

## **Day 1 Procedure**

1. Dilute a 2 mg/mL solution of ampicillin in Luria Broth (LB) to 40 mL of each of the desired concentrations (0, 1, 2, 5, 10, 20 µg/mL). Distribute and mix the solution evenly for approximately 10 seconds.
2. Prepare 21 culture tubes total, with 3 for each concentration AND negative control. Transfer 5 mL of each solution into each tube using a p1000 micropipette.
3. Submerge a metal inoculation tube in 70% isopropyl alcohol for 30 seconds. Then, sterilize the loop at the tip of the inner flame of a lit Bunsen burner.
4. Place a single colony of E.coli bacteria in each tube using the inoculation loop.
5. Using sterile forceps, place a black bead into each culture tube excluding the negative controls, which contain no bead.
6. Preset a water bath with a submerged orbital shaker to 37 degrees Celsius and a shaking speed of 60 rpm. Grow the tubes in the orbital shaker overnight.