Digital and Paper Microfluidic Diagnostic Devices

Project Plan

Iowa State University
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Summary

The project aims to design two complementary systems for performing biological assays and diagnostic tests. The first system, using digital microfluidics, is reusable and allows for precise fluid control. The second system, paper microfluidics, uses inexpensive materials to provide fast results.

Introduction

Microfluidics is a field of science and engineering which aims to study and develop systems to manipulate small (µL to nL scale) volumes of fluid. The field was enabled by microfabrication techniques developed for integrated circuit fabrication. These techniques allow researchers to create micrometer-scale channels capable of manipulating fluids. Inkjet printers, most ubiquitous example of microfluidic technology, rely on small chambers of fluid and integrated heating elements to propel droplets of ink onto the paper.

A large part of the rapidly developing field of microfluidics focusses on "lab on a chip" (LOC) applications. LOC systems integrate laboratory functions, like detecting biological compounds, on a single microfluidic device. Traditional microfluidic devices are fabricated using an elastic polymer, PDMS, bonded to a glass slide. The most sophisticated devices may have embedded electrodes for applying electric fields to the fluid flows. Recently, two alternative systems, digital microfluidics and paper microfluidics, have gained attention.

Digital microfluidics involves transporting droplets of fluid in two dimensions over a grid or electrodes using only electric fields. The system works in the following way: first, a droplet is placed on a grid of electrodes, similar to a pixel grid on an LCD display. Then, a voltage is applied to an electrode adjacent to the electrode containing the droplet. A phenomenon called electrowetting causes the droplet to move towards the new electrode. The ability to move discrete droplets in two dimensions is a unique advantage of digital microfluidics. Digital microfluidic (DMF) devices offer a flexible platform for lab-on-a-chip applications.

Paper microfluidics is a method of manipulating fluid flows using the wicking property of paper. Several systems have been developed for creating fluidic channels in paper, with wax printing being the most promising option. Melting the wax through the paper forms a fluid barrier, enabling channels to be created. Paper microfluidics can be used to develop more sophisticated lateral flow assays, or test strips, which are commonly used for point of care testing.

Problem Statement

Digital

Medical diagnostic tests are currently expensive, time consuming, and prone to human error when performed by lab technicians. Such tests involve numerous pipetting and mixing steps, requiring either expensive pipetting machines or skilled human hands. Digital microfluidic devices have the potential to automate diagnostic tests. All the required fluidic operations; transport, dispensing, merging, and mixing, can be carried out on a DMF device. Unlike traditional microfluidic devices, a DMF device is flexible enough to be used for a variety of different diagnostic tests. DMF devices have the potential to benefit resource-poor hospitals where lower cost fluid handling systems would improve the speed, accuracy, and throughput of diagnostic tests.

Paper

The current market for paper-based low-cost diagnostic devices is in its very early stages. While there are several small groups leading the market, there is still much room for growth. The goal is to, from research done on current technology in the market, create a paper-based diagnostic device capable of being competitive with the current market. To achieve this, we hope to first create a device to test the pH of saliva, and then next semester take it a step further and modify the device to diagnose a condition that will be determined next semester. Amylase in saliva is a compound that we may pursue.

Concept Sketch

Digital

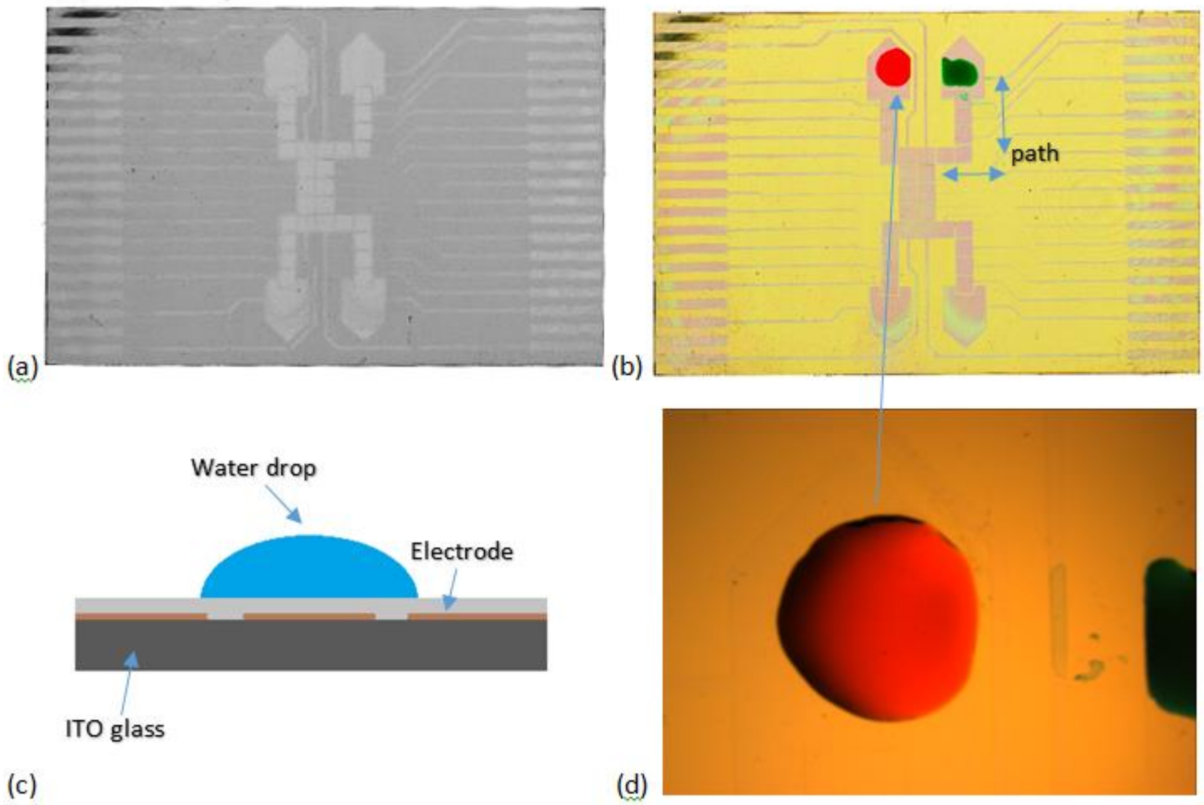


Figure 1. Concept sketch of our hydrophobic insulator: (a) ITO glass layout design, (b) Water drop moving across the path, (c) Horizontal view of hydrophobic insulator, and (d) A magnified view of water drop on the surface

Paper

Saliva collection:

We are designing a device which can collect and test the concentration of pH and viscosity in the saliva. Our saliva collection devices includes a hand hold plastic "stick" and on the other half of the stick is going to be our absorbent polymer, which will collect saliva from a collection tube where we can spit our saliva into.

Saliva testing:

On the testing part, we primarily are setting our goal to test saliva for things like alcohol level, pH, viscosity, hormone level, and our ultimate goal is to design and fabricate an at-home device that can measure and display colorimetric results for easy interpretation.

System Block Diagrams

Digital

User Interface

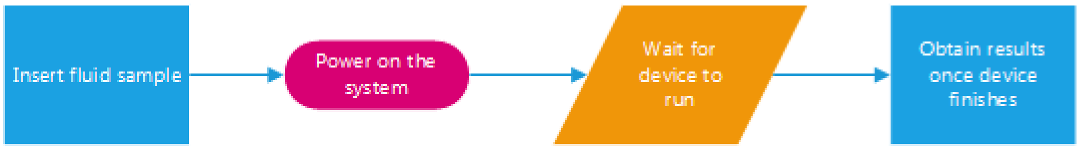


Figure 2. User Interface Block Diagram

Device Function

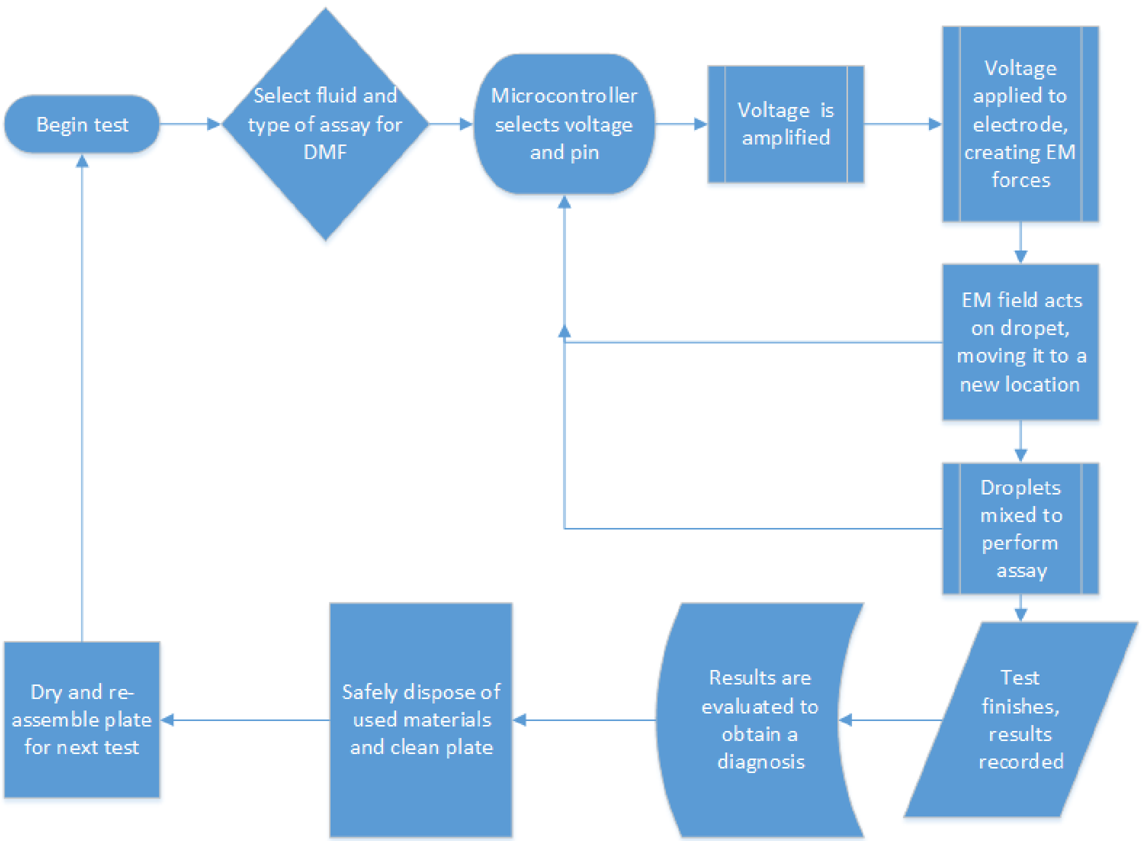


Figure 3. Device Function Block Diagram

Paper:

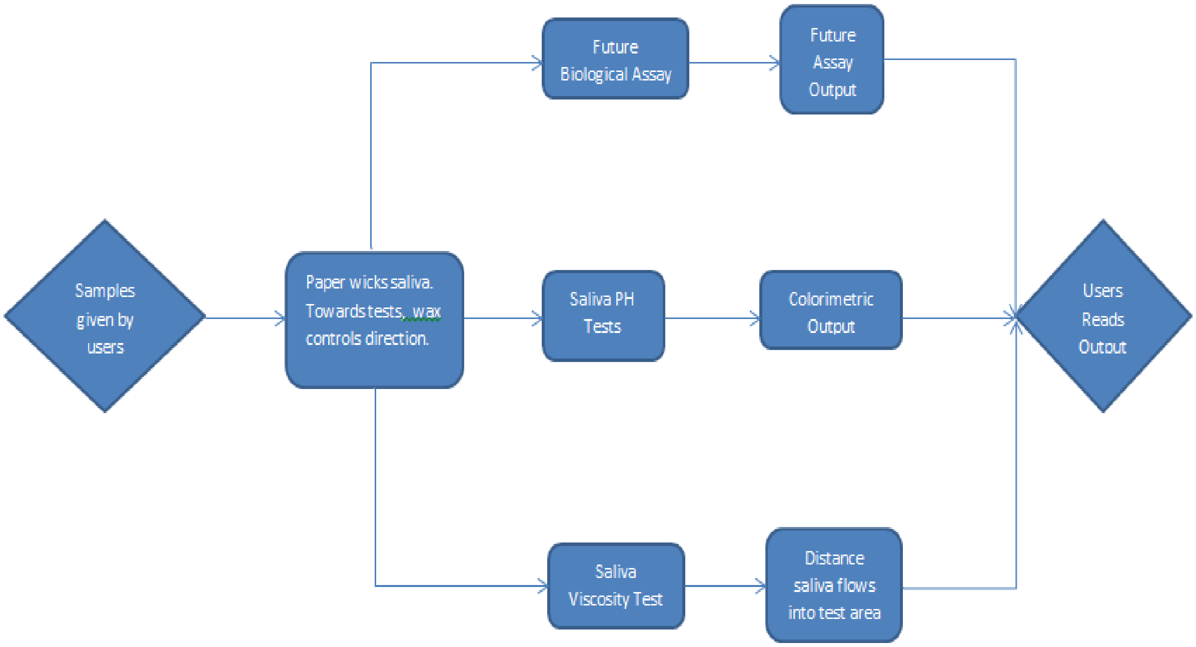


Figure 4. Paper Microfluidics Block Diagram

System Description

Digital

Hardware

DMF Control Board :

The control board receives commands from the computer, transmits a signal to the switching board and sends feedback to the computer. It relies on an Arduino Mega 2560 connected to a computer via USB. Also, the control board communicates with the driver boards and each relay switch connects to a single electrode on the DMF device via a custom pogo-pin connector. The board also measures the amount of current passing through the device according to the device impedance and amplifier output.

40 Channel High Voltage Switching Board:

The switching board receives command signals from the control board and transmits a high voltage signal to pogo pin board. It has a 40 high voltage relays that allow it to transmit the high voltage signal on 40 channels.

High Voltage Amplifier:
The amplifier converts a 0.5 Vrms input signal from the controller board to 100 Vrms output and produce 0.1~30kHz. It is connected through a feedback resistor back to the control board to facilitate amplifier-output monitoring.

120 Channel Pogo Pin Board:
The Pogo Pin Board is connected directly to an array of electrodes patterned on Indium Tin Oxide (ITO) glass. The signal is received from the high voltage switching board.

Webcam / High Speed Camera:
A webcam will record the real time events on the device's surface electrodes.

Software
A graphical user interface is used for the DMF automation system. Software can communicate with the control board through the Arduino. Also, a high speed camera records and displays the real time result on the device surface. The software can record sequences of electrode activation steps needed to manipulate the discrete droplets to perform an assay.

Paper
The sample is deposited on the paper substrate by the patient. The saliva flows through the paper through the designed pattern. The wax barriers of the design keep the sample within the boundaries of the pattern. The saliva flows to the designated test areas, which could include, but is not limited to: viscosity, pH, alcohol, or other analyte. The viscosity test will give results based on distance that the saliva flows into the test area. The pH, alcohol, or other analyte tests will provide colorimetric results for visual observation.

Operating Environment

Digital
The proper operating environment will be an indoor laboratory. The system for the digital microfluidic (DMF) device will be operated by a computer program. A power source is needed for both the DMF device and the computer. The device manipulates liquid droplet to such as blood, saliva, or sweat. These liquid droplets are sensitive to temperature changes, so a constant room temperature is required to use the device. The device should be operated on a flat, level surface to prevent the droplets from being influenced by gravity. Vibrations should be minimized to ensure consistent droplet motion Indium Tin Oxide (ITO) glass is fragile, so the operating area

needs to prevent outer shocks from damaging the ITO glass. Overall, the indoor laboratory which can supply the electricity is the optimal place to operate the device.

Paper

The operating environment will be at a user's home. The design includes two parts, one part is the saliva collector, the other part will be the polymer indicator, which is supposed to show different colors and indicate different health condition for users. The requirement of the operating environment should be relatively low, under cold or hot weather, different humidity, different strength of light the device should eventually get the same result. The devices should be easy to use and the result should be easy to read and understand.

User Interface Description

Digital

The user will interact with software and hardware interfaces of the device. The software interface will allow users to graphically control which electrodes of the device are actuated. The hardware interface involves two components; loading the sample onto the electrodes, and connecting the electronics.

Loading the device involves pipetting the sample and the required reagents onto the reservoir electrodes, connecting the electrode array to the high voltage switching board using an edge connector, and covering the bottom electrode array with a cover glass (sandwiching the droplets in between). The electronics require a standard 120Vac power supply from a wall socket. The device connects to a computer via USB. Optionally, a webcam can be used to display the electrodes on ITO glass and confirm that the droplet moved to the correct location after clicking the mouse on the screen to activate the electrode.

Paper

The devices should be portable, handheld, light small, easy and safe to test, and the test result should also be fast, safe and easy to indicate the health condition for the user. The difference of color for different results should be discernible by eyes directly.

Functional Requirements

Digital

The main goal of our project is to make a device that enables manipulation of the droplets on an array of electrodes. Also, the device should be portable and easy to use. To operate the device, we must have a power source and electricity. The power source will be used to generate the signal and provide a voltage to each electrode. The lists below are the requirements for the DMF device.

- Indium Tin Oxide (ITO) glass
    - All the wires and electrodes are connected.
    - The glass is coated with ~50nm of Teflon-AF 1600 by spin-coating (1000 rpm, 30s)

- Signal
    - DMF device uses an AC signal.
    - The signal is amplified (around 100~150V) by an amplifier.
    - High voltage driver boards give the system around 100 channels.

- Software program
    - Each electrode is matched with the electrode on the screen.
    - Appropriate voltage should be applied to the DMF device (100~150V)

Paper

The functional requirements of our project are that it can run several different tests and display the results calorimetrically for easy interpretation: pH, viscosity and hydration of saliva. Our saliva testing device is at-home test kit, and it can only be used one time. There is no power source or electricity needed for our saliva testing device. Our saliva testing device can only handle with small amount of fluid, usually less than 5 ml. With our testing device, you will be able to test at least one analyte to determine people's health condition.

Non-Functional Requirements

Digital

In order to make the digital device successfully work as a low cost medical device it will need to be portable, easy to setup, safe, and modular. Over time pieces of the system will experience wear and tear, so parts of it such as the ITO circuit board will need to be replaced without having to purchase an entirely new system. The device uses high voltages (over 50V) which require safety features to prevent users from hurting themselves. The system design is complex and uses multiple components including high voltage amplifiers, microcontrollers, sensors, electromagnetic fields and more that will have a level of complexity that a user will not be expected to fully understand, but must be able to utilize simply and effectively. Finally a degree of portability will be needed; currently an external power source is required to operate the machine. Users should be able to assemble it in various locations while minimizing required space.

Paper

Our non-functional requirement of this project is that the saliva testing device needs to be relatively cheap. This is one of our goals of the project to replace the current method which is higher cost with this cheaper alternative. Another requirement for this project is that this device needs to be very easy to use. Users who don't have professional background should be able to do the tests at home by following the instructions. Lastly, the entire testing process should be relatively fast, taking less than 30 minutes.

Market and literature survey

Paper

The leader in paper microfluidics is the research group of Dr. George Whitesides. In the last five to ten years, Whitesides has published several papers regarding low-cost diagnostics with a number of them being directly related to paper microfluidics. Our group has studied some of the papers published by the Whitesides research group and is using the information we have gleaned from them to begin our own work. We have also looked into many other devices currently on the market that are used to test saliva for a variety of characteristics such as pH, viscosity, alcohol content, and hormone levels.

Some of the research companies and their products are listed below:

1. http://www.healthchemdiagnostics.com/

Saliva Collection Device

Figure 5. Saliva Collection Device

The polymer used in this device can concentrate the protein in saliva.
Market: available in Italy, Span, Not in the US.
Price: $9 each for orders over $900

2. http://www.chematics.com/alcoscreen.php

Alco-Screen 2 Minute Saliva Alcohol Test

Figure 6. Alco-Screen 2 Minute Saliva Alcohol Test

The test indicates blood alcohol concentration.

Market: available worldwide.
Price: $9.99/3pack $23.99/10pack.

3. http://www.americanscreeningcorp.com/Drug-Testing-Saliva-C21.aspx

Discover 5 Panel OneStep Oral Fluid Test

Figure 7. Discover 5 Panel OneStep Oral Fluid Test

The saliva test indicates the presence of five different drugs.
Market: available worldwide.
Price: $150 per device

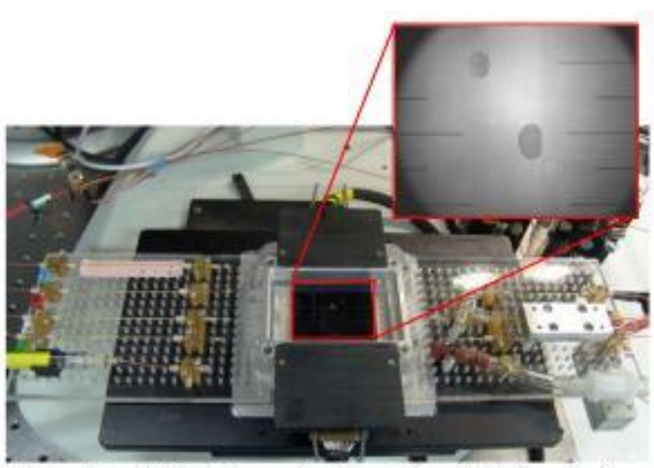
Digital

There are a few big names in the Digital Microfluidics market, including Advanced Liquid Logic, Sandia National Laboratories, and Dr. Aaron Wheeler.

Advanced Liquid Logic is the only company in the market that makes digital microfluidic devices and has a product that is called the LSD-100 Lysosomal Storage Enzyme Analysis system which uses similar eletrowetting principles to perform assays on dried blood spot extracts. The system exists for running many tests at once in a lab but is not marketed as a point of care medical diagnostic that could be used in homes.

Figure 8. LSD-100 Analyzer

Sandia National Laboratories takes the digital microfluidic devices in the direction of DNA sequencing. The goal of their digital microfluidic system is to produce devices around $3000, which is well suited for doing lab work and not for personal diagnosis or low cost. Therefore while they utilize the theory and fundamentals of digital microfluidics, the application of the device has a different purpose than what we propose.



Integrated AMB platform for formatting DNA for Illumina sequencers.

Figure 9.

Much of the literature published on digital microfluidics comes from Dr. Aaron Wheeler's group. Currently the research is broken down into 8 different subcategories from clinical applications, extraction techniques, digital chemical reactions, and more. Approximately 50 publications on digital microfluidics is the primary source that we refer to for information on digital microfluidics. For the first stage of the project we are modeling our prototype based on an open source system listed below.

Fobel, R.; Fobel, C.; Wheeler, A.R. "Dropbot: An Open-Source Digital Microfluidic Control System with Precise Control of Electrostatic Driving Force and Instantaneous Drop Velocity Measurement" Appl. Phys. Lett. 2013, 102, 193513.

Deliverables

Digital

We will deliver a DMF device that can manipulate discrete fluidic droplets on the surface of an array of electrodes coated with a hydrophobic insulator by May 2014.

For fall semester of 2013, we will deliver circuit board schematics and electrode layouts for a DMF device. We will also deliver a prototype device based on existing DMF designs.

Next semester will focus on creating a robust and reliable DMF device. The final deliverables include the hardware (the device itself) and software needed to run the device. A biological assay should be demonstrated on the device.

Paper

For the fall semester, we will deliver a saliva testing unit using paper as a substrate. The patient will deliver a sample of saliva and the device will measure the pH of the sample. For the spring semester, we will deliver a device that incorporates more biological testing to the sample provided. Pre-concentration of the sample, vertical and horizontal flow patterns, colorimetric results, and biological assays are all things that could be included in the final black box design.

Work Plan

Timeline

Digital

October

- Fabricate ITO glass electrode arrays
- Sent ITO glass to Specialty Coating Systems for Parylene coating
- Verify functionality of control boards

November

- Finish fully functional prototype capable of moving a droplet
- Design enclosure for high voltage amplifier
- Redesign circuit boards

December

- Order parts for new circuit boards
- Write code to control the circuit boards through a computer
- Design enclosure for final system

Spring Semester

	Jan			Feb				Mar						Apr				May
Week of:	13	20	27	3	10	17	24	3	10	17	24	29	7	14	21	28	5
Task																	
Fabricate new circuit boards	■	■															
Design new electrode array	■	■															
Test board functionality		■	■	■													
Fabricate ITO electrode arrays		■	■	■	■												
Research biological tests			■	■	■	■											
Build enclosures					■	■	■										
Battery powered device							■	■									
Characterize system							■	■	■	■							
Implement biological test								■	■	■							
Prepare Final Documents										■	■	■	■	■			
Prepare Final Presentation															■	■	■

Paper

October:

- Physical system design
- Order products - pH indicator, polymer to act as saliva substitute.
- Fabrication of the system design

November:

- Continued fabrication and modifications to system design if necessary
- Demonstrate pH test
- Depending on time and resources, integrate pH test and viscosity test into one single device

December:

- Final testing of device to show capability of measuring viscosity and pH of a saliva sample or substitute test medium.

Spring Semester

	Jan			Feb				Mar					Apr				May
Week of:	13	20	27	3	10	17	24	3	10	17	24	29	7	14	21	28	5
Task																	
Research biological tests	■	■	■	■	■												
Build multi-analyte capable device			■	■	■	■											
Integrate non-paper substrates				■	■	■	■										
Implement analyte concentration						■	■	■	■								
Implement function test								■	■	■							
Prepare Final Documents										■	■	■	■	■			
Prepare Final Presentation															■	■	■

Member contributions

Digital

Jared Anderson

- Market research and literature survey
- ITO glass fabrication

Riley Brien

- Group leader
- Design enclosures for high voltage amplifier and final system
- Research biological tests

Taejoon Kong

- Control board circuit design
- Research biological tests

Chee Kang Tan

- Program circuit boards
- Develop user interface

Paper

Nick Dudak

- Research biological tests
- Integrate non-paper substrates to device design (for concentration of sample)
- Creation of pH solutions

Ben Hagarty

- Subgroup leader
- Fabrication of wax barriers on paper
- Creation and testing of pH solutions

Jingxuan Sun

- Research Patents
- Literature review
- Design of paper structure

Qi Sun

- Fabrication of paper and wax barriers
- Design multi-analyte device