

Construction and Validation of an In Vitro Smoke  
Exposure System

Blaine Shillington, Undergraduate Student



Cindy Knall, Advising Faculty Member



## **Abstract**

This project proposes to research the effect of atmospheric pollutants in cigarette smoke on live respiratory cells. I will be assisting in this study by helping design and modify a preexisting plan, and then by assembling said design. This design system will then be used as an exposure system to study the controlled exposure of smoke and nicotine on live cells. I will be responsible with the assembly and testing of the exposure system, before laboratory testing can begin.

My goals for this project are to choose, acquire, and assemble all parts necessary for an exposure system used to expose cells to atmospheric pollutants. I will be researching the necessary parts needed for our system. I will then be responsible to make certain that all parts are received. Finally, assembly of the complete system and testing of its functionality will be required. I will be verifying that the system works properly and is easily manageable to be used in laboratory experiments. My final goal will be to create a system that properly exposes cultured cells to atmospheric pollutants according to research specifications.

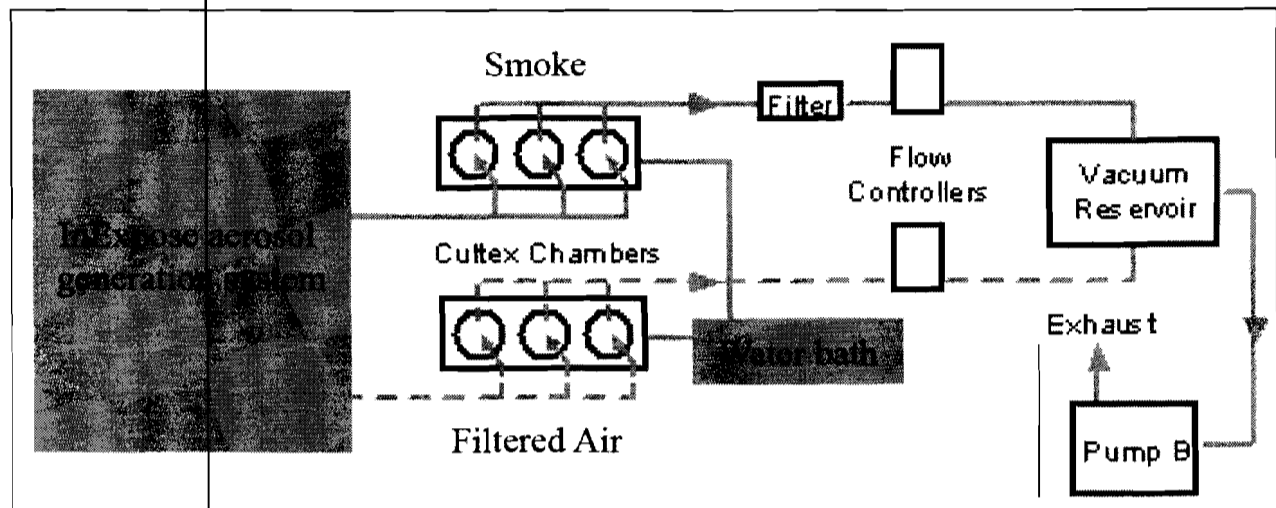
## **Introduction**

Little current information is known about the results of exposing live cultured cells to raw cigarette smoke. This project proposes to study the changes of respiratory cells after a complete and accurate exposure of certain concentrations of cigarette smoke and nicotine using a reliable exposure system. Historically, it has been known that exposure to atmospheric pollutants has long-term negative effects. It has not, however been properly understood to what extent certain doses of pollutants affect respiratory cells (Aufderheide and Mohr 2000). An air-liquid interface is ideal for culturing respiratory cells as it closely mimics morphology of mucosal epithelial cells and has advanced the culture of airway epithelial cells (Adler et al. 1990; Gruenert et al. 1995). Present knowledge has been difficult to attain, due to inconsistent exposure environments; specifically concerning dosage relationships. In past exposures, it became difficult to expose cells properly and completely. Newer technologies have also allowed for controlled exposure to certain toxins, and have helped to target certain airborne

pollutants (Bombick et al. 1997). These new technologies allow us to properly and evenly expose cultured cells and control exposure doses. It is with these technologies that we expect to advance knowledge of this field of study.

### Research Design

The proposed design consists of a smoking machine, used to smoke cigarettes and produce pollutants, flow control systems, and an exposure system, used to directly pass smoke over and through respiratory cells.



A sample method to be used is as follows.

#### *For assembly*

- Decide on parts needed.
- Order required parts and pieces.
- Assemble Preliminary design.
- Disassemble and modify.
- Reassemble and finalize design

#### *For Testing*

- Functionality test.
- Review/ analyze completed design.
- Test for Flow rates.

- Use flow rate controllers to verify constant flow
- Verify flow pattern
- Test for exposure system.
- Verify completed design.

Upon testing, tables will be produced representing various flow rates according to smoking machine control. I will relate the flow of the system to the parameters of the laboratory requirements, and verify that completed system functions according to specifications.

**Anticipated Results**

Upon testing the completed exposure system, I expect to be able to create a data table(s) representing the various flow rates capable of being produced by the exposure system. I believe that lower flow rates will clearly be relational to lower amounts of cigarette smoke/ nicotine exposure, and that higher flow rates will be representative of higher smoke/nicotine exposure. I expect that the system will ultimately function to properly expose cells to smoke at a controlled variable rate. It is also to be expected that cells will exhibit change after significant exposure to atmospheric pollutants.

**Budget**

Stainless steel and plastic tubing	Various quantity	Various cost each	\$500.00
Bath, Brinkmann heating circulator	1ea	\$1300.00	\$1300.00
Flow controllers, Sierra MattTrack	2 ea	\$1100.00	\$2200.00
Air Pump	1 ea	\$1000.00	\$1000.00
<b>Total</b>			<b>\$5000.00</b>

## **Budget Justification**

- Stainless steel and plastic tubing.

Tubing is necessary to connect all parts of system. This permits controlled air flow throughout.

- Bath, Brinkmann heating circulator.

Bath is used to regulate temperatures in the exposure system. Bath helps to create a climate much resembling the human respiratory tract.

- Flow controllers, Sierra MassTrack.

Flow controllers are required to monitor and regulate the flow of air and smoke through the exposure system. They allow for controlled exposure and dosage of pollutants.

- Air pump.

The Air pump provides the power to pull air through our exposure system and over our cells, exposing them to the airborne pollutants. It is therefore a crucial part of our system.

## **Project References**

Aufderheide M, Knebel JW, Ritter D. (2003) An improved in vitro model for testing the pulmonary toxicity of complex mixtures such as cigarette smoke. *Exp Toxicol Pathol.* 55:51-7.

Adler KB, Cheng PW, Kim KC. (1990) Characterization of guinea pig tracheal epithelial cells maintained in biphasic organotypic culture: cellular composition and biochemical analysis of released glycoconjugates. *Am J Respir Cell Mol Biol.* 2:145-54.

Gruenert DC, Finkbeiner WE, Widdicombe JH. (1995) Culture and transformation of human airway epithelial cells. *Am J Physiol.* 268:L347-60.

Bombick DW, Bombick BR, Ayres PH, Putnam K, Avalos J, Borgerding MF, Doolittle DJ. (1997) Evaluation of the genotoxic and cytotoxic potential of mainstream whole smoke and smoke condensate from a cigarette containing a novel carbon filter. *Fundam Appl Toxicol.* 39:11-7.

## **Project Timeline**

November until Early January: Brainstorm Parts plan, Order crucial pieces.

January: Begin Spring semester; All parts ordered by Feb. 1<sup>st</sup> deadline.

February 15<sup>th</sup>: Receipt deadline; begin assembly.

Late January until February: Assemble exposure system for March 1<sup>st</sup> deadline.

March: Testing, design verification. Data collection complete for March 30<sup>th</sup> deadline.

Early April: Data processing, final project details.

April 13<sup>th</sup>: Project Completion date.

## **Appendix**

### *Areas of Study*

2005 to present

Undergraduate study, University of Alaska Anchorage. Department of Engineering, Mechanical Engineering

2001 to 2005

High school study. Chugiak High School(graduated) and Sacred Heart Catholic High School.

### *Additional Work*

2005 to present

Volunteer work with youth, Skyline Family Fellowship Free Methodist Church.

2002 to present

Volunteer work, music ministry. Skyline Family Fellowship Free Methodist Church.