3: Growth Curves for Lactobacillus Plantarum HS

*Josh Derrick 2021-04*

**Overview and goals:**

The goal of this experiment is to determine the growth curve parameters for our two Lactobacillus strains: one with GFP and one with mCherry. This should give us a baseline dilution rate to calibrate a chemostat with the robot. These strains are different from the other strains we already have growth curves for because they are isolated from human saliva and not from wild fly guts

At the end of this experiment we should:

* Know the growth rate of LP\_HS\_GFP and LP\_HS\_mCherry

**Three days before Experiment(4/11):**

* Streak LP\_HS\_GFP and LP\_HS\_mCherry from Glycerol on plates of MSR+CAM. Grow up to 24 hours. Also streak LP\_WT as a control
* Prepare MSR+CAM
* Prepare PBS+CAM

**One day before the experiment (4/13):**

* Grow 3 tubes of LP\_WF\_GFP and LP\_WF\_mCherry from 3 separate colonies from plates in 3 mL MSR+CAM. Grow a tube of LP\_WT as a control.

**Day 1 (4/14):**

* Dilute LP replicates in MSR+CAM to an OD of 0.5. Read the OD using the cuvette reader in lab
* Mix 20 uL of LP with 180 uL of MSR+CAM according to the spreadsheet in this folder. This is our 1x.
* Use multichannel to transfer 20 uL to 10x dilution in PBS+CAM.
* Take out 20 uL to normalize volume of the 10x dilution.
* Grow in plate reader for 24 hr

**Day 3 (4/15):**

* Collect OD data and analyse for time to exponential threshold (dilution time) and/or growth rate, which we could use for dilution rate.