A NOVEL BIOREACTOR FOR MUSCLE TISSUE ENGINEERING

THE IMPACT OF SOCIAL MORES ON VACCINE DEVELOPMENT AND DISTRIBUTION DURING PUBLIC HEALTH CRISES

A Thesis Prospectus
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By
Caroline Roden

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Technical Team Members: Benedict Albergo, Curtis Creech, Aparna Kola

On my honor as a University student, I have neither given nor received unauthorized aid on this assignment as defined by the Honor Guidelines for Thesis-Related Assignments.

ADVISORS

Catherine Baritaud, Department of Engineering and Society

Steven Caliari, Departments of Chemical and Biomedical Engineering

Volumetric muscle loss (VML) is a broad term used to describe permanent, large-scale damage to muscle tissue that results in some form of decreased function (Corona et al., 2012; Machingal et al., 2011). It is extremely common in military settings, but also affects many civilian populations (Corona et al., 2015; Grogan et al., 2011). Traditional approaches to treat VML have had limited effectiveness, as muscle grafts require large volumes of tissue which can lead to donor site morbidity (Corona et al., 2015; Sarrafian et al., 2018). Additionally, most approaches do not account for the frequent comorbidity of VML with damage to nerve and connective tissues (Gilbert-Honick & Grayson, 2020; Yang & Temenoff, 2009). As such, functional repair of VML is difficult to achieve with traditional methods.

Biomaterials-based approaches have been attempted in recent years, but most are only effective at treating simple injuries (Gilbert-Honick & Grayson, 2020; Owens et al., 2008). Muscle function is highly dependent on innervation and a strong connection at the musculotendinous junction (MTJ). As such, this project's technical advisor, Steven Caliari of the Chemical and Biomedical Engineering departments, has worked with his lab to create a new scaffold for regenerating skeletal muscle to address these needs (Basurto et al., 2021). The goal of this scaffold is to encourage myogenic cells to mature and proliferate according to spatial and environmental cues to regenerate VML/MTJ injuries. Past studies have indicated the beneficial effects of mechanical or electrical stimulation on tissue development. In this research project, the existing scaffold will be tested in a custom designed bioreactor while undergoing combined electromechanical stimulation (Goldberg, 1967; Goldspink, 1999; Maleiner et al., 2018). This technical project will take place over the whole school year, with design and prototyping beginning in November and testing taking place during the Spring semester. By the end of the project, a functional bioreactor will be fabricated and tested for future use by the Caliari lab. This

device will first be virtually designed and simulated, before being fabricated using university equipment and grant funding given to the advisor's research group.

The STS research project will examine the link between evolving societal mores and the development and acceptance of vaccine technologies during public health crises. As the Covid-19 pandemic continues to spread, widespread hesitance to accept newer vaccine technology has hindered efforts to reach herd immunity. Since the invention of the first true vaccine in response to Smallpox, scientists have had access to an ever-expanding toolbox of technologies to inoculate populations against diseases. However, the implementation of these tools and the overall acceptance of the final products has been inconsistent. Public and governmental opinions regarding what constitutes a true crisis often dictate what projects get adequate attention and funding, which hinders scientists' abilities to develop necessary vaccine technologies. While most vaccinations that are developed are consistently effective and safe for public use, society's acceptance of the technologies fluctuate frequently. As is often the case, history may provide a window into the causes of these patterns. By examining the social contexts surrounding a variety of public health crises with both good and bad outcomes, this project aims to determine how social mores influence the development and diffusion of vaccination technologies. These historical case studies will then be contextualized within the Handoff model and the Technology and Social Relationships (TSR) model. More specifically, the end user in the Handoff model will be considered as the central figure in the TSR model to show how the actors involved in getting the vaccine from the bench to the clinic can reach patients effectively, and how those patients then influence their own social spheres with their acceptance of that same technology. While this subject is not tightly coupled with the technical topic, further researching regarding views on

bodily autonomy and the ethics of growing human tissues *ex vivo* may reveal how widely the products developed out of the technical project will be accepted as well.

DESIGNING A NOVEL BIOREACTOR FOR ELECTROMECHANICAL STIMULATION OF TISSUE-ENGINEERED SCAFFOLDS

Muscle tissues are highly complex and difficult to recreate with adequate functionality. When a large amount of muscle is lost, the body is unable to heal itself and patients are often left with poor functionality and potential disability. Central to the proper function of muscle tissue is tissue contractility prompted by electrical stimulation from the nervous system. When the structures necessary to facilitate this coordinated action are damaged extensively, as they frequently are in volumetric muscle loss (VML), the body lacks the ability to repair the region. Many tissue-engineered muscle repair constructs have been designed over the years with varying protocols for tissue preconditioning via bioreactors. Most studies either conditioned the tissues with mechanical or electrical stimulation. This technical project, however, aims to design a bioreactor capable of providing coordinated electrical and mechanical stimuli to an anisotropically-aligned and electrically conductive scaffold.

Open-source bioreactor designs will be used as a starting point for this technical project. Following an extensive literature review of the current state-of-the-art, initial designing of the bioreactor will be done using 3D modeling software. After running simulations and optimizing the design, the initial prototype will be constructed and assembled. Scaffolds provided by the associated lab will be used to test the initial prototype, and longer-term tests will be carried out to ensure durability of the machine.

As depicted in Figure 1 on page 4, the bioreactor will first consist of a basin for holding cell culture media, in which the scaffold will reside for the duration of its preconditioning. The scaffold will be held on both ends by clamps. One end will be rigid, and the other will be attached to a servo motor, which will be responsible for providing the cyclic strain to the scaffold. Electrical stimulation will pass through the scaffold via either electrodes on the scaffold, or via the clamps themselves. The cell culture basin will have electrical directionality built in by giving each end polarity. The servo motor and the electrical stimulation will be controlled by microprocessors that will be programmed by members of the team.

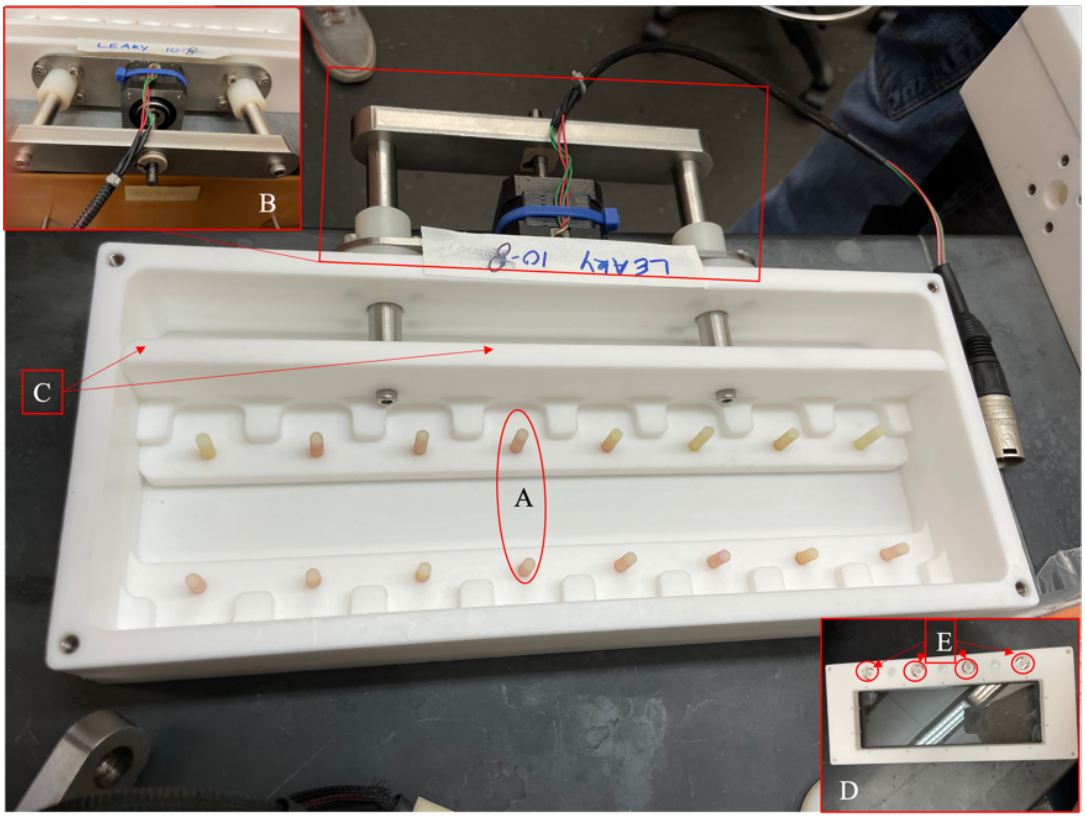

Figure 1: Christ Lab skeletal muscle tissue bioreactor with mechanical stimulation. A. Muscle tissue construct is clamped between two screws. B. Mechanical stimulation motor control. C. Wall attached to clamps moves to stretch tissue. D. Lid allows for monitoring tissue culture. E. Filters for proper exchange of gasses. (Roden, 2021)

This technical project is being completed under the advisement of Steven Caliari of the Chemical and Biomedical Engineering departments at the University of Virginia, as well as graduate student of Chemical Engineering, Ivan Basurto. Dr. George Christ's lab has also offered insight and resources to aid in this project's completion. The team members on this project are fellow Biomedical Engineering Majors Benedict Albergo, Curtis Creech, and Aparna Kola. The project at hand is also being completed as part of a larger, ongoing project by the Caliari and Christ labs at the University of Virginia. As such, grant money is available for use in purchasing materials needed to fabricate the bioreactor. The 3D printing labs and the design labs at Stacey Hall will be used for design and fabrication of the bioreactor as well. Arduino microprocessors will be used to program the bioreactor. Myogenic cells will be sourced from either the Caliari lab or from Fischer Scientific. The expected product of the technical project is a functional bioreactor capable of providing tunable electromechanical stimulation to a given cell-seeded scaffold in order to induce a quantifiable myogenic response in the cells. The project will be documented in a technical report.

SOCIAL MORES AND PUBLIC HEALTH CRISIS RESPONSE

The Covid-19 pandemic has highlighted many egregious gaps in the public chain of communication necessary for a proper and effective crisis response. These poor communication skills, however, are not the sole arbiter of the public's response to new vaccine technologies. Society is more connected now by rapid communication via the internet than it was during any previous pandemic. This "word-of-mouth" communication should have provided a boost to scientists attempting to convince laypeople to trust the vaccine. Many pockets of society, however, have continued to resist it and view anyone pushing them to get vaccinated in a deeply

negative light. Why is it that getting vaccinated used to be the "patriotic" thing to do, where now people view it as a symbol of an overbearing government? In this research project, generational social mores will be studied in correlation with public attitudes towards newer vaccine technologies. Additionally, the impact of these communal ideals on deciding which technologies deserve investment will be studied.

CHANGING SOCIAL MORES

A community's mores are "centrally important and accepted folkways, and cultural norms that embody the fundamental moral views of a group," (Farlex Partner Medical Dictionary, 2012). Since the invention of the first vaccine by Edward Jenner in 1796, it is fair to say that society and what society accepts as 'normal' has changed drastically (The College of Physicians of Philadelphia, n.d.). The individual's view of their own place in a given society has also changed. Political polarizations have made many people view calls to action for the 'betterment of society' as affronts to their personal liberties, where they might have previously seen those same actions as 'patriotic'. A shifting American attitude from commonality towards individualism may be partially to blame for some changing attitudes toward public health measures intended to better society. In searching for correlations between social mores and community attitudes regarding selflessness, personal liberties, and what every person owes to society, as well as the public's acceptance of and response to novel vaccine technologies, this project hopes to illuminate ways in which engineers can more effectively market these technologies in the modern era to improve outcomes during public health crises.

The barriers to an effective study of social mores are abundant. It can be difficult to define the morals of a society, but this study will attempt to do so by examining one country, the

United States, and will attempt to discover public views on society through legislation of the time, religious views, and by viewing era-appropriate propaganda. Polling data will be utilized when possible. Additionally, tracing the flow of money in vaccine research funding may illuminate overall public opinions, as many companies invest in what is viewed as being publicly favorable.

This research project will be completed by investigating various public health crises throughout the past century. Particular interest will be paid to those where social factors seemed to influence the progression of treatments and solutions heavily, such as Polio, HIV, and Covid-19. The Social construction of technology (SCOT) method will be utilized to examine the implementation and acceptance of varying public-health related technologies (Kline & Pinch, 1999). The 'technology and social relationships' model of researching be utilized to illuminate the decision-making process of specific social groups as they do or do not accept a vaccine, as shown in Figure 2 (Baritaud & Carlson, 2009). Additionally, the 'handoff model' shown in Figure 3 on page 8 will be utilized when discussing which vaccines actually make it into the market, as there are many social and political factors which can influence the marketability of certain drugs, as seen during the HIV crisis (Baritaud & Carlson, 2009). These

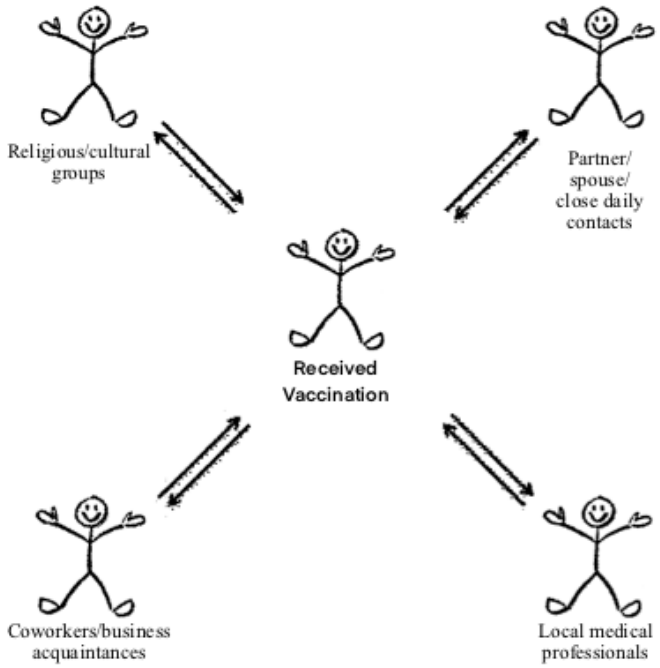

Figure 2: The Technology and Social Relationships model will illuminate how certain groups respond to vaccine technologies. (Adapted by Roden (2021) and Baritaud (2009) from Carlson).

two models will be considered in connection to each other, where the vaccinated end user of the handoff model will be considered as the central figure in the TSR model.

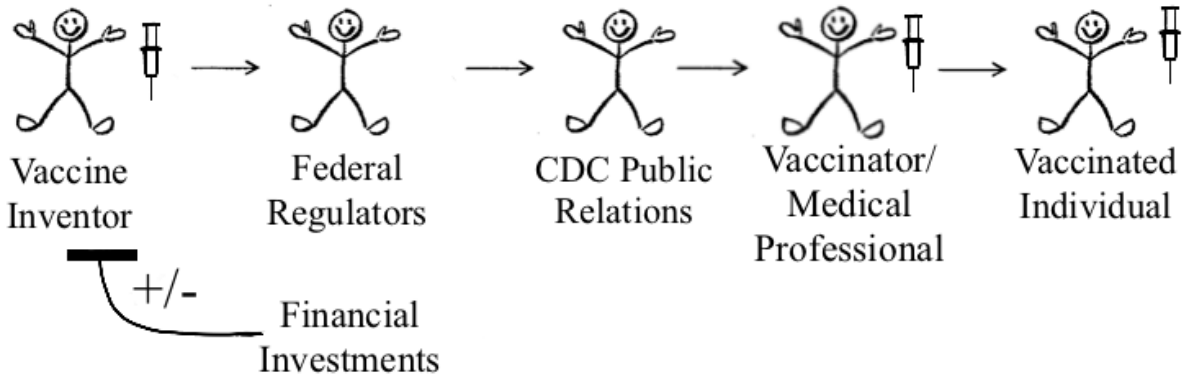

Figure 3: Handoff model utilized to show commercialization of vaccine technology. (Adapted by Roden (2021) and Baritaud (2009), Carlson).

This paper will be in the form of a scholarly article. Through a deep dive into historical responses to public health crises, this project will hopefully provide evidence that indicates that community efforts and attitudes towards embracing and understanding technologies are crucial to the adoption of these technologies on a large scale. Hopefully, this can show scientists how best to market modern vaccine technologies.

LOOKING FORWARD

The fabrication of a multi-chamber bioreactor capable of electromechanical stimulation preferential to MDC myogenesis will allow for the construction of a 3D multi-compartment scaffold capable of mimicking native skeletal muscle. Establishing a computational simulation protocol capable of controlling and predicting the behavior of the bioreactor system may also provide an effective model for other electromechanically responsive tissue (e.g., cardiac muscle). In the future, bioreactor preconditioning of myogenic cells within this tissue construct may guide

the repair of clinically relevant VML/MTJ injuries. Overall, this novel biomaterial approach will be significant to restoring muscle function and improving the lifestyle of thousands of civilians and military personnel suffering from VML.

Using a historical mindset towards vaccine acceptance and development, this project will hopefully illuminate a path forward for scientists and society. While the Covid-19 pandemic continues, it may be useful to scientists to look at society closely to determine how they market the vaccine. Future pandemics may have the same issues, so looking at society may be the key to determining the best way to respond to future pandemics.

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