Microfluidic Device mimicking Bifurcated Bronchioles for Analysis of Particle Flow in Pulmonary Environments

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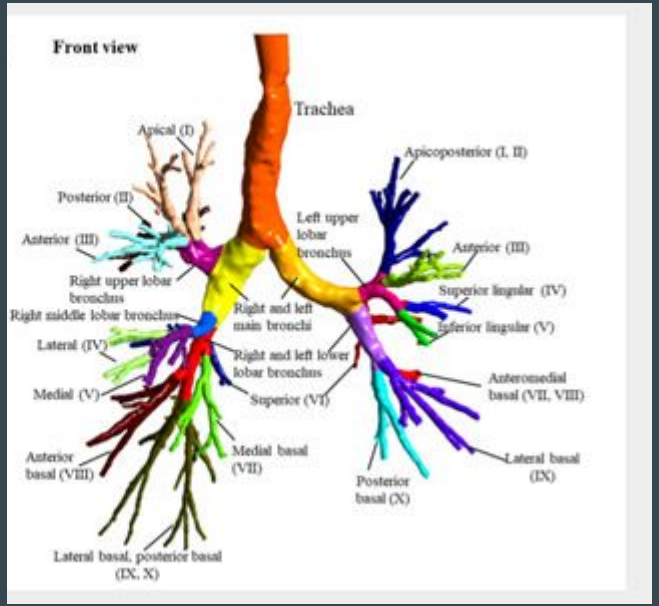
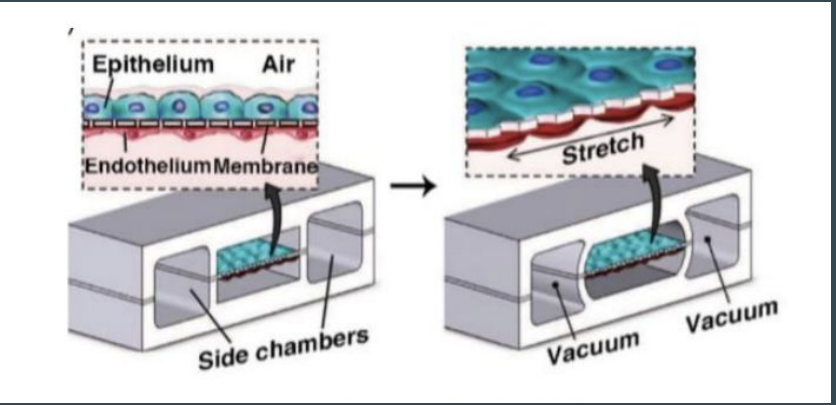
Background

- Bronchioles are the structures within the lungs that are responsible for delivering oxygen-rich gas to the alveoli
- Medical conditions that can affect and restrict the bronchioles include bronchitis, asthma, and chronic obstructive pulmonary disease (COPD).
- These condition effect over 35 million people each year.

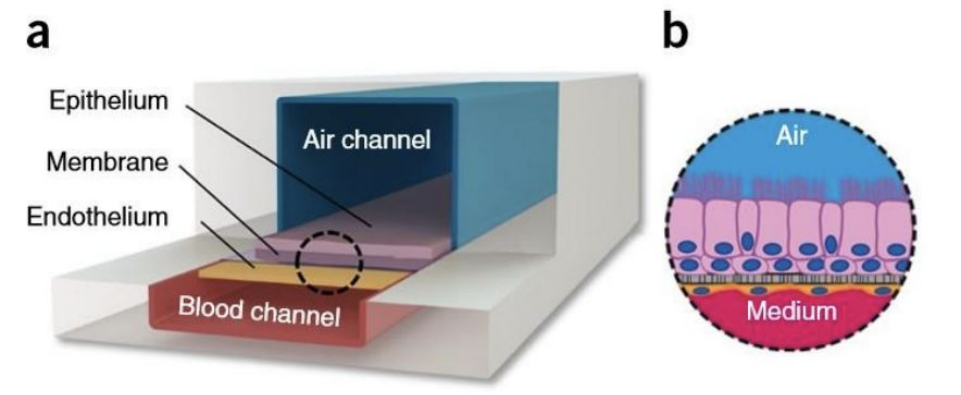


(Xu et al. 2019)

Current Lung on a Chip Technology



(Huh et al. 2010)

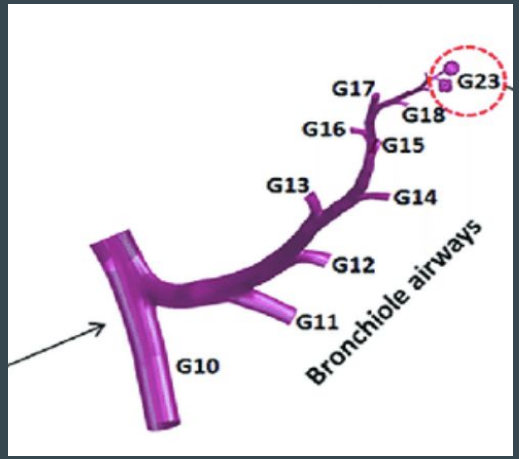


(Benam et al. 2016)

Why this Device is Needed

- Purpose: Create a cell-specific bifurcated analog of bronchioles to analyze particle flow in pulmonary environments

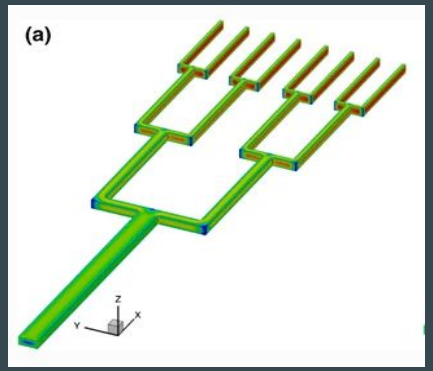
- Device (Bronchioles on a chip) Benefits:
  - A lung disease simulation
    - Asthma, inflammation, lung cancer, pulmonary fibrosis, lung injuries, etc
  - Realistic simulation of lung cells/tissue interactions by controlling their microenvironment
  - Provide a tool for studying a drug
  - More cheaper, accurate, and less invasive than animal experiments.



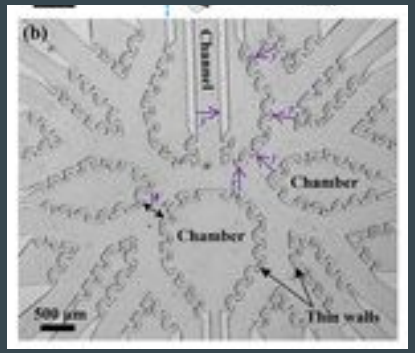
(Kim et al. 2017)

Device Proposal

- The proposed device:
  - Shall mimic the natural bronchial tree using multiple branching microfluidic channels
  - Shall have more vascular networks and surface areas for cell growth and interaction
  - Shall be lined with simple columnar epithelial cells (SCES) progressing to cuboidal epithelial cells (CES) to mimic the natural anatomy



(Zografos et al. 2015)

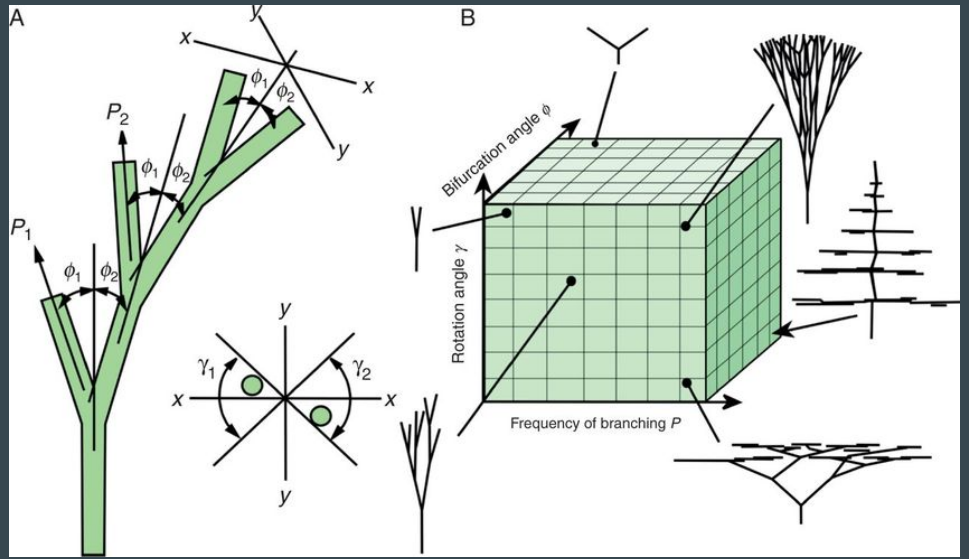


(Sznitman. 2022)

(Sznitman. 2022)

Device Theory

- The airways of the human lung (sometimes referred to as the bronchial tree) contain 23 levels of bifurcation (Reis, A.H. et Al.)
- Bifurcating channels can be created to design the microfluidic path. Challenges include:
  - How many bifurcations can fit on a chip
  - What is the smallest diameter (Feature) which can be designed into the chip
- Chips can be connected in series to allow additional levels, if sufficiently analogous connections can be made. (Can be characterized based on application).

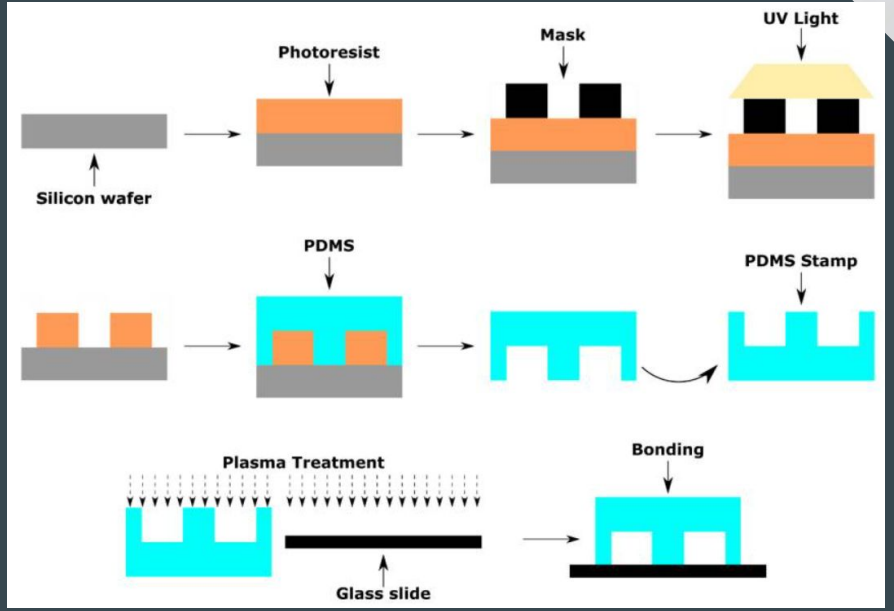


(J. Oyston et. Al.)

Device Fabrication Step 1

- Top and bottom microfluidic chambers
  - Polydimethylsiloxane-based (PDMS)
  - A soft-lithography technique
    - replica molding (REM):

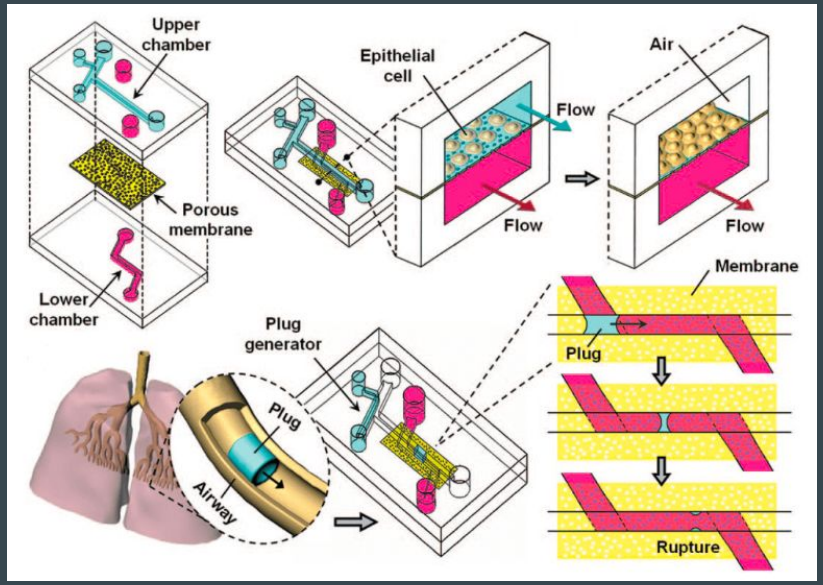
a. Computer aided design (CAD) to create a pattern
b. Photolithography techniques to develop a master
c. Fill the master mold with PDMS and cure
d. Remove the PDMS from the master
e. Bond the PDMS to a glass slide and performing plasma oxidation



(Puryear III et al. 2015)

Device Fabrication Step 2

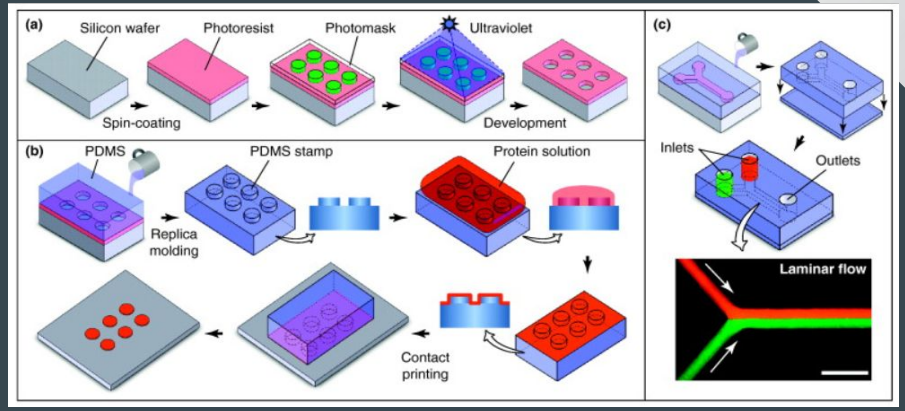
- A porous PDMS membrane (400 nm holes)
  - Cast against a deep reactive ion etching (DRIE) patterned silicon wafer (50 x 50 mm)
  - Located between top and bottom chambers.
- The microchannel size
  - 300 μm in width and 100 μm in height
  - Diameters of respiratory bronchioles and distal conducting airways
- Cell culture and stimulation
- Blood samples and flow conditions



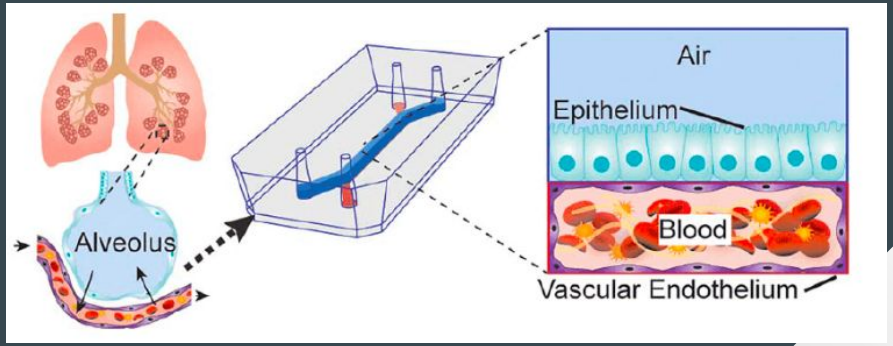
(Huh et al. 2018)

Device Fabrication - Materials

- PDMS
  - Coated with fibronectin and collagen to achieve a hydrophilic airway environment (Biocompatible)
- A semi-permeable PDMS membrane (400 nm holes)
- Silicon wafer
- photoresist material
- Glass slide
- Human alveolar type II epithelial cells (HAT2EC)
- Human lung microvascular endothelial cells (HLMEC)
- Human blood sample



(Huh et al. 2018)

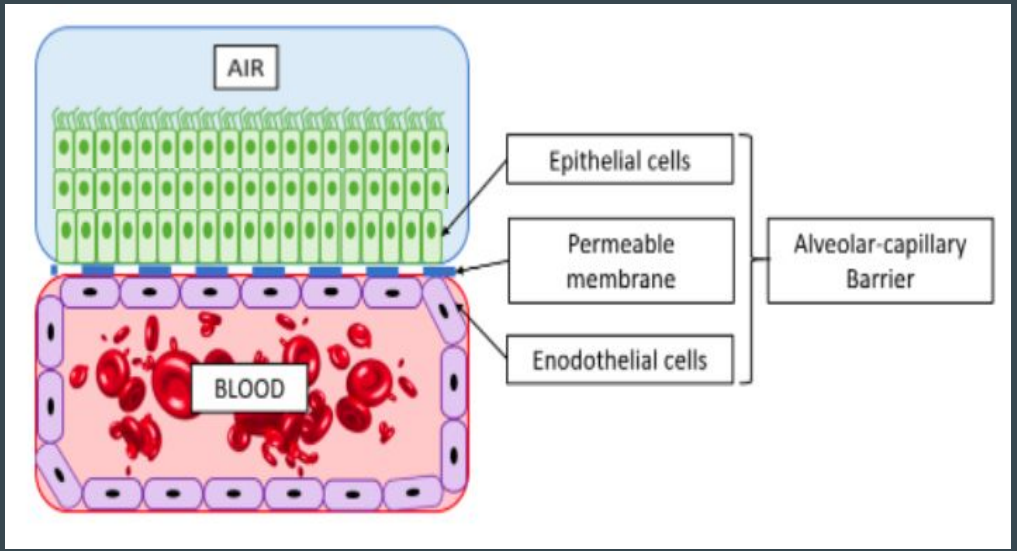


(Jain et al. 2017)

Device Fabrication - Techniques & Biocompatibility

- Why PDMS as a device material?
  - Good oxygen permeability & biocompatibility & low toxicity for long-term cell culture in an enclosed device.
  - High elasticity, allowing for on-chip cell manipulation
  - Optical transparency for cell imaging and assessment
  - Low-cost
- Why DRIE?
  - Achieve narrow structures on the silicon master mold.
  - Reduced material consumption
- Why soft lithography (SL) & Plasma oxidation (PO)?
  - SL:
    - A pattern resolution ranging from nanometer to micrometer precision
    - a relatively lower cost, easier setup, high compatibility with various materials
  - PO:
    - Assists in the bonding process and converts the PDMS surface from hydrophobic to hydrophilic
    - Provide a better biocompatible environment for the cell attachment
    - Better imitate *in vivo* fluid interactions.

Cell Harvesting

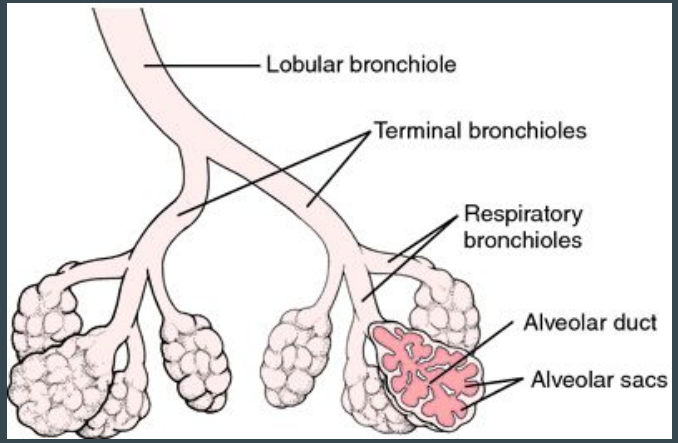


(Magnetics and Microhydrodynamics)

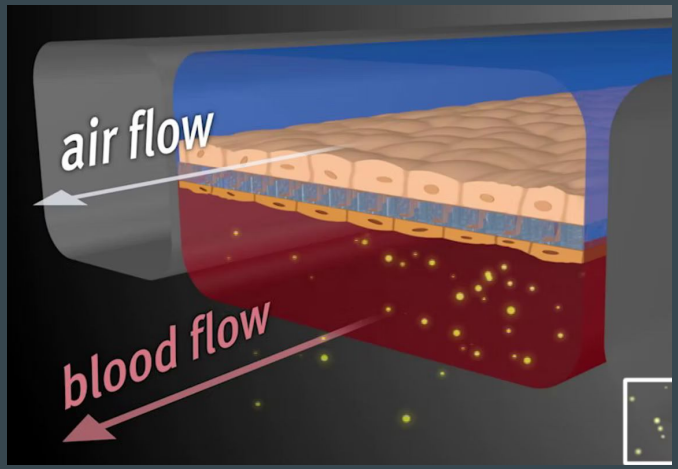
- Human alveolar type II epithelial cells (HAT2EC)
  - Cuboidal
  - Seeded into the top chamber
  - Cultured on the semi-permeable membrane while the apical and basal sides are perfused with culture media.
- Human lung microvascular endothelial cells (HLMEC)
  - Seeded on the ceiling and walls of the bottom chamber.

Device Considerations

- The device must be an accurate representation of diverging bronchioles
- Bronchiole lumen should be similar
- Cell lining must be similar to that of the true respiratory environment
- Mass exchange must occur in flowing synthetic blood



Miller-Keane Encyclopedia and Dictionary of Medicine, Nursing, and Allied Health, Seventh Edition.



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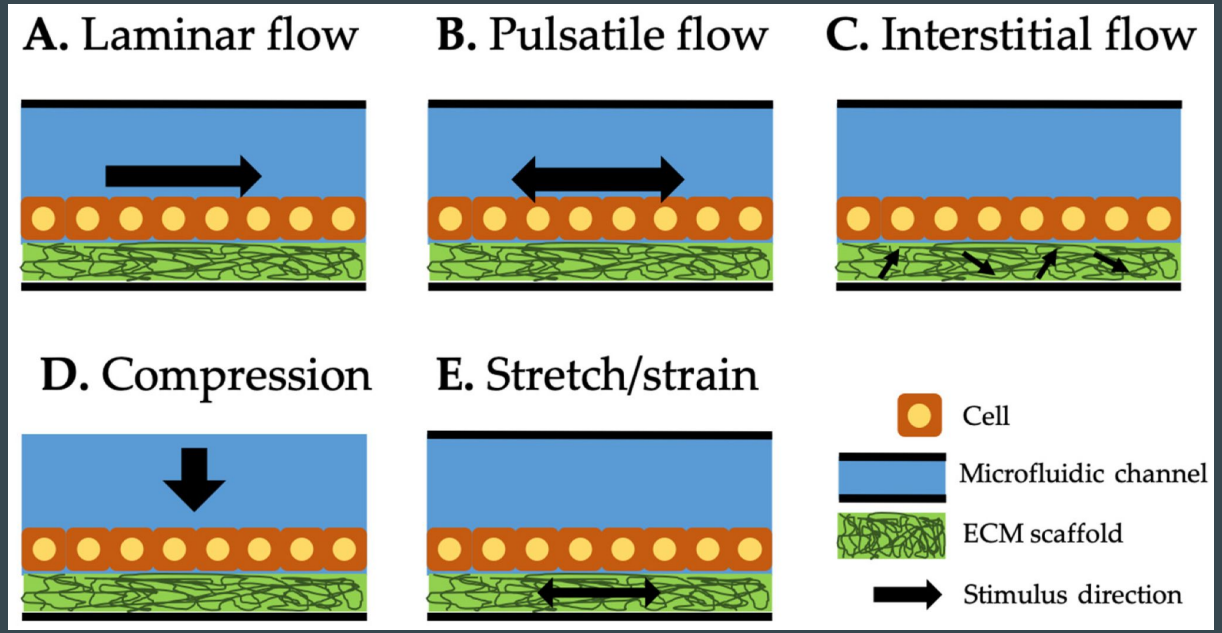
Device Testing

- Testing the device must be quantifiable
- Measuring mass transport
- Measuring air flow in mass per minute especially through bifurcations
- Measuring cell density



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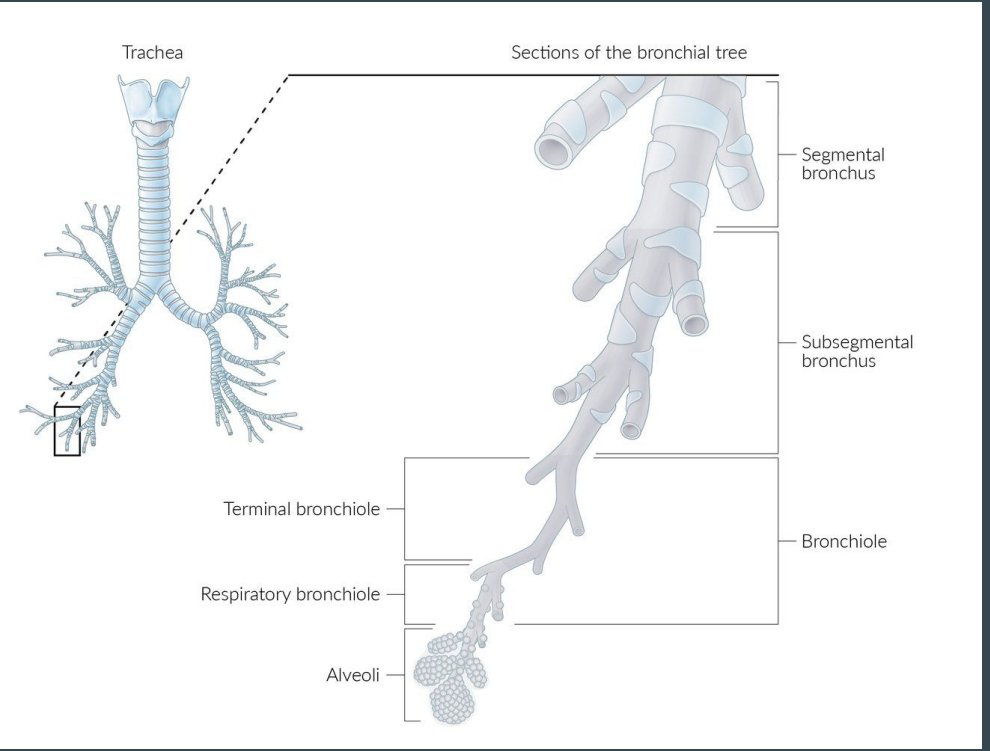
Device Limitations

- Blood/Bronchiole relation will be limited
- Cells will not regenerate or replace each other when damaged in the designed model
- Total modeled bifurcating volume will be far less than in a real lung



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Future Step

- Serve as a platform for testing nanoparticles entering the microenvironment
- Investigate the effects of HAT2EC and HLMEC on alveolar functions and pathologies, such as infections
- A more realistic airway shaped model can be created, advancing the understanding of disease mechanism
- The need for both conventional 2D cell culture methods and animal experiments will decrease.

Questions?

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