Transformation of E. coli with G1

Participants: Brianna Branson, Kylie Hartana, Rori Hoover, Patrick Jiang, Niam LeStourgeon

Date: Friday, April 28, 2023

Protocol:

1. Thawed NEB 10-beta Competent E. coli cells on ice for 10 minutes
2. Added 25 µL E. coli cells to two microfuge tubes
3. Added 2 µL G1 to one tube and 2 µL control reaction to the other
4. Flicked the tubes to mix and briefly centrifuged the samples down
5. Incubated the tubes on ice for 30 minutes
6. Heat shocked the E. coli in a 42°C water bath for 30 seconds
7. Incubated the tubes on ice for 5 minutes
8. Added 475 µL outgrowth medium to each tube
9. Incubated the E. coli at 37°C while shaking at 400 rpm for 1 hour

Results: N/A

Conclusion: N/A

Plating G1 Transformed E. coli

Participants: Gabriella Cerna (mentor)

Date: Friday, April 28, 2023

Protocol:

1. Warmed 2 LB-agar kanamycin plates in a 37°C incubator
2. Pipetted 50 µL E. coli cells from each microfuge tube onto their respective plate
3. Placed 3 plating glass beads on each plate and tipped the plates back and forth to spread the cells
4. Incubated both plates at 37°C overnight

Results: Colonies grew on the plate with E. coli that had G1 transformed into them, and there was no growth on the control plate.

Conclusion:

Our results indicate that we were successful in generating G1. For further explanation, please refer to the conclusion section of the "G1 Generation" entry from April 26, 2023.

We want to extract this DNA from our E. coli, so our next steps are to grow the cells in a liquid culture and perform a miniprep.