First, I gathered previously collected cultures of Arthrobacter from around the world. I made an R2A media , which contains low concentration of nutrients to allow more sensitive bacteria the chance to grow. I poured the agar into a rectangular 24-well plate and allowed it to set at room temperature. Next, I made a polyethylene glycol (PEG) solution at 5 different concentrations ranging from -0.25 to -2.5 megapascals. I filter sterilized this solution and added it to the set agar for 24 hours. PEG diffused into the R2A media during this time, and excess PEG was poured off.

I then inoculated the prepared wells with Arthrobacter, using a single point inoculation at the center of each well in quadruplets. I put the inoculated plates into incubators with constant temperature of 4°, 11°, 18°, 25°, and 32° C overnight. After 24 hours, I took pictures of the growth, then again every 2 days for a week and a half. I uploaded the photos onto a computer and used an image processor to trace the bacterial growth over time for each incubation temperature. The image processor estimated bacterial growth via the colony surface area . I input this data into a software code made by Matt Gebert and modified by Nick Dragone to estimate the rate of growth for each bacteria, and compare growth rate and size across the temperature and desiccation treatments. This will help me identify temperature preferences and dryness tolerance of each bacteria of interest. Finally, I isolated the microbes using streak plating, extracted and PCR amplified the DNA for Sanger sequencing, so I could verify the identities of the isolates.

Data Set:

I used cultures that were acquired by other researchers for prior research projects. Previous researchers collected soil samples from Antarctica over the past 10 years. Others collected cultures from Panama, Tonga, and Mt Everest within the last 5 years. These isolates were stored at -80 C, until we pulled them out of storage for this project. These isolates were chosen to represent the variety of environments on Earth from which *Arthrobacter* had been isolated. The data was collected as photographs of the growth of each bacterial colony over time with a ruler for scale. I organized the data in tables matching the 24 well plates and in Excel.

Comment

Identified and cultured samples

Made R2A plates and infused with Polyethylene glycol solution

Plated samples into wells and incubated at 4°-32° C for 1.5 weeks

Inserted data into a previously-made code for growth analysis

Took pictures of plated samples every 2 days and uploaded onto computer

Outlined bacterial growth in computer app and estimated colony size via surface area

Abstract

Antarctica is 98% ice, making it a very cold, dry, inhospitable place for organisms to live. Despite this, microbes were isolated and cultured from the 1% of Antarctic land that is uncovered soil. Most Antarctic soil hasn't seen liquid water in tens of thousands of years and the soil temperatures in Antarctica only reach above freezing for less than a few hours per year. To understand why bacteria can survive in Antarctic soil despite these stressors, we can begin by comparing growth preferences and tolerances to those in other environments around the globe. To test this, I acquired samples of *Arthrobacter*, a bacterial genus found in soil across the globe, from 5 different global locations and tested the growth of each under different temperatures and water availability in the lab. By inoculating plates with water availability ranging from -0.25 to -2.5 MPA and temperatures from 4°-32° C, I measured the growth rate of each isolate. I then found the temperature preference of the isolate and its tolerance to lack of water. My research addresses how bacteria adapt to different climates and the impact of temperature preference and water tolerance on growth rate.