Every Breath You Take
Exposure Science & the Virtual Respiratory System

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SOT RASS Webinar
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Outline

- Imaging-based CFD models of the respiratory system
  - A decade’s worth of infrastructure development in one slide!
- Application #1: CFD models for vapor exposures
  - Ex: Comparative risk of reactive aldehydes in cigarette smoke
- Application #2: CFD models for aerosol exposures
  - Ex: Comparative risk of pathogen exposures
  - Ex: Personalized dosimetry models
- Emerging capabilities for multi-scale models
  - Airway & tissue mechanics
  - Impact of disease
  - NHLBI’s new LungMAP consortium
Our research program builds upon the foundation established by Julie Kimbell & colleagues who demonstrated the value of anatomically correct dosimetry models in assessing potential health risks for formaldehyde inhalation exposures.
Imaging Based Model Development

- 3D/4D MRI and CT
  - Mod-High resolution
  - Dynamic
  - Structure & Function
- What once took months, can now be done in days
- Personalized models are on the horizon

Challenges:
- Automating the process
- Improving resolution
- Decreasing X-Ray dose (CT) or imaging time (MR)
Suite of CFD Models

If you want a model, just ask!

Rabbit

Monkey

Rats & Mice

Human
In-vivo $^3$He MRI in live rats vs. CFD at steady-state inhalation

Basic Respiratory Maneuver

- Inhale
- Exhale
- Image Data Collection

Application 1:
CFD Models for Volatile Materials
Published assessments of the relative risk of cigarette smoke constituents based upon rankings of:
- RfC, RfD, Cancer Potency, etc. vs. Cigarette yields
- Acrolein > Acetaldehyde > Formaldehyde

Direct comparisons of site-specific doses at rat NOAELs under bioassay conditions vs. realistic smoking profiles provides a different (improved) perspective.

- Contact site irritation, inflammation, mutations
- Cytotoxicity & tumors in nasal tissues of rodents
  - Obligate nose breathers
  - Human oral inhalation

Obligate nose breathers vs. Human oral inhalation
CFD/PBPK Model for Aldehydes

- CFD with PBPK Airway Boundary
  - Each surface facet has 1D PBPK
  - Relied on published CFD & PBPK models for acrolein, formaldehyde, & acetaldehyde
  - Followed convention of Schroeter et al. (2008) nasal models for acrolein
  - Metabolism distributed between epithelium & subepithelium by cell type & region according to Bogdanffy et al. (1986), Keller et al. (1990), & Franks (2005)
  - Each compartment has 100 layers

- Transient CFD/PBPK simulation over full breathing cycle
  - Fully 2-way coupled 3D & 1D domains for wash-in vs. wash-out

\[
\begin{align*}
\frac{\partial C_a}{\partial t} + (U \cdot \nabla) C_a &= D_a \nabla^2 C_a \\
\frac{\partial C_t}{\partial t} &= -\left(\frac{V_{\text{max}}}{V_t} \right) \cdot C_t - K_f \cdot C_t + D_t \frac{\partial^2 C_t}{\partial x^2} \\
\frac{\partial C_b}{\partial t} &= -K_f \cdot C_b - \left(\frac{Q_b}{V_t} \right) \cdot C_b + D_b \frac{\partial^2 C_b}{\partial x^2}
\end{align*}
\]
CFD Models With PBPK Boundary Conditions

Airway surfaces blocked by cell type/region

Corley et al. (2012) *Toxicol Sci* 128, 500-516
CFD/PBPK Model Calibrations Against Nasal Extraction Data

Formaldehyde

- Kimbell et al. (2001)
- CFD/PBPK

Concentration (ppm):
- 2
- 6
- 15

Nasal Extraction (%):
- Morris & Blanchard (1992)
- CFD/PBPK Metabolism from Teegarden et al. (2008)
- CFD/PBPK Metabolism Reoptimized

Acetaldehyde

- Morris & Blanchard (1992)
- CFD/PBPK Metabolism from Teegarden et al. (2008)
- CFD/PBPK Metabolism Reoptimized

Concentration (ppm):
- 1
- 10
- 100
- 1000
- 1 (Cyclic)

Nasal Extraction (%):
- Kimbell et al. (2001)
- CFD/PBPK

Acrolein

- CFD/PBPK

Concentration (ppm):
- 0.6
- 0.9
- 1.8
- 3.6
- 4.5
- 9.1
- 4.4 (Cyclic)

Nasal Extraction (%):
Human Exposures to 0.6 ppm Acrolein, 6.9 LPM

Cmax (µg/g)

Steady State vs. Transient Inhalation

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Steady-State vs. Transient Inhalation

Human Exposures to 0.6 ppm Acrolein, 6.9 LPM

- Steady State
- Transient

C x T (µg*s/g)

Resp./Trans.  Olfactory  Nasopharynx  Larynx  Trachea  Right Main Bronchus  Left Main Bronchus  Right Superior Lobe  Left Superior Lobe  Right Middle & Inferior Lobes  Left Superior Lobe  Left Inferior Lobe
Simulations of Rat Inhalation Exposures

Acetaldehyde, 50 ppm

Acrolein, 0.2 ppm

Formaldehyde, 1 ppm
Tissue Concentrations – Rat Acrolein

Maximum Facet Concentration Profiles

Anterior Respiratory/Transitional

Epithelial/Subepithelial Boundary (40 µm)

Olfactory

Epithelial/Subepithelial Boundary (70 µm)

Concentration (ng/g)

Depth (µm)
Maximum Facet Concentration Profiles

Anterior Respiratory/Transitional

Olfactory
Tissue Concentrations – Rat Acetaldehyde

Maximum Facet Concentration Profiles

Anterior Respiratory/Transitional

Olfactory

Epithelial/Subepithelial Boundary (40 µm)

Epithelial/Subepithelial Boundary (70 µm)

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Simulation of Cigarette Puff


  - Acetaldehyde – 1028 ppm (857 µg/cig)
  - Acrolein – 94 ppm (100 µg/cig)
  - Formaldehyde – 108 ppm (61 µg/cig)
Simulation of Cigarette Puff

Ex: Acetaldehyde, 1028 ppm

Human Smoking Profile

Puff

Breath

Breath

Flow Rate (m/s)

Time (s)
Potential Dose Metrics

- Peak flux at any point along airway wall
- \( C_{\text{max}} \) for each cell type or region (compartment averaged)
- AUC for each cell type or region (compartment averaged)
- “Hot spots” as a function of concentration, surface area & depth within a cell type or region
  - Ex: Top 2.5% of AUC’s for all facets in each region
Tissue “Hot Spots” – Rat
Rank Ordered, Surface Areal Normalized, Facet AUC
(Integrated Conc. Over Time & Depth)

Acrolein
(Rat NOAEL = 0.2 ppm)

Formaldehyde
(Rat NOAEL = 1 ppm)

Acetaldehyde
(Rat NOAEL = 0.2 ppm)
Tissue “Hot Spots” – Human
Rank Ordered, Surface Areal Normalized, Facet AUC
(Integrated Conc. Over Time & Depth)

Acrolein
(Puff = 94 ppm)

Formaldehyde
(Puff = 108 ppm)

Acetaldehyde
(Puff = 50 ppm)
Flux vs. Tissue “AUC”

Acrolein
(Rat NOAEL = 0.2 ppm)

Surface Fluxes

AUC Tissue Concentrations

Flux (pg/cm² sec)
- 7.00E+02
- 6.12E+02
- 5.25E+02
- 4.38E+02
- 3.50E+02
- 2.62E+02
- 1.75E+02
- 8.75E+01
- 0.00E+00

AUC (Kg/s/m²)
- 1.30E-10
- 1.14E-10
- 9.75E-11
- 8.12E-11
- 6.50E-11
- 4.87E-11
- 3.25E-11
- 1.62E-11
- 0.00E+00
Flux vs. Tissue “AUC”

Formaldehyde
(Rat NOAEL = 1 ppm)

Surface Fluxes

AUC Tissue Concentrations

Flux pg/cm² sec
- 1.80E+03
- 1.58E+03
- 1.35E+03
- 1.12E+03
- 9.00E+02
- 6.75E+02
- 4.50E+02
- 2.25E+02
- 0.00E+00

AUC (Kg*s/m²)
- 4.50E-10
- 3.94E-10
- 3.38E-10
- 2.81E-10
- 2.25E-10
- 1.69E-10
- 1.12E-10
- 5.62E-11
- 0.00E+00
Flux vs. Tissue “AUC”

Acetaldehyde
(Rat NOAEL = 50 ppm)

Surface Fluxes

AUC Tissue Concentrations

Flux (pg/cm²·sec)
- 6.00E+04
- 5.00E+04
- 4.00E+04
- 3.00E+04
- 2.00E+04
- 1.00E+04
- 0.00E+00

AUC (Kg·s/m²)
- 3.00E-07
- 2.62E-07
- 2.25E-07
- 1.88E-07
- 1.50E-07
- 1.12E-07
- 7.50E-08
- 3.75E-08
- 0.00E+00
Potential Dose Metrics

- Peak flux at any point along airway wall
- Cmax for each cell type or region (compartment averaged)
- AUC for each cell type or region (compartment averaged)
- “Hot spots” as a function of concentration, surface area & depth within a cell type or region

- Ex: Top 2.5% of AUC’s for all facets in each region
- Combines attributes of peak concentrations and C x T
- Similar in concept to “Flux Bins” used by Kimbell et al. for formaldehyde
- Used to calculate Lifetime Average Daily Dose (LADD) for each species
  - Rat Anterior Resp./Trans. Epith & Olfactory Epithelium under inhalation bioassay conditions
    - Rat NOAEL: AUC_{10}/breath * bpm * 360 min/d * 5 d/7 d
  - Human oral inhalation using realistic smoking profile and cigarette yield
    - Human: AUC_{10}/puff * 11 puff/cig * no. cigs/d
  - Assume no site concordance (i.e. rat nose is sentinel for all human airways)
Comparative Dose Cigarette Smoke Constituents

**Acrolein**

(Rat NOAEL = 0.2 ppm; Puff = 94 ppm)

---Rat Olfactory LADD
---Rat Respiratory/Transitional LADD

Rank order:
Acrolein > Formaldehyde > Acetaldehyde

No significant differences when simulated as a mixture with competitive metabolism

**Formaldehyde**

(Rat NOAEL = 1 ppm; Puff = 108 ppm)

**Acetaldehyde**

(Rat NOAEL = 50 ppm; Puff = 1024 ppm)
Application 2: CFD Models for Aerosols
Example with *Bacillus anthracis*

  - Outside of the accidental release of anthrax in the former Soviet Union (FSU), the “Amerithrax” intentional release is the only modern human data set we have for inhalation anthrax exposure.
    - 22 cases, 11 pulmonary 50% mortality.
    - FSU – 77 confirmed cases, 66 deaths.

- Low level exposure (“wool sorters disease”).
  - Estimated 600-1300 particles – relatively little effects.

- Problem: we really do not know true risk of morbidity and mortality from exposure to anthrax spores.
  - What tolerances need to be established for exposure & cleanup?
  - How do animal studies relate to human exposures?
  - How long do we have to initiate treatment?

- Significant effort by EPA, DoD, DHS to develop predictive exposure-dose-response models based upon
  - In vivo studies with *rabbits*
  - In vitro studies with rabbits & human cell & tissue cultures
Species Differences

Rabbit

Rat
Key Processes in Disease

Inhaled spores
- **Deposition**
  - Location: Lung
  - Relevant processes/parameters: Gravitation, diffusion, convection, particle size, breathing rates

Deposited spores
- **Uptake/transport**
  - Location: Inside macrophages & dendritic cells (lung and lymph node)
  - Relevant processes/parameters: Uptake efficiencies, migration rates

Intracellular spores
- **Germination**
  - Location: Inside macrophages & dendritic cells (lymph node or during transit)
  - Relevant processes/parameters: Germination efficiencies, vegetative bacteria killing, toxin activity

Intracellular vegetative bacteria
- **Proliferation**
  - Location: Lymph node
  - Relevant processes/parameters: Bacterial replication rates, bacterial killing and inhibition of replication in the lymph node

Extracellular vegetative bacteria

Toxemia, bacteremia, death
- **Disease processes**
  - Location: Systemic circulation, target organs
  - Relevant processes/parameters: Host immune status (priming or immunocompromised), relationship between toxemia and death

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Comparisons of Spore Deposition

End Inhalation
Key Processes in Disease Progression

- Inhaled spores
- Deposited spores
- Intracellular spores
- Intracellular vegetative bacteria
- Extracellular vegetative bacteria
- Toxemia, bacteremia, death

**Deposition**

**Uptake/transport**

**Germination**

**Proliferation**

**Disease processes**

**LUNG**
- Inhalation
- Spore clearance from lung
- Spore clearance from MLN

**MEDIASTINAL LYMPH NODE**
- Uptake/Transport to MLN
- Spore clearance from MLN

**Spore clearance from lung**

**Spore clearance from MLN**

**Bacterial replication**

**Bacterial clearance from MLN**

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Personalized Models & Aerosol Deposition Data

- Benchmarking against regional deposition literature
- Ongoing human studies that compare healthy vs. diseased individuals (Darquenne, UCSD & Glenny, UW)

CT Scans Each Volunteer @ FRC and FRC + 1 L fitted with mask used in studies

Regional (bolus) vs total (continuous) deposition with measured ventilation and +/- Heliox in same position (supine) as CT scans
Personalized Models & Data

PD02
PD03
PD07
PD08
PD09
PD10
Personalized Models & Data

DEPOSITION IN HEALTHY
0.75L/S

- Nasal Breathing
- Oral Breathing
Emerging Capabilities:
Multi-Scale Models for Tissue Mechanics, Disease (& Aerosols)
Accounting for Tissue Mechanics
Multi-Scale Hybrid CFD Models

Computational models dependent upon understanding interactions between force transmission, mechanics & ECM (from Suki et al. (2005) J. Appl. Physiol. 98, 1892-1899)

4D µCT of rat lung during mechanical ventilation
Jacob & Lamm, Stable small animal ventilation for dynamic lung imaging to support computational fluid dynamics models, PLoS One, 6 (11), 2011

Strong need for high-resolution in vivo measurement tools & organotypic culture systems that address mechanics
Mechanics of the lung is implicit in its motion

Rat model of COPD
- Elastase-dosed rat (left lobe only)
- CT images 11 times over 1-sec breathing cycle
- Develop maps of ventilation and stress/strain relationships
Aerosol Studies in Rats with Lung Disease

Can CT-derived ventilation maps predict aerosol deposition patterns?
- 6 male SD rats administered elastase to one lobe
- 4C-CT imaging to develop airflow maps
- Exposed to 1µm FMS aerosols for 5 min followed by cryomicrotome/optical imaging
- Co-registered CT with cryomicrotome imaging

Multi-Scale Coupling for Disease

- Replace unrealistic airway outlet boundary conditions with 1D ODE System
  - Ex: Ginzburg RLC Circuits for the human lung (up to 23 generations)
    - R & L come from airway geometry
    - C comes from dynamic CT & MR P-V imaging

- Airflows driven from alveolar region
- Critical for improving pulmonary drug delivery under disease conditions

Kuprat et al. (2013) J. Comp. Phys. 244, 148-167
Multi-Scale Coupling for Aerosol Deposition

Bi-Directional Coupling Approach
(Asgharian & Price)

Ongoing studies with healthy vs. diseased rats & human volunteers

1D Navier Stokes + Mechanics + MPPD

Gen 4

Gen 5-23

Less CFD, More 1D

<5

10

15

20+

More CFD, Less 1D

Lung Generations for CFD

Applications

³He Gas

Microparticles

Nanoparticles

Gas Exchange

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Emerging Capabilities: LungMAP
Extending to the Cell

Cell Morphology Maps Exist for the Nose—Functional (e.g. metabolic) Maps of the Full Respiratory System Are **Inadequate** or **Non-existent**

Rat

Olfactory epithelium
Respiratory epithelium
Transitional epithelium
Squamous epithelium
Nostril

Monkey

Squamous epithelium
Respiratory epithelium
Olfactory epithelium
Nonciliated transitional epithelium
Lymphoepithelium

Human

Squamous epithelium
Respiratory/transitional epithelium
Olfactory epithelium

Create an open-access reference resource and comprehensive molecular atlas of the late-stage developing lung

- Utilize state-of-the-art molecular and imaging technologies to map and annotate the cell types and functions of the developing MOUSE and HUMAN lung

- Fill the knowledge gap in molecular/cellular events that drive lung development (alveologenesis)

- Provide tissues, reagents and data to the medical research community
NHLBI LungMAP Consortium

Four Research Centers
- Cincinnati Children’s Hospital Medical Center (Whitsett, PI)
- Children’s Hospital of Los Angeles (Wharburton, PI)
- University of Alabama at Birmingham (Ambalavanan, PI)
  - Yale School of Medicine (Kaminski, MPI)
  - UCSD (Hagood)
  - CMU (Bar-Joseph)
- PNNL (Ansong, Corley, Carson, Co-PIs)
  - Texas Advanced Computing Center (Carson)
  - Baylor College of Medicine (Ljungberg)
  - University of Washington (Frevert, Gharib)

Data Coordinating Center
- Duke University (Palmer, PI)
- RTI International (Clark, MPI)
- CCHMC (Whitsett)

Human Tissue Core
- University of Rochester Medical Center (Pryhuber, PI)
  - Seattle Children’s Hospital (Deutsch)
  - National Disease Research Interchange (Lonsdale)
  - International Institute for the Advancement of Medicine (Smith)
# Data Types Produced (Consortium)

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<th>UAB</th>
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Summary

- Time to develop 3D CFD models greatly reduced
- Models based upon realistic anatomy, physiology, physics of airflow, and material transport
  - Minimizes assumptions and extrapolations
  - Improves resolution in exposure-dose-response assessments
- Realistic breathing patterns matter (vs. steady-state)
- Site-specific cellular dose matters (vs. flux rates)
- Foundation established for multi-scale linkages:
  - Airway and tissue mechanics
  - Impact of disease on airflow and dosimetry
    - Sensitive populations
    - Pulmonary drug delivery
  - Cell functional atlas
- All models and data available
  - Templates enhance new model applications
Collaborators & Funding

**UW**
- Glenny
- Frevert
- Robertson
- Hlastala
- Pipavath
- Lamm
- Krueger
- Gharib
- Bassingthwaigte Cox

**MWLSC**
- Polissar
- Neradilek

**UC Davis**
- Hyde
- Plopper
- Buckpitt

**UCSD**
- Darquenne

**UT-TACC**
- Carson

**UC**
- Ljunberg

**CGC**
- Fowler

**BCM**
- Fanucchi
- Postlethwait

**PNNL**
- Corley
- Trease
- Ansong

- Minard
- Laskin
- Metz

- Einstein
- McDermot
- Qian

- Kuprat
- Wright
- Shankaram

- Kabilan
- Jacob
- Timchalk
- Straub

- Teegarden
- Richmond
- Perkins

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- Praxair 52168
- RJR OP53160R2
- EPA EP-C-09-006
- DHS HSHQPM-14-X-00037
- VAC 65907

- **NIEHS P01 ES011617**
- **NIBIB R21 EB008192**
- **NHLBI U01 HL122703**
- **DOE LDRD 40403 & 46109**
“All models are wrong, but some are useful”
-George Box

“It is better to light one candle than to sit and curse the darkness”
-Anon