The goal of this project is to explore the mechanism of cannabidiol (CBD) as a therapeutic for epilepsy. Cannabidiol, the non-psychoactive component of cannabis, has been shown to reduce the intensity and frequency of seizures in both animal models and humans. Nonetheless, it remains uncertain how CBD produces these effects. It is believed that CBD does so by taking advantage of the LPI-GPR55 signaling pathway. Lysophosphatidylinositol (LPI) is a lipid produced endogenously by the brain that serves as an agonist of the receptor GPR55. Many have shown that CBD functions as an antagonist of this receptor, thereby inhibiting its activity. Moreover, whereas LPI increases excitability within the brain, which can induce seizure-like activity, CBD prevents this increase in excitation. By doing so, CBD has the potential to serve as a therapeutic for epilepsy. This research aims to further elucidate cannabidiol’s role in reducing seizure severity through a GPR55-dependent mechanism.

My study will focus on cannabidiol’s action on a synaptic level within the hippocampus as I examine the type of synapses at which it is expressed. Our previous data has revealed that GPR55 colocalizes with both vesicular glutamate transporter 1 (VGLUT1), a presynaptic marker of excitatory synapses, and vesicular GABA transporter (VGAT), a presynaptic marker of inhibitory synapses. Specifically, the receptor is found more commonly in terminals labeled by VGLUT1 than VGAT. I plan to investigate how this colocalization changes when animals experience seizures as well as differences in GPR55 expression between animals who do and do not receive CBD treatment. In order to carry out this investigation, I will use in vivo seizure induction; I will do so via the seizure models of pentylenetetrazol, lithium-pilocarpine, and kainic acid. This will allow me to also investigate the effect of cannabidiol on seizure severity as part of our study. Thereafter, I will assess GPR55 colocalization with VGLUT1 and VGAT in acute hippocampal slices from these animals. By completing this project, I will be able to obtain a better understanding of cannabidiol’s activity at specific synapses throughout the hippocampus.

My role in this project during the coming semester will involve both in vivo seizure induction and immunohistochemistry. I will be spending approximately twenty-two hours in the lab per week, which will be spread over Monday afternoons and all day Wednesdays, Thursdays, and Fridays.

On Mondays, the majority of my time in the lab will be devoted to attending lab meeting. This will include both discussing the research of others in the lab as well as presenting my own work at least once this semester.
On Wednesdays, Thursdays, and Fridays, I will focus on the completion of my project itself. Towards the beginning of the semester, this will involve working with a MD/PhD student in our lab, to induce seizures in mice using the pharmacologic agents of pentylenetetrazol, lithium-pilocarpine, and kainic acid. The mice in our cohort will be sorted into three groups: vehicle non-seizure, vehicle seizure, and cannabidiol seizure. I will be in charge of carrying out the intraperitoneal injections of these drugs while the MD/PhD student remains blinded and grades the resulting seizures using the Racine scale. Thereafter, we will dissect the brains of these mice and fix them using paraformaldehyde. I will then slice the brains using a cryostat, mount them on slides, and carry out the immunohistochemistry protocol. The majority of my time following this will be dedicated to imaging the slides using the confocal microscope. Lastly, I will use a code written in the program ‘Icy’ to calculate the extent of colocalization between GPR55 and the markers VGLUT1 and VGAT among the groups of animals.

Throughout this semester, my work in this lab will grant me a better understanding of the anti-seizure mechanism of cannabidiol. I will also be able to complete any other small projects that may arise as I work to complete my senior thesis.