7: Invasion of L Plantarum

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**Overview and goals:**

The goal of this experiment is to determine how different invasion rates determine species composition. Hopefully we would find that the results match our voltage equation. At the end of this experiment we should know:

* If the OD of the invader causes a difference in final community composition.
* How to perform experiments like this with flow cytometer
* Feasibly of PBS as a fixative.

**Two days before Experiment(4/26):**

* 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.

**One day before the experiment (4/27):**

* Back dilute each tube 1/1000 LP\_WF\_GFP and LP\_WF\_mCherry from 3 separate colonies from plates in 3 mL MSR+CAM.

**Day 1 (4/28):**

* Back dilute each culture to an OD of 0.1. Grow up to about an OD of 0.7-1. We want many mL of indigenous culture and about 1 tube of 3 mL culture for each dilution of invader.
* Spin down invader and resuspend in PBS (2x)
* Setup plate. Vertically will be dilutions of invader, horizontally will be replicates of indigenous bacteria. 1 mL of each indigenous culture in each well of the deep well. Add 500uL of 2x MRS, 500uL of 0.25,0.5,1, 2x invader. Remove 1 mL.
* Dilute every 1.5 hrs with 500uL of 2x MRS and 500 uL invader, sample every 2uL 30 minutes onto 198 uL of PBS.

**Day 1 Onward (4/28):**

* Obtain cell counts with flow cytometer