ANTIBIOTICS PREPARATION

ADAPTED FROM THE BONNET TEAM PROTOCOL REPOSITORY

IMPORTANT: **Dissolve antibiotic powder in solution and prepare in sterile conditions.**

MATERIALS:

- Antibiotic powders (usually in the common fridge in the flow cytometer room)
- ddH2O
- 100% ethanol solution (under the chemical hood)
- Syringes
- 0.2µm filters
- 50mL tubes
- 1.5mL sterile centrifuge tubes
- 10mL sterile pipettes

PROTOCOL:

. Weigh the powder.
. Add ~50% of desired liquid to a 50mL tube.
. Add powder to liquid in tube and mix to dissolve.
. Adjust the volume to the desired final value.
. Filter the solution, ONLY if the antibiotic is diluted in water, under the hood PSM.
. Aliquot the solution under the hood PSM by 1mL in 1.5mL tubes using a 10mL pipette.
. Store aliquots at -20°C.

***Note: Carbenicillin can be used instead of ampicillin. Carbenicillin is more stable, so it is potentially more effective at selecting only bacteria containing the plasmids of interest (for example, fewer satellite colonies will grow). However, it is also more expensive.**

***To use : dilute antibiotic into your LB medium at 1:1,000. For example, to make 100 mL of LB/ampicillin growth media, add 100 µL of a 100 mg/mL ampicillin stock (1000X stock) to 100 mL of LB.**

Stability of antibiotics in agar plates
Ampicillin, Carbenicillin, Penicillin ->1 month
Kanamycin, Spectinomycin, Chloramphenicol -> 2 months
Streptomycin, Erythromycin, Neomycin -> 2 months