Laryngeal Bioreactor

Midsemester Presentation

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Rebecca Stoebe – Communicator

Dan Thompson – Team Leader

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Dr. Nathan Welham, Dr. Yutaka Yoya, Dr. Steve Lee - Clients

Outline

- Problem Statement
- Client Description
- Significance
- Last Semester Summary
- Initial Testing
- Goals
  - Testing
  - Design Improvements
  - Documentation
- Budget
- Timeline

Problem Statement

- Bioreactor capable of decellularization and recellularization of laryngeal tissue
- Allow horizontal fixation of larynx and ability to expose tissue to both air and media through automated controls
- Provide separate environments for vasculature, inner lumen and exterior of tissue
- Allow easy access to tissue while minimizing media used
- Use of biocompatible, autoclavable materials
- Allow function of bioreactor in lab, incubator, and refrigerator

Larynx Anatomy
<http://www.intechopen.com/books/innovative-rheumatology>

Client Description

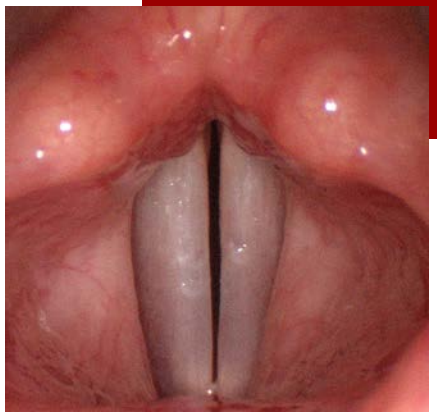
- Dr. Nathan Welham
- Assistant Professor at UW School of Medicine
- Specializations:
  - Speech-language pathology
  - Treatment of disorders of airway and voice
- Dr. Yutaka Toya, Dr. Steve Lee

Dr. Nathan Welham

http://www.surgery.wisc.edu/system/assets/177/Welham_Nathan_2013_ForWeb_profile.jpg%3F1373387179

Significance

- Need for larynx transplant:
  - 136,00 cases of laryngeal cancer per year globally
- Low success rate for laryngeal transplant
- No known currently available bioreactor for laryngeal tissue



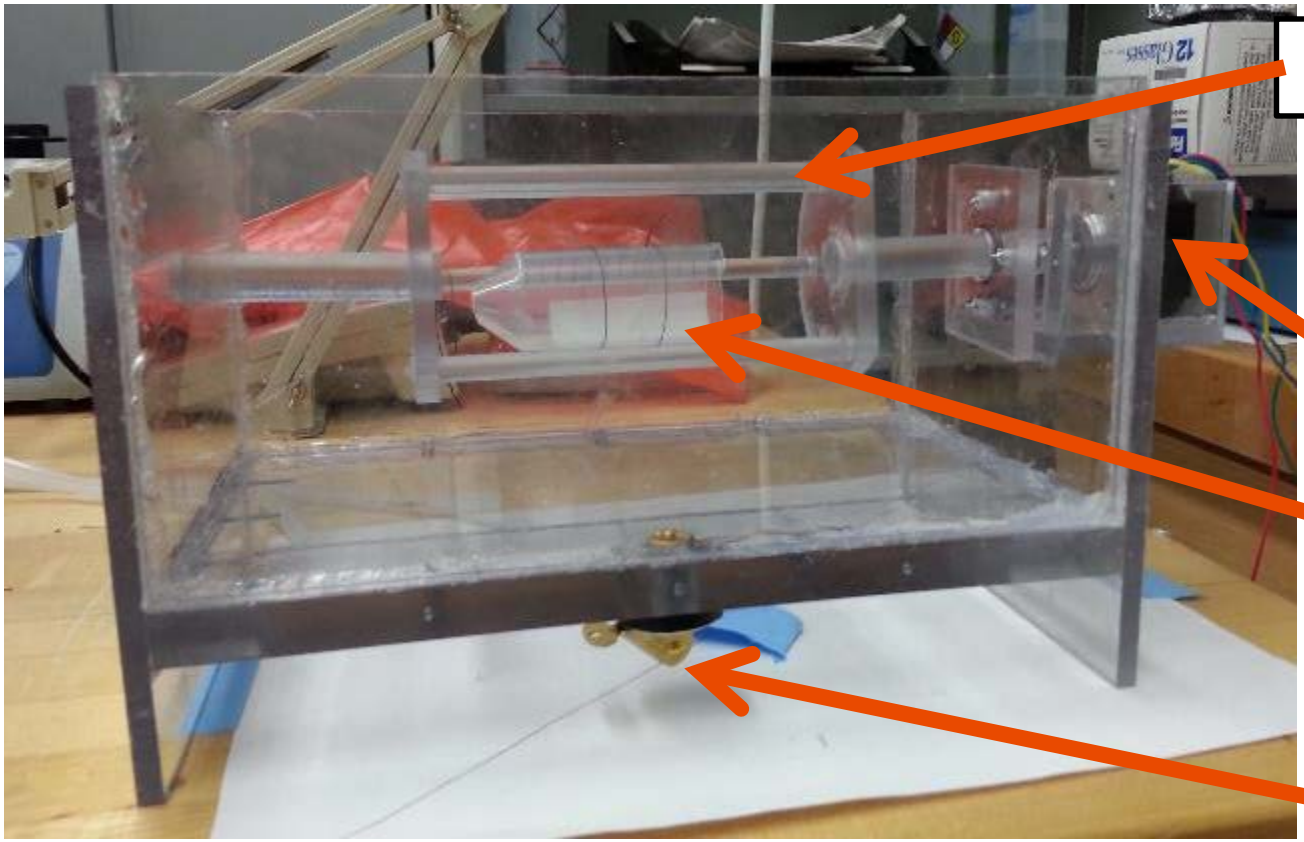
Healthy Vocal Folds

<http://voicedoctor.net/media/normal-vocal-cord/normal-female-vocal-cords>.

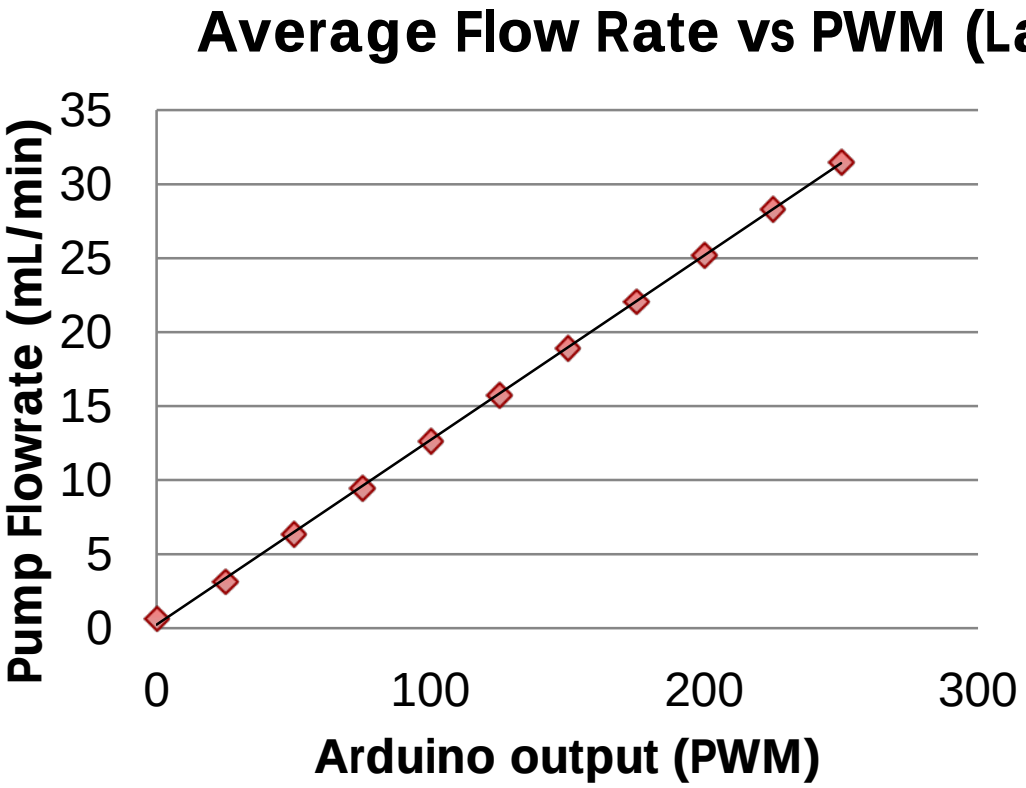
Cancerous Vocal Folds

<http://www.massgeneral.org/voicecenter/multimedia/>

Last Semester – Final Design



Last Semester – Testing

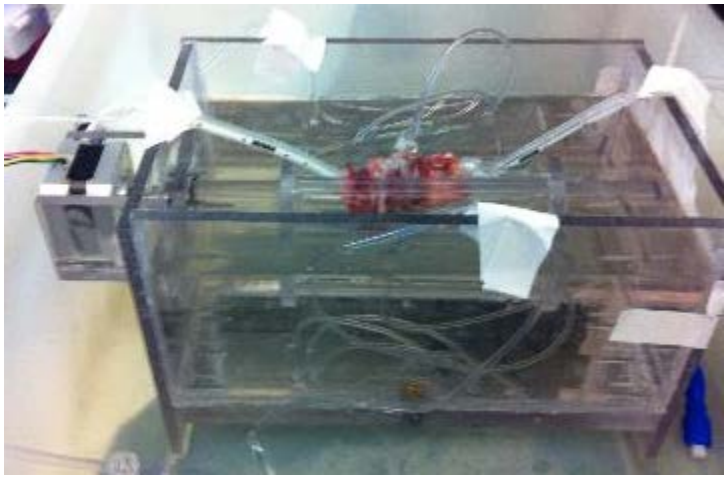


◆ Average Flow…
$y = 0.1247x + 0.2308$
$R^2 = 0.9998$

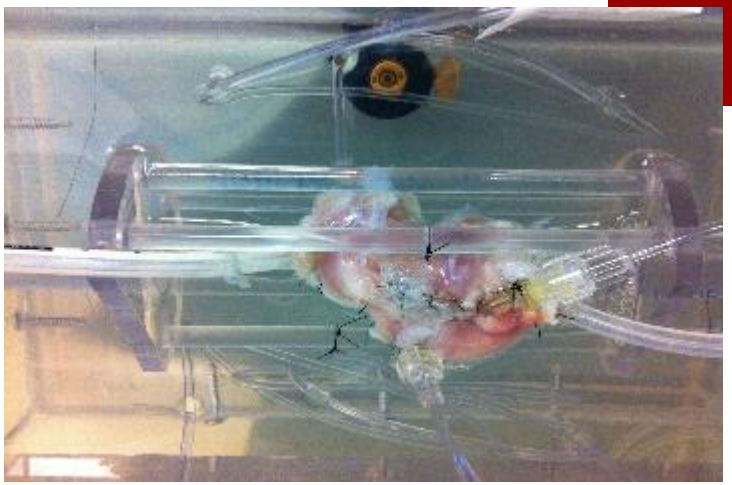
Note: Error bars not included due to data point consistency

Pump testing

Current preliminary testing



Current prototype undergoing testing



Partially decellularized larynx in prototype

- Completed modifications:
  - Program modified for greater ease of use
  - Bioreactor resealed
  - Hole drilled to prevent liquid interfacing with motor
  - Holes drilled for access

Testing: Fluid Dynamics Modeling

- Oxygen penetration calculations
  - Predict oxygenation profiles within tissue constructs
  - Goal: determine optimal rotation profile for tissue
- Solidworks/Ansys-based computational fluid dynamics modeling
  - Assess impact of fluid flow on bioreactor, tissue
  - Goal: develop basic preliminary model for predicting tissue response under bioreactor conditions

Sample fluid dynamics modeling with Solidworks

http://blog.capinc.com/wp-content/uploads/2012/02/2012-02-08-Flow-Simulation-2012-SP2-HVAC-Module.png

Testing: Decellularization

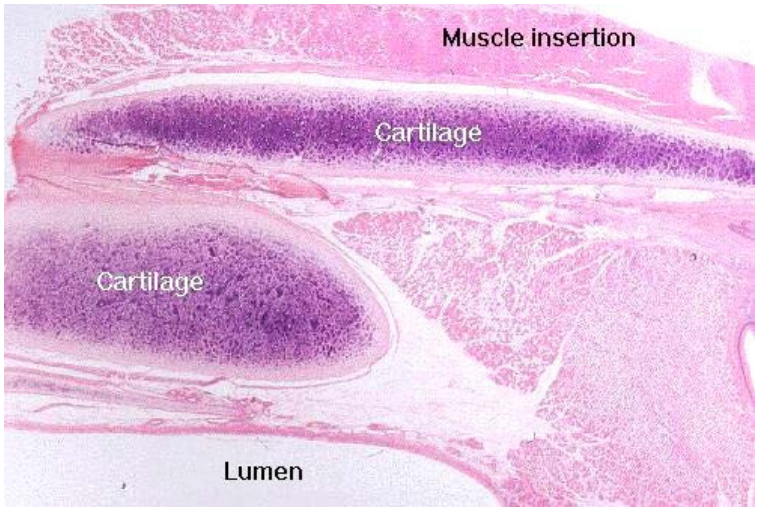
- SDS:
  - Perfuse through vasculature
  - Circulate through inner lumen
- Assays:
  - Progressive biopsy
  - Histological staining
- Variables considered:
  - Time spent in SDS
  - CHAPs as an alternative SDS
  - Detergent concentration
  - Larynx movement
  - Flow profiles in bioreactor

Fresh larynx extracted from laryngectomy

Partially decellularized larynx

Testing: Recellularization

- Fibroblasts and media perfused through decellularized scaffold
- Evaluation of cell uptake:
  - Histological examinations
  - Assay for engraftment, apoptosis, proliferation, cell survival
- Variables to consider:
  - Direct seeding vs. perfusion
  - Incorporation of multiple cell types
  - Flow profiles

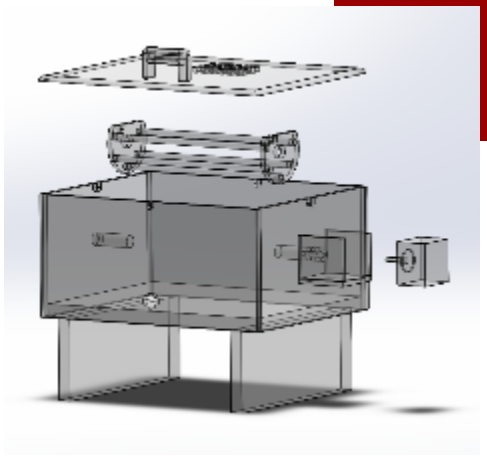


Example H&E stain for intact larynx

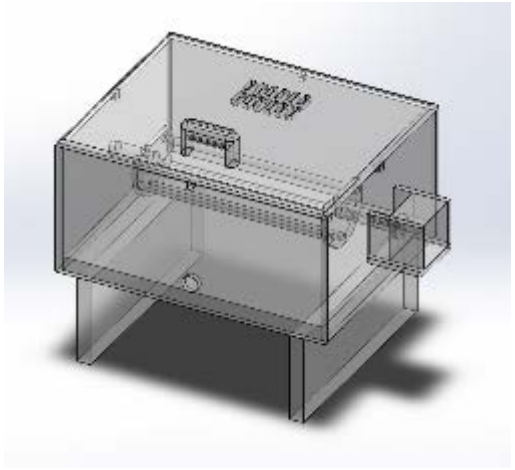
http://www.vetmed.vt.edu/education/curriculum/vm8304/lab_companion/histo-path/vm8054/labs/Lab25/EXAMPLES/EXLARYNX.HTM

Design Improvements

- Alter product dimensions
- Use of new materials
  - Sealant
- Incorporation of fabricated details for ease of use
  - Securing tissue
  - Allowing better placement of tubing
- Different incorporation of electronics
  - Change motor housing
- Use of a more accurate fabrication technique
- Alteration of drain placement
- Incorporation of a lid



Device redesign



Documentation

- User Manual
  - Manipulation of electronics
    - Altering program to suit individual need
    - Use of different programs for more complex applications
  - Product assembly, disassembly
  - Basic troubleshooting
- Service instructions
  - Product sanitation
  - List of parts for replacement, repair

Arduino Uno

http://www.liquidware.com/system/0000/3648/Arduino_Uno_Angle.jpg

Budget

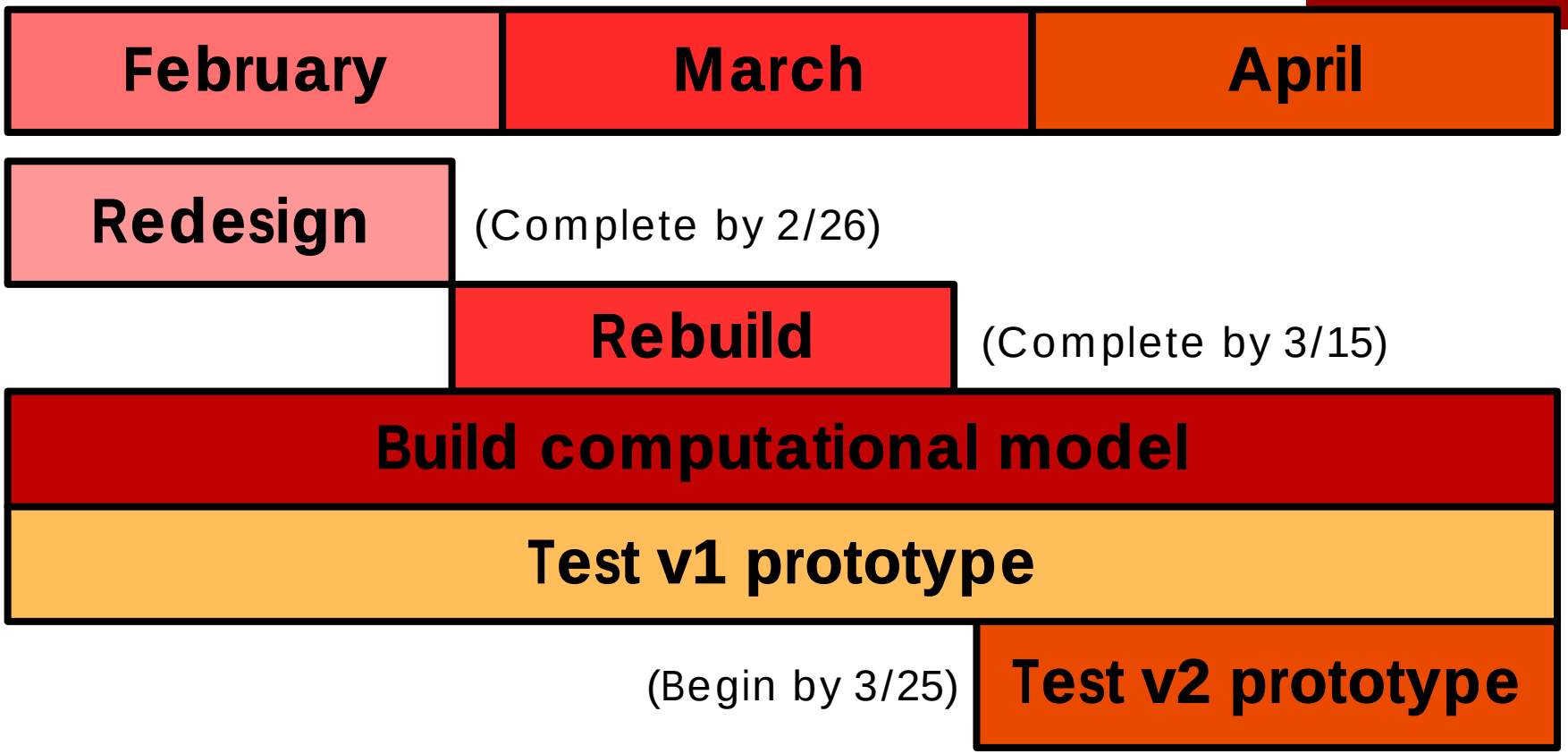
- Past Expenditures

Details	Cost
Bioreactor Raw Materials: Polycarbonate	$110
Pumps: Langer Instruments peristaltic pumps	$1,295
Electronics: Arduino Uno, Stepper Motor, Stepper Driver, etc.	$113
Miscellaneous: Hardware, seals, glue, etc.	$20
Total Cost	**$1,538**

- Future Expenditures

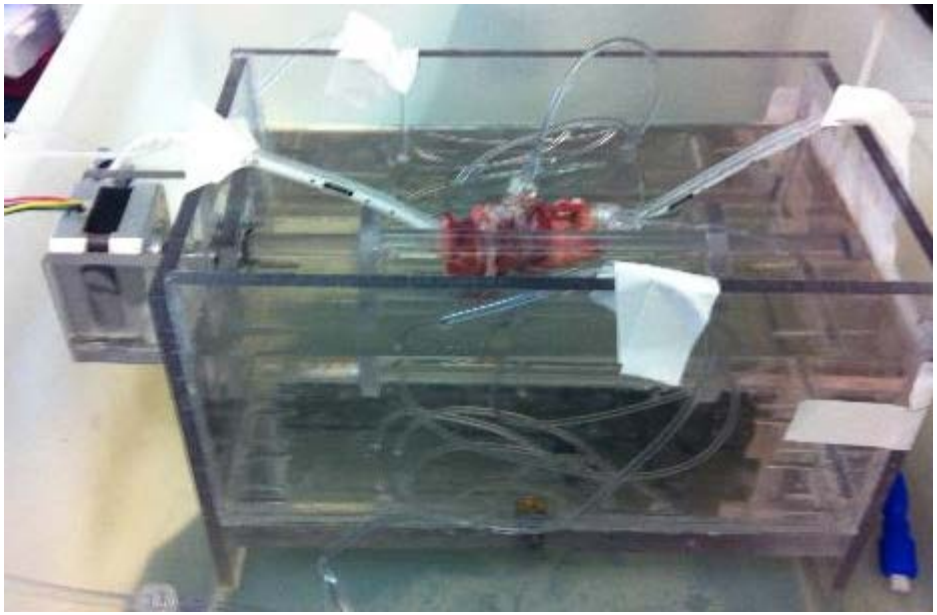
Details	Cost
Bioreactor Raw Materials: Polycarbonate	$60
Electronics: Arduino Uno, Stepper Motor, Stepper Driver, etc.	$75
Miscellaneous: Hardware, seals, glue, etc.	$40
Total Cost	**~$175**

Timeline


February
March
April
Redesign
(Complete by 2/26)
Rebuild
(Complete by 3/15)
Build computational model
Test v1 prototype
(Begin by 3/25)
Test v2 prototype

Summary

- Bioreactor to decellularize/recellularize a larynx
- Rotates to promote fluid diffusion
- Fluid access to exterior, vasculature, and inner lumen
- Automation of rotation and pump flow



Acknowledgements

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Questions?