Microbiology Lab Experiment Changes

Experiment #: none

Title: Gradient Plate: Isolation of Streptomycin Resistant Mutant

Live Organisms: *E. coli*

Changes: Procedure
(Work in groups, do two groups per table)

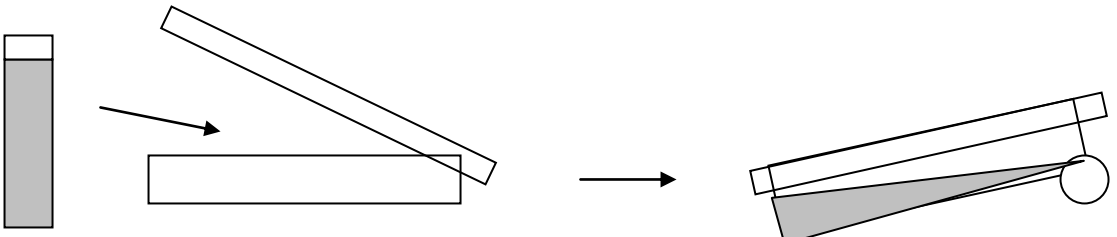
1. TSA deeps have been melted and are being held at 45-50°C in a water bath.

2. Follow diagrams below. Pour one melted agar deep into an empty Petri dish, then put a colored pencil (it has the right thickness) under one end and wait 15 minutes.

3. Next remove pencil from under Petri dish. Add 1.0 mL of colored Streptomycin solution to the remaining melted agar deep. Invert tube to mix and pour on top of first layer of agar. The streptomycin solution contains a colored dye to allow the gradient to be visualized. Wait 15 more minutes.

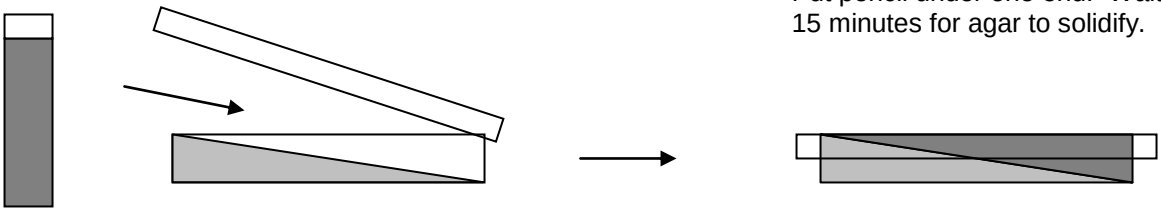
4. After the agar plate has completely solidified, spread 0.2 mL of the *E. coli* culture onto the surface of the agar. Your instructor will demonstrate this technique.

Take Home Lesson: Why would you expect to find any streptomycin resistant mutants? What is the purpose of the streptomycin? Does streptomycin induce (cause) mutations? What are some of the possible mechanisms a mutant might use to circumvent the streptomycin?



First agar deep

Put pencil under one end. Wait 15 minutes for agar to solidify.



Second agar deep + 1.0 mL streptomycin

Wait 15 minutes then spread 2 drops of *E. coli* on surface.