**Minipreps (Alkaline Lysis)**

1) Pick a single colony from plate using a 200 µl pipet tip.

2) Place in 2 ml of LB + Amp and shake ON at 37°C. (1 ul amp for every mL of LB)

3) Transfer 1 ml of culture to an eppendorf and spin @ 12K 1 min to pellet.

4) Aspirate LB.

5) Resuspend the pellet in 125 µl Solution I by pipetting (run on rack to resuspend pellet before adding solution I)

6) Add 125 µl Solution II and vortex to mix. Solution II should not be more than 1-2 months old. **Do not** let the minipreps sit in Solution II for more than 10 minutes before the next step.

7) Add 125 µl of Solution III and vortex. At this point a white ppte will form.

8) Spin @ 12K for 10 minutes.

9) Meanwhile, label a new set of eppendorfs and add 1 ml of 100% EtOH to each.

10) Add supernatant from miniprep to the EtOH and invert to mix (4-5 times).

11) Spin @ 12K for 10 minutes to ppte DNA (you will be able to see a white pellet but most of this is RNA).

12) Aspirate liquid and wash with 500 µl 70% EtOH.

13) Spin @ 12K for 5 min.

14) Aspirate liquid and quick spin again to bring down any remaining liquid. Aspirate the remaining liquid. Be careful not to suck up the pellet! I put a yellow tip on my aspirator to reduce the power.

15) Add 50 µl TE containing 500 µg/ml RNase A (50 µl of 10 mg/ml stock in 1 ml of TE).

16) Set up digest of DNA using 5 µl of miniprep.