

6/28/17

Wednesday, June 28, 2017 12:48 PM

Who's in Lab Today: Nathan, Ayesha, Jeff, Ana

Made LB/Amp agar plates and LB/CAM/Kan agar plates to streak out strains from the stabs.

Strains are:

LC1539 - BL21(DE3)pLysS/pPhusion (grow this in LB + Kan + Cam (10 ug/ml))

LC1853 - TOP10/pTM0100 (LB + Amp)

Working conc.:

Amp: 100 ug/mL

CAM: 10 ug/mL

Kan: 50 ug/mL

We did not see any colonies in the stabs. Hence, we plated the strains using a different protocol.

The protocol used is as follows:

1. 50 uL of sterile LB media is added to the stab.
2. Invert the stab back and forth for approximately 5 minutes.
3. Take a micropipette and extract the 50 uL of LB from the stab.
 - a. **After inverting for five minutes, the idea is that the cells have dislodged and the LB media contains cell now.**
4. This 50 uL of LB media + cells can be deposited on the respective agar plate.
5. Use glass beads (approximately 10 glass beads) to spread out the media on the plate.
6. Make sure to label the plate.
7. Invert the plate and place in the 37 degree room.

Black stripes denote KAN/CAM LB Media

Green and brown stripes denote Ampicillin LB Media