

[slide 1]

The Waterloo iGEM 2018 team is excited to present *E.co-light*: Dynamic Optogenetic Control of Co-cultures.

[slide 2]

In nature, microbes exist as part of complex and dynamic communities.

[slide 3]

But in the lab, it can be difficult to grow more than one species together in a co-culture. There are many reasons for this, a major one being competition. When two microorganisms are grown in the same media, they compete for resources. One often grows faster, uses up more nutrients, and takes over.

[slide 4]

In order to solve this, our team is working on a system that would allow us to control growth of bacterial populations using optogenetics. Optogenetics is the use of light to trigger genetic and/or physiological changes in a living cell. Light-sensitive systems can be found in many plants and microorganisms. These systems allow them to respond to changes in their environment. Optogenetics has many applications in synthetic biology as well, and was chosen as Method of the Year of 2010 by Nature Methods.

[slide 5]

Our project uses optogenetic control to regulate *E. coli* populations in co-cultures (cultures that contain multiple different populations)

[slide 6]

Controlling co-culture population ratios has potential applications for biosynthesis projects, chemical engineering, and many more fields which use multiple strains of bacteria to generate useful chemicals in sequence. One

bacterial strain would produce a compound, then the next strain would modify it into its final form. Working together, these microorganisms act as a factory. The better we can control the microscopic machines in this factory, the more efficient and useful it can be in producing important food products, cosmetics, fuels, or pharmaceuticals.

[slide 7]

Our goal is to control two bacterial populations independently and simultaneously! But how? We're going to control the production of MetE, an enzyme required for the synthesis of methionine. Methionine is an important amino acid bacteria need to make proteins and grow. Bacteria can get it from their environment or produce it themselves using the MetE enzyme. If placed in a medium that doesn't contain methionine, bacteria can only grow if they produce this enzyme. So by placing the MetE gene under the control of an optogenetically-regulated promoter we are able to fine-tune population growth. Our optogenetic system is CcaS CcaR, which is active under green light, and inactive under red light. So if we shine green light on our engineered bacteria, they'll express MetE, produce methionine, and grow!

[slide 8]

Alternatively, we can shut off MetE expression with red light and thereby stall growth.

[slide 9]

Let's see how this might work... Without our system: two populations are grown together, but one grows faster and takes over the culture.

[slide 10]

Let's say our system is controlling the green cells in this scenario. As soon as we notice them starting to take over the culture, we can switch their light from

green to red, slowing their growth and allowing the golden population to catch up. In order to find out the amount of light the bacteria need to grow, we can use a computer program, which runs something called Model Predictive Control. This uses our system's math model to generate an optimal strategy for shining the light.

[slide 11]

With the work done by all of our amazing teammates, we hope our project will inspire some awesome applications for future research!