7/19/17

Wednesday, July 19, 2017 9:37 AM

Who is in Lab: Salma (9-11:30, 2:30-4), Ayesha

GIBSON ASSEMBLY POSITIVE CONTROL:

Did Miniprep of YCP Overnight Culture

46 uL of concentrated DNA

- Conc. 67.4 ng/uL
- Ratio 1.91

Stored in -20^{o}C freezer in iGEM box (same place as JOE DNA is stored)

INTERLAB & GIBSON ASSEMBLY:

The cell competency test performed yesterday (7/18/17) with DH5alpha cells was not successful because the transformed bacteria was plated on Kan/CAM plated when they should have been plated on AMP plates.

Hence, new aliquots of DH5alpha cells were made chemically competent and transformed with DNA. **"Making Chemically Competent Cell"** and **"Chemical Transformation"** protocols were used to plate the samples listed below.

Duplicates were made of each of the following:

-DH5alpha with 10 pg/uL concentration of pUC19 plasmid.
-DH5alpha with 50 pg/uL concentration of pUC19 plasmid
-BL21 (DE3) with 10 pg/uL concentration of pUC19 plasmid
-BL21(DE3) with 50 pg/uL concentration of pUC19 plasmid

A total of 8 centrifuge tubes were used for this setup and 8 AMP plates (green and brown stripes).

For samples with 10pg/uL, a 6 uL stock solution was made which was diluted stock of the 50 pg/uL of the pUC19 plasmid.

Calculations:

C1V1= C2V2
(50 pg/uL) (V1)= (10pg/uL)(6uL)

V1= 1.2 uL of pUC19 plasmid must be diluted in 4.8 uL of DI H2O.

TO do tomorrow:

- Quantify # of colonies on the cell competency test
- Autoclave 1000 uL pipette tips