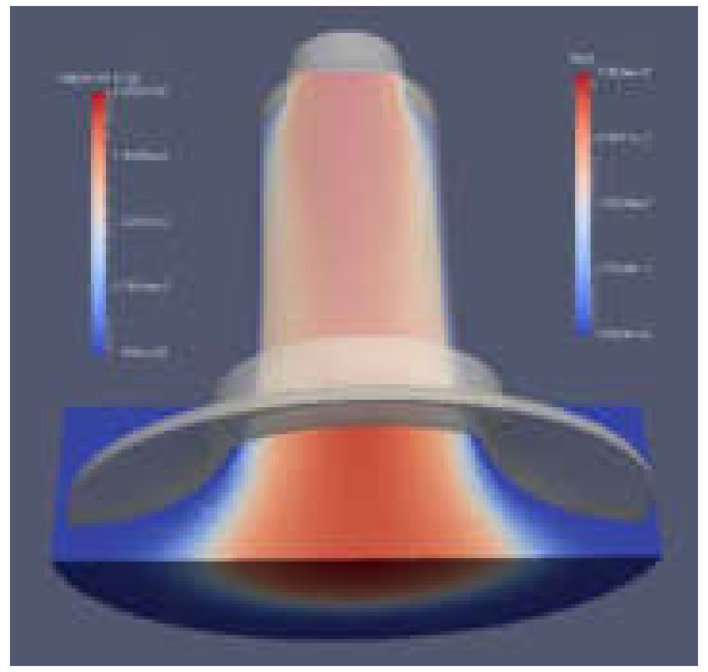
Computational fluid dynamics (CFD) code based on the OpenFOAM platform, for simulating generation, transport, evolution, and deposition of multispecies aerosol

AeroSolved

www.aerosolved.com

Open-source and publicly available at github





<u>Applications</u>

- Research on aerosol physics
- Development, characterization, and validation:
  - Inhalation devices
  - Aerosol generators
  - Aerosol delivery and exposure systems
- Wide range of applications, from industrial processes (such as spraying and emission reduction) to environmental and atmospheric sciences

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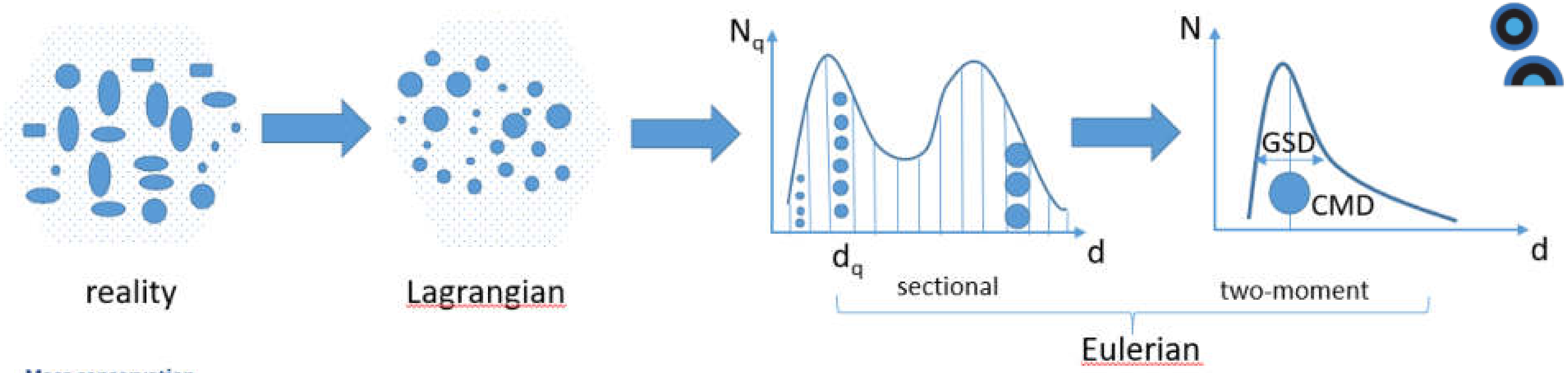
AeroSolved

Increasing complexity and level of details



Mass conservation $\partial_t \rho + \partial_j(\rho u_j) = -\partial_j[(1-\gamma)f_j]$

Momentum conservation $\partial_t(\rho u_i) + \partial_j(\rho u_i u_j) = -\partial_i p + \partial_j(\mu \tau_{ij})$; $i = 1,\ldots,3$

Energy conservation $\rho c_p(\partial_t T + u_j \partial_j T) = \partial_j(k \partial_j T) + \partial_j(\mu u_k \tau_{kj}) + \Im_h + Dp/Dt$

Transport of compounds in gas phase $\partial_t(\rho Y_n) + \partial_j(\rho Y_n u_j) = \partial_j(Y^{-1} f_j Y_n) + \Im_{Y_n}$; $n = 1,\ldots,N$

Transport of compounds in liquid phase $\partial_t(\rho Z_n) + \partial_j(\rho Z_n u_j) = \partial_j(Z^{-1} \gamma f_j Z_n) + \Im_{Z_n}$

Transport of particle number density $\partial_t(\rho M_q) + \partial_j(\rho M_q u_j) = -\partial_j(\rho M_q u^l_{j,q}) + \partial_j(\rho D_q \partial_j M_q) + \Im_{M_q}$; $q = 1,\ldots,Q$

Key development publications:

Frederix, E. M. A. Eulerian modeling of aerosol dynamics. From nucleation to deposition, PhD thesis, 2016

Frederix, E. M. A. et al. Characteristics-based sectional modeling of aerosol nucleation and condensation, J. Computational Physics, 2016

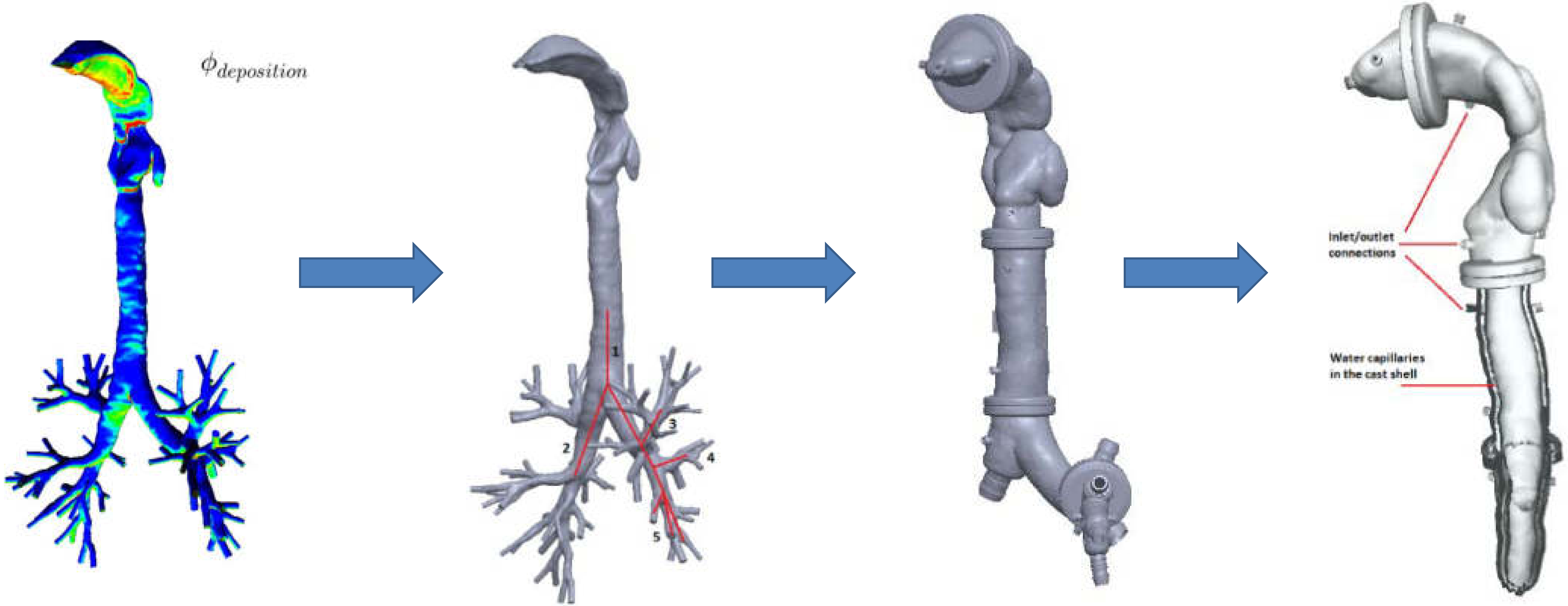
Frederix, E. M. A. et al. Application of the characteristics-based sectional method to spatially varying aerosol formation and transport. J. Aerosol Science, 2017

Full list of publications available at: www.aerosolved.com

- Longest PW, Kleinstreuer C, Computational Models for Simulating Multicomponent Aerosol Evaporation in the Upper Respiratory Airways, Aerosol Science and Technology, 2005, 39:2, 124-138, DOI: 10.1080/027868290908786
- Rostami AA, Computational Modeling of Aerosol Deposition in Respiratory Tract: A Review, Inhalation Toxicology, 2009, 21:4, 262-290, DOI: 10.1080/08958370802448987
- Corley RA, Kabilan S, Kuprat AP, Carson JP, Minard KR, Jacob RE, Timchalk C, Glenny R, Pipavath S, Cox T, Wallis CD, Larson RF, Fanucchi MV, Postlethwait EM, Einstein DR. Comparative computational modeling of airflows and vapor dosimetry in the respiratory tracts of rat, monkey, and human. Toxicol Sci. 2012 Aug;128(2):500-16. doi: 10.1093/toxsci/kfs168. Epub 2012 May 12. PMID: 22584687; PMCID: PMC3524950.
- Hofmann W, Modeling techniques for inhaled particle deposition: the state of the art. J Aerosol Med. 1996;9(3):369-88. doi: 10.1089/jam.1996.9.369. PMID: 10163662.
- Feng Y, Zhao J, Hayati H, Sperry T, Yi H, Tutorial: Understanding the transport, deposition, and translocation of particles in human respiratory systems using Computational Fluid-Particle Dynamics and Physiologically Based Toxicokinetic models, Journal of Aerosol Science, 151, 2021, 105672, https://doi.org/10.1016/j.jaerosci.2020.105672.
- Phalen RF, Hoover MD, Oldham MJ, Jarabek AM, Inhaled aerosol dosimetry: Research-related needs and recommendations, Journal of Aerosol Science, 155, 2021, 105755, https://doi.org/10.1016/j.jaerosci.2021.105755.
- Finlay WH, The mechanics of inhaled pharmaceutical aerosols – an introduction, 2001, Elsevier, https://doi.org/10.1016/B978-0-12-256971-5.X5000-7

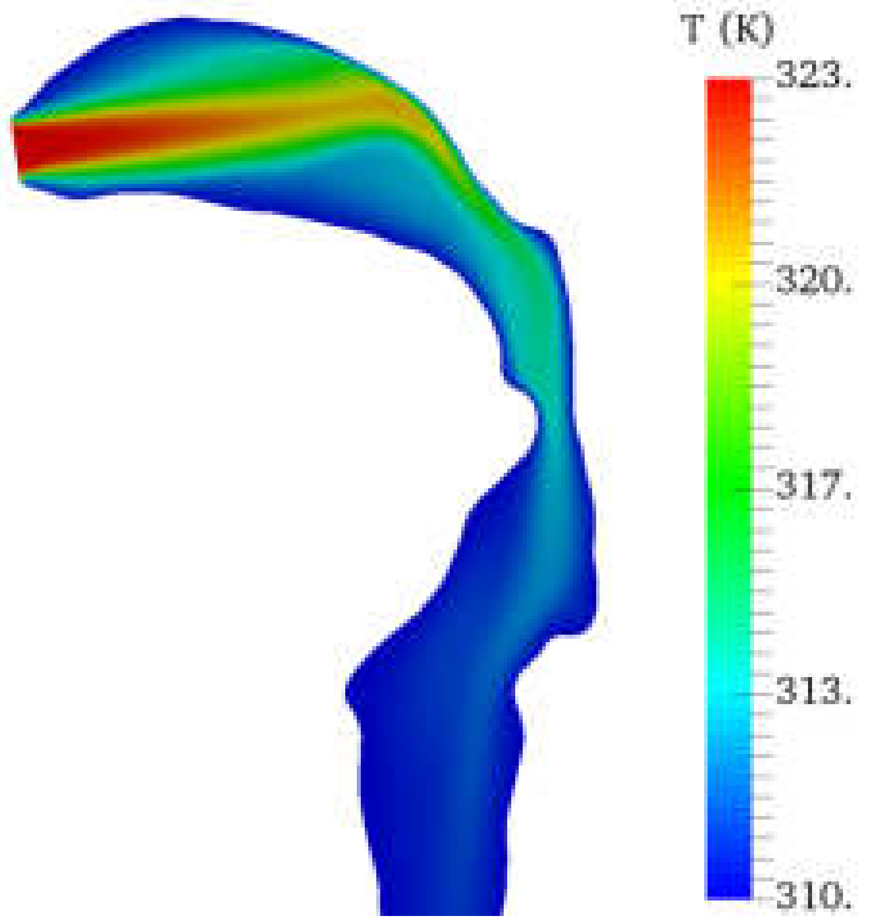
Developed human lung cast for experimental investigation

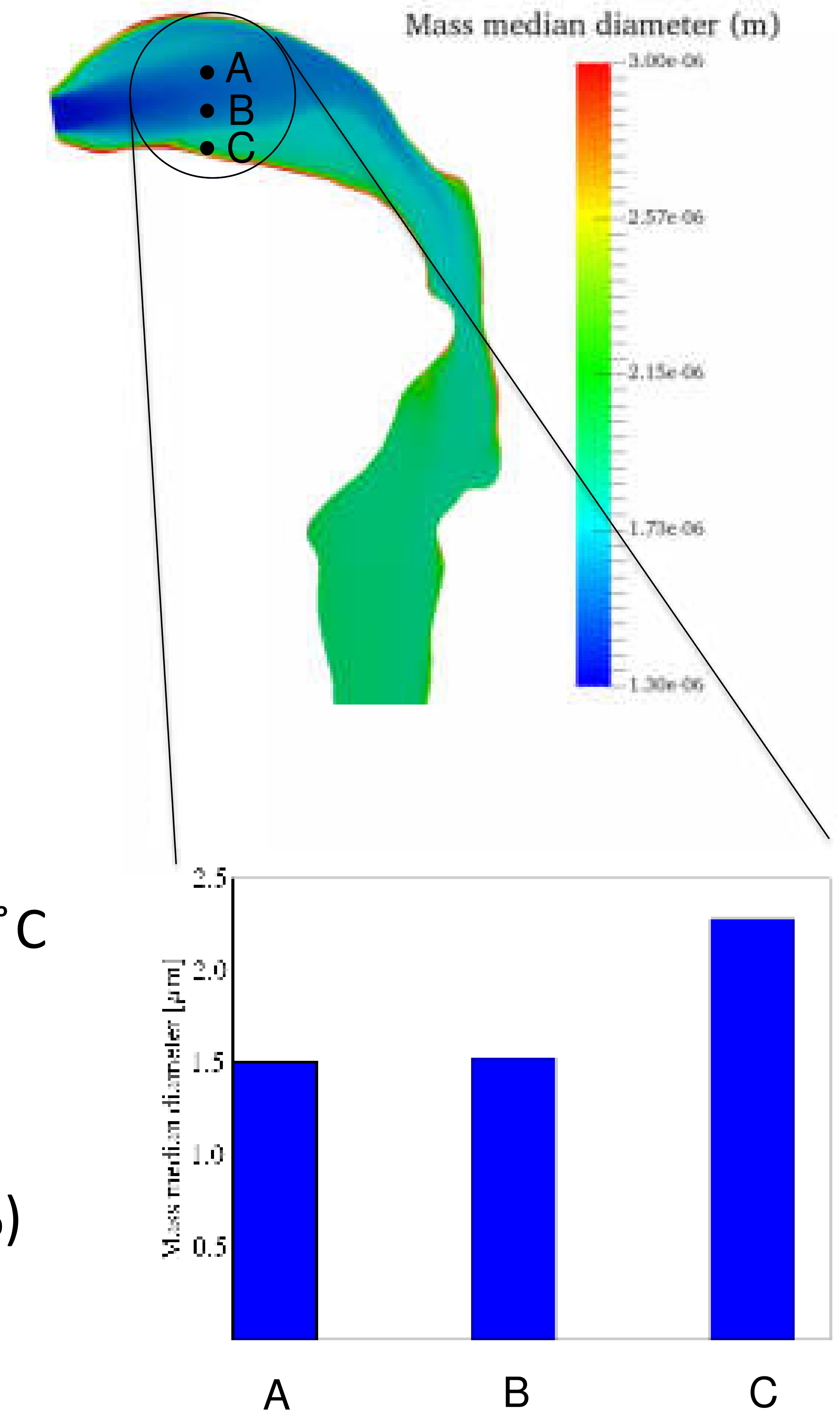
Cast geometry:
Zhang, Z., C. Kleinstreuer, and S. Hyun, *Size-change and deposition of conventional and composite cigarette smoke particles during inhalation in a subject-specific airway model*, Journal of Aerosol Science 46, 2012

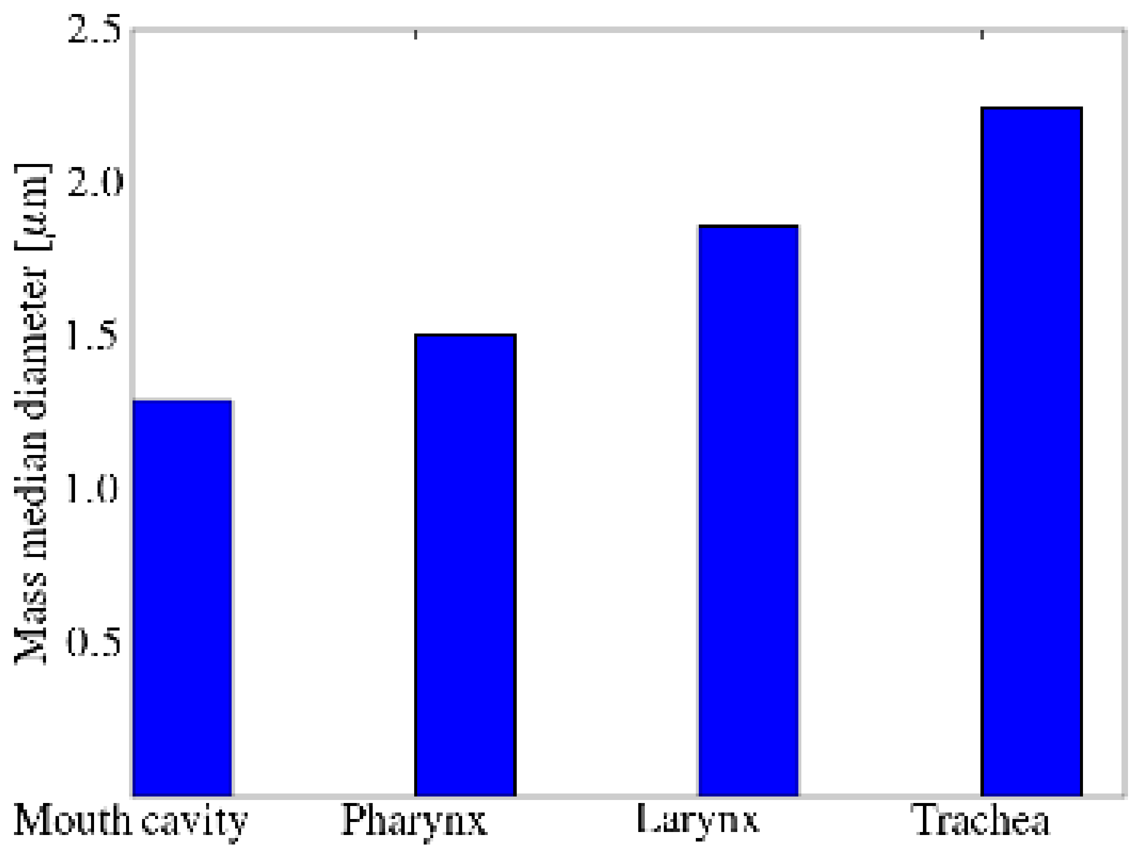


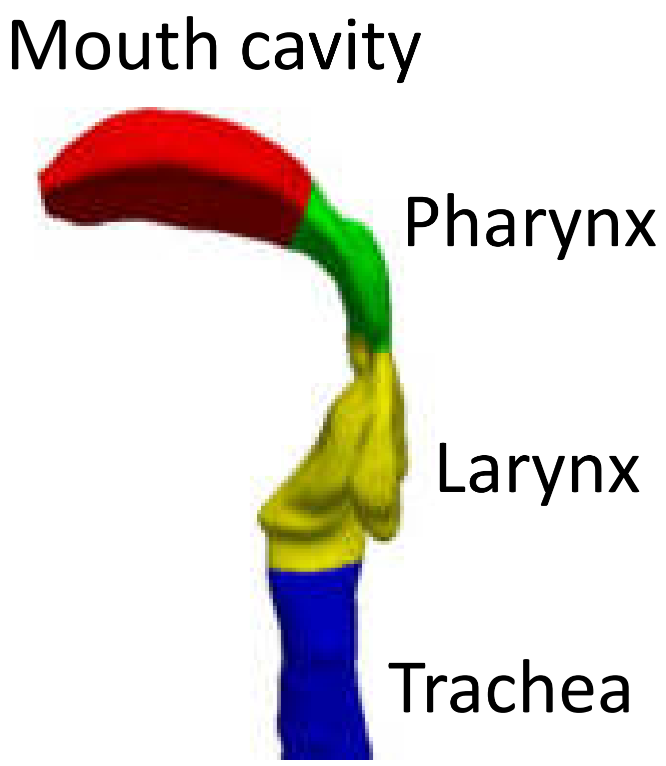
M. Asgari, F. Lucci, J. Bialek, B. Dunan, G. Andreatta, R. Smajda, S. Lani, N. Blondiaux, S. Majeed, S. Steiner, J.-P. Schaller, S. Frentzel, J. Hoeng, A.K. Kuczaj, (2019), Development of a realistic human respiratory tract cast representing physiological thermal conditions, Aerosol Science and Technology, 53:8, 860-870, DOI: [10.1080/02786826.2019.1612839](10.1080/02786826.2019.1612839)

Aerosol evolution in the cast model due to condensation







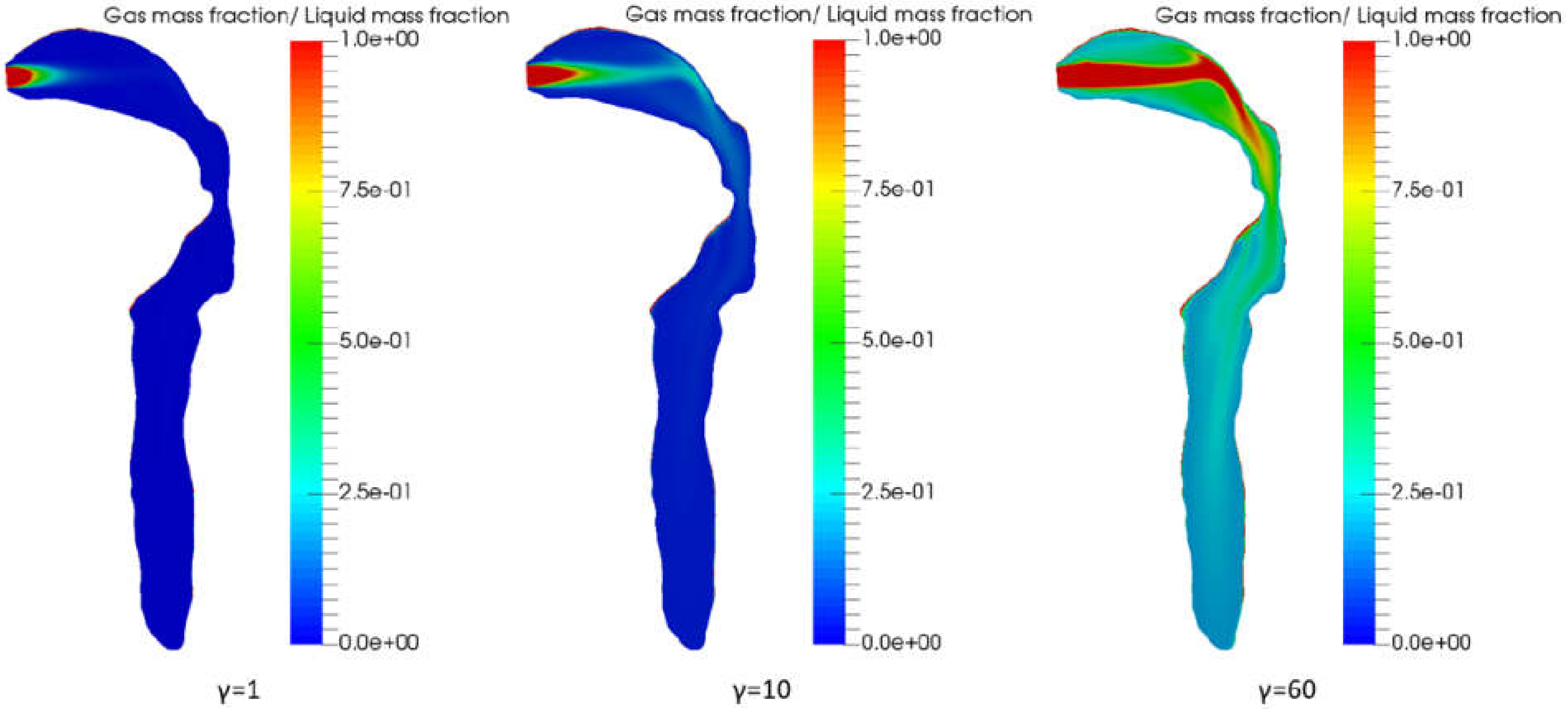


- Saturated aerosol inflow at 50˚C
- Flow rate of 1.5 L/min
- Glycerol (80%) and water (20%) mixture

- Decreased temperature: from 50˚C to 37˚C
- 98% relative humidity in the respiratory tract
- Particle condensational growth
- Change in deposition pattern due to evolution

M. Asgari, F. Lucci, A.K. Kuczaj, Multispecies aerosol evolution and deposition in a human respiratory tract cast model, Journal of Aerosol Science, 153, 2021
https://www.sciencedirect.com/science/article/pii/S0021850220302056

Influence of the activity coefficient for multispecies mixture (parameter study)

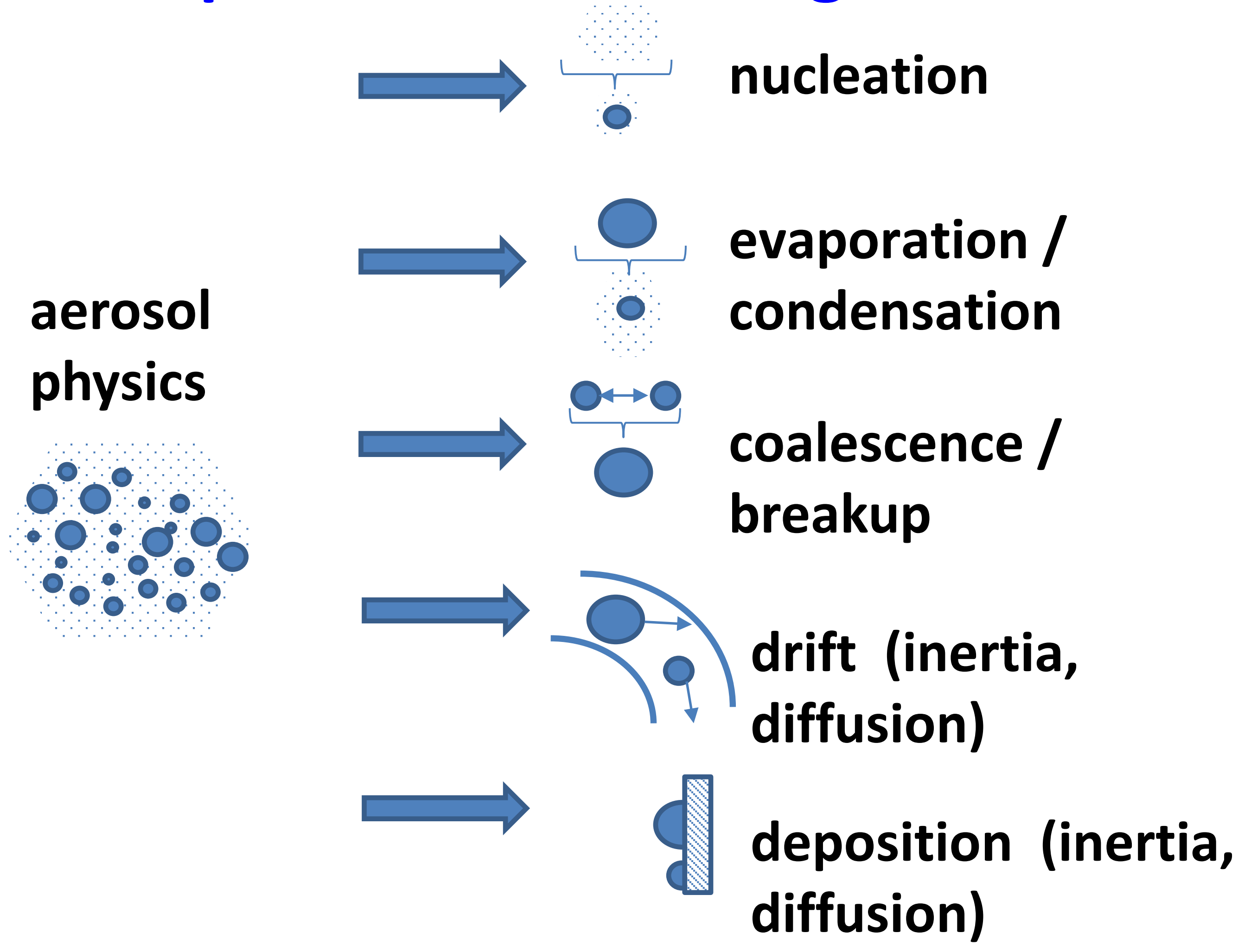


Increased gas phase mass concentration of traced substance owing to larger activity coefficient value for the same settings and liquid mixture composition.

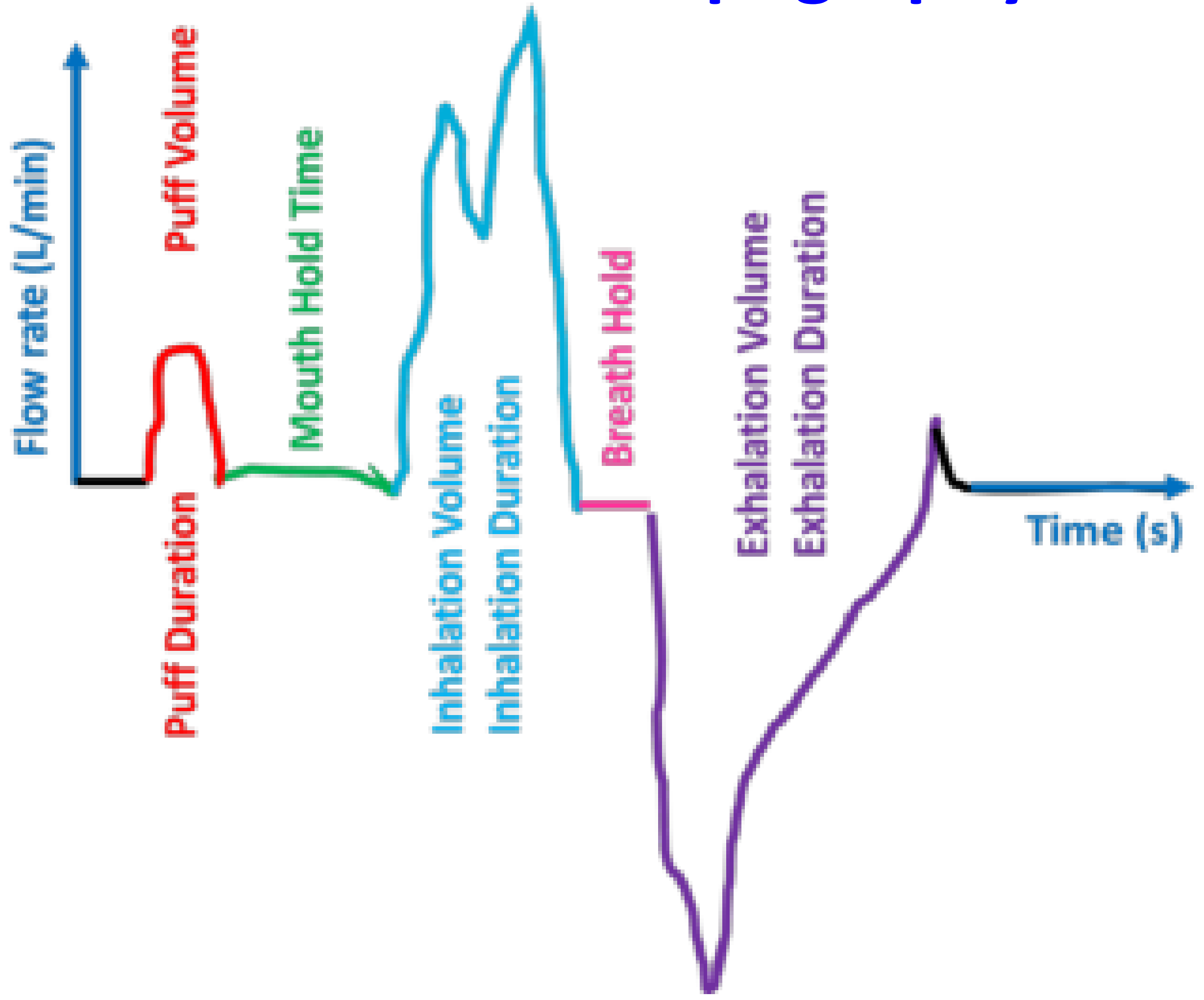
Development of 1D evolving aerosol dosimetry system codes

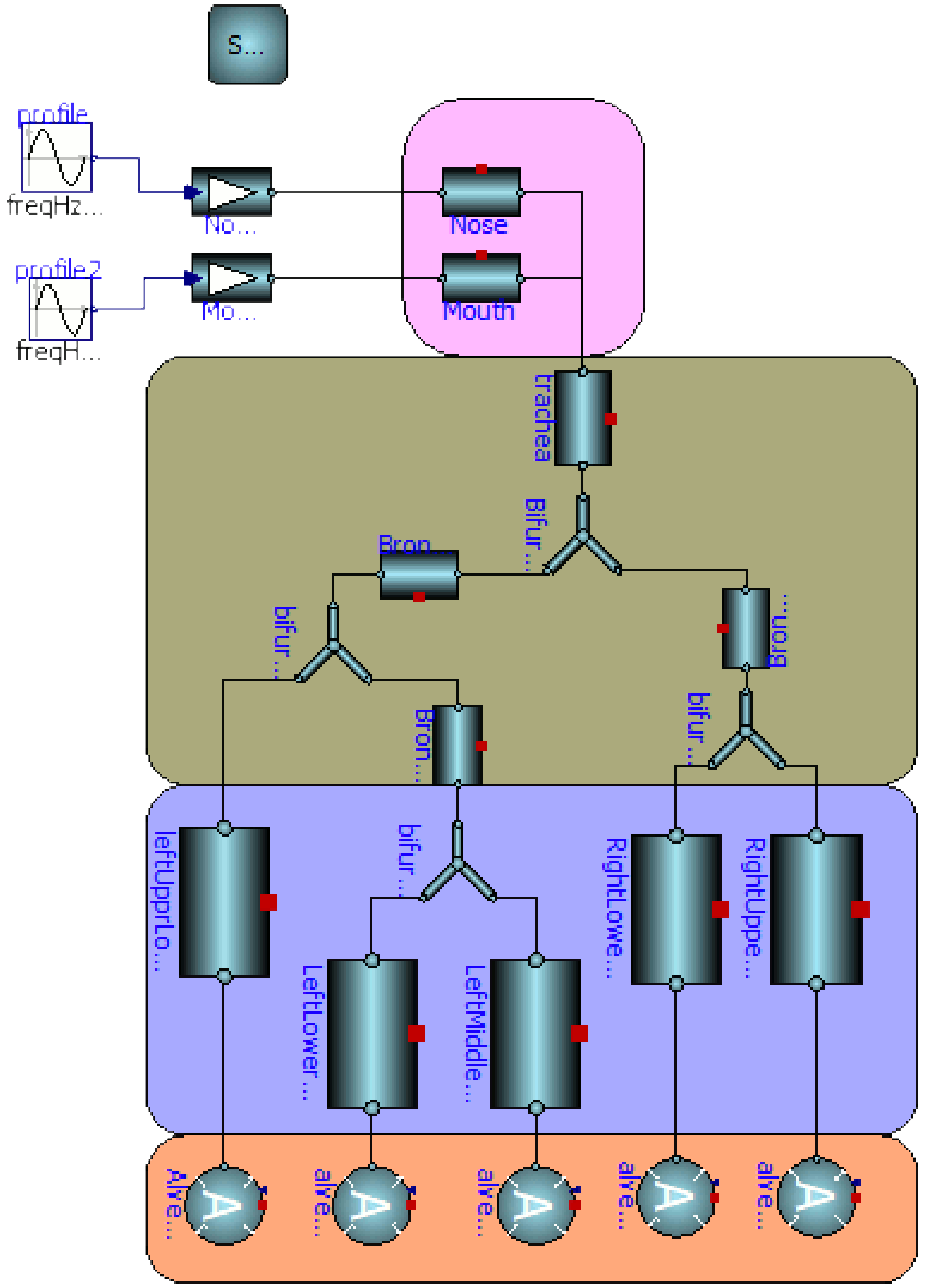
Airway anatomy

Deposition of evolving aerosol



Inhalation topography





Dosimetry predictions for evolving aerosols with complex physics and temporal/spatial resolution linked to PBPK (efficacy and tox assessment tools)

Summary

SOURCE → EXPOSURE → DOSE → BIOLOGICAL RESPONSE

- Exposure measurements are not sufficient for calculating delivered dose

- Aerosol delivery depends on the evolution of particles during inhalation, implying that often simple flow-based dosimetry scaling has limited application

- Chemical composition and deposition of aerosols depend on particle size (gas–liquid partitioning is important [e.g., activity coefficients for non-ideal mixtures])

- Development of simplified modeling approaches together with experimental validation and computational fluid dynamics (CFD) support has potential to improve dosimetry calculations

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