An Updated Device for Time Lapse Microscopy to Study *Toxoplasma Gondii* Invasion in Intestinal Epithelial Cells

(Technical Paper)

A Framework for Improving Access to Nutritional and Mental Healthcare to Decrease the Prevalence of Preventable Diseases in the United States

(STS Paper)

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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.

Introduction

Chronic diseases are a leading cause of death in the United States, despite the fact that the prevalence of chronic diseases, such as coronary heart disease and type 2 diabetes mellitus, could be decreased by as much as 90% if preventative measures were taken to eliminate traditional risk factors (Greenberg & Pi-Sunyer, 2019; Hu et al., 2001; Stampfer et al., 2000). The United States Healthcare system is reactive, focused on fixing health issues after they occur, instead of preventing health issues from occurring in the first place. Therefore, the proposed research will explore methods for treating the underlying problems behind diseases, in order to minimize the need for further, reactive treatment. For the technical topic, a mechanobiology approach will be used to treat the peristaltic abnormalities that increase risk of toxoplasmosis. Meanwhile, the proposed research paper will concern improved nutritional and mental healthcare and their connection as a means for preventing a plethora of diseases before they set in.

The technical topic will study the relationship between the stretching capabilities of the intestinal epithelial cells, which line the gut wall, and their impact on the invasion of a parasite known as *toxoplasma gondii*. Most people infected with *T. gondii* are asymptomatic, but people with inflammatory bowel disease (IBD), a condition characterized by irregular peristaltic movements, are at a higher risk for developing toxoplasmosis, an illness similar to the flu (Kato, 2018). Current treatments for toxoplasmosis focus on the therapeutic remedies for curing toxoplasmosis once it has set in, but this technical report will focus on the mechanobiology of the intestinal wall with the goal of developing toxoplasmosis treatments that address the underlying causes instead of treating only the toxoplasmosis.

Similarly, the STS research question that will be investigated is how access to nutritional education and ability to act on nutritional information can be improved to decrease the

prevalence of mental health problems, along with improved access to mental healthcare, since both nutritional education and mental healthcare have potential as preventative healthcare methods to decrease the prevalence of many preventable diseases, therefore decreasing the overall economic and social costs of these diseases within the United States Healthcare system.

Technical Topic

It is estimated that roughly 30% of the world's population is infected with *T. gondii*, yet the majority of people are asymptomatic (Kato, 2018). However, *T. gondii* infection still poses a high risk to immunocompromised people, people with IBD, and pregnant women (Halliez & Buret, 2015). IBD is an umbrella term used to describe Crohn's disease and ulcerative colitis, both of which can cause extreme discomfort and debilitation. IBD affects roughly 6.8 million people worldwide, with the United States having one of the highest prevalence rates in the world (Alatab et al., 2020; Kaplan, 2015). This disease impacts the muscular contractions of the intestine, commonly referred to as peristalsis, which are hypothesized to play a role in *T. gondii* invasion (Alatab et al., 2020; Fakhoury et al., 2014).

The current standard of care for toxoplasmosis patients includes antimalarial drugs and antibiotics, but these prescriptions have been shown to only be effective in 60-70% of patients. Thus, while treatments exist for this potentially life-threatening illness, more research must be done to develop more effective treatment options. Along with this motivation for improving care for patients, the relationship between *T. gondii* invasion and the health of epithelial cells, such as those that line the intestine, must be better understood. While it is known that diseased cells have a distinct cobblestone-like appearance that is unlike the normal columnal phenotype of gut epithelial cells, it is unknown how this cell shape contributes to the increased susceptibility of *T. gondii* invasion (Booth, 1970). A mechanobiology-focused approach is required to determine

whether *T. gondii* invasion is a consequence of the non-stretch adapted epithelial cells associated with the disease state, or whether *T. gondii* invasion produces the diseased cell state. The experiments involved in this research will involve stretching epithelial cells using a stretch device that mimics the contractile movements of the intestine and infecting the cells with *T. gondii.* In order to study these interactions in real time using live-cell microscopy, the cells must remain in an environmental chamber that mimics physiological conditions. *T. gondii* invasion rates*,* epithelial cell responses, and structural dynamics can all be analyzed with this imaging technique and the presence of such an environmental chamber.

Therefore, the primary goal of this technical project is to design an environmental chamber in which the relationship between *T. gondii* invasion and the health of epithelial cells will be observed. In February 2021, the capstone team will design a 3D printed enclosed chamber using AutoDesk Fusion that will serve as a mountable microscope incubator. Specifications such as maintaining incubator conditions and protecting the microscope will be incorporated into the design and will allow the team to conduct accurate experiments in an efficient manner and produce consistent data. Once the environmental chamber is completed and printed by early March 2021, cells will be plated, infected with *T. gondii*, and stretched in a way that mimics diseased or healthy cells. Invasion mechanisms can then be observed and effective research can be done to understand the relationship between *Toxoplasma gondii* and gut diseases like IBD. Furthermore, a better understanding of these invasion mechanisms can provide a new angle from which to approach the development of treatment options for toxoplasmosis that target the mechanobiology of the intestine and have some potential to treat underlying conditions such as IBD as well. This project will directly result in a novel environmental chamber to study this topic and a technical report will be written to present the various results of the project.

STS Topic

Nutritional psychiatry is a relatively new field that studies the link between diet and mental health (Adan et al., 2019; Sarris, 2019). Researchers (Conner et al., 2017; Dinan et al., 2019; Moreno-Agostino et al., 2019) have found evidence of a link between poor diet and mood disorders such as depression and anxiety (Adan et al., 2019), suggesting that a healthy diet can decrease one's risk of developing a mood disorder. While there are many contributing factors for poor mental health, this research paper will focus on the nutritional component. Furthermore, diet is linked to physical health conditions such as cancer (Di Daniele et al., 2016). Therefore, the direct effects of nutrition on physical health must be taken into consideration, in addition to the indirect effects of nutrition on physical health via mental health. One link between nutrition and overall health that will be discussed is the impact of the gut microbiome, which has been identified as a potential therapeutic target for both mental and physical health problems, as it plays a role in gastrointestinal disorders, metabolic disease, and other conditions (Hills et al., 2019; Malan-Muller et al., 2018). The final connection that must be considered is the relationship between mental health and physical health. Mental health is known to impact physical health, and it is worth noting that physical health has additionally been shown to impact mental health, with many direct and indirect connections between the two (Ohrnberger et al., 2017).

This research paper will explore methods of improving access to nutritional healthcare in order to improve mental healthcare, and therefore improve physical healthcare. There is a need for this area of research since food insecurity is a major public health problem in the United States, affecting approximately 12% of the population, and information around what makes a diet nutritious is lacking for the general public (Brown et al., 2019; Velardo, 2015). This research

paper will compare the potential efficacy of various methods of improving nutritional education and access, since different programs have yielded different results in terms of nutritional behavioral changes (Murimi et al., 2017). Furthermore, there are many barriers to mental health access, especially among low-income communities (Hodgkinson et al., 2017). Beyond economic barriers to access, there is also a lot of stigma around seeking mental healthcare, and many methods exist that seek to improve access and decrease the stigma surrounding mental healthcare (Mehta et al., 2015). For example, it has been shown that improved education about mental health and mental healthcare has the potential to decrease this stigma (Milin et al., 2016).

There are many stakeholders in terms of the intertwined connections between nutrition, mental health, and education, including all Americans who are a part of the reactive American healthcare system, and who are affected by the increasing economic and social burden on the US healthcare system (Patlak & Levit, 2009). Young Americans in particular are stakeholders in this topic since prevention of physical health problems is more effective when prevention begins at a younger age when health problems have had less of an opportunity to develop, so the ensuing research will primarily focus on how to improve education and access to nutritional healthcare and mental healthcare for children and young adults. Physicians also have a stake in the development of programs to prevent disease, since the burden that is placed on healthcare system workers can be decreased if the prevalence of preventable diseases is decreased.

This analysis will use actor-network theory to evaluate how best to improve access to and education about nutrition and mental healthcare (Cressman, 2009). Actor-network theory is a framework that describes the constantly changing relationships between actors involved in a system (Rodger et al., 2009). However, actor-network theory has been criticized for being subjective in which actors are included, so care must be taken to ensure that the network contains

all of the most relevant actors but does not become infinite (Radder, 1992). Improving the healthcare system is an incredibly intricate problem, due to the involvement of politics and policies, the economy, cultural and religious beliefs, complexities of the education system in the United States, and more. Actor-network theory will allow for the relationships between the various actors of this problem to be analyzed.

Research Question and Methods

The research question is how can access to nutritional education and ability to act on nutritional information can be improved to decrease the prevalence of mental and physical health problems, along with improved access to mental healthcare? The research methods that will be used are policy analysis, network analysis, and documentary research methods. Policy analysis will be used to evaluate the various policies currently in place to address portions of the research question and their efficacy, and network analysis will be employed to evaluate how new policies could be implemented within existing networks. The documentary research methods will include keyword searches for phrases such as "nutrition security," "mental health access," and "brain-gut-microbiome axis." The policies and other information found in this research will be organized thematically to give a clear picture of each layer of the multilayered relationship between nutrition, mental health, and physical health. This research will be conducted over the course of the Spring 2021 semester.

Conclusion

The anticipated deliverable of the technical project will be an environmental chamber to accurately study the invasion mechanisms of toxoplasma gondii in intestinal epithelial cells, and a technical report discussing further findings related to the technical topic. Meanwhile, the deliverable of the STS research project will be a proposed framework and next steps for

improving access to nutritional and mental healthcare as forms of preventative healthcare. Both of these projects will provide a new avenue for improving part of the healthcare system by addressing the underlying causes of health problems instead of primarily seeking to fix them after they develop.

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