BIOL 4140/8146
Fall 2016

Protocol

Aseptic Technique and Media Preparation

One of the most important skills you can learn in cell culture is proper aseptic technique. Cells in culture do not have the immune system or barriers that cells in your body have to protect them from infection. They are very vulnerable to bacterial, viral or fungal infections. Additionally, when cell lines become infected it can be disastrous for experiments! We will be practicing aseptic technique today by working in the laminar flow hood to prepare mock media.

Materials

Sterile

- 1 T25 flask
- DMEM media
- 1x PBS (in place of Bovine Serum)

Procedure

1. You will be preparing a T25 flask of cell culture media. A T25 flask holds 5mL of media. You will be making D10 media. Please calculate the amount of each material that will needed to be added to your T25 flask before proceeding. The formula is:
   D10 = 90% DMEM + 10% Bovine Serum (use PBS in place of BS)
2. Prepare the laminar hood (BSC) for the aseptic technique.
3. Make a T25 flask of D10 media aseptically using the techniques you have learned and the calculations you prepared.
4. Prepare the laminar hood for the next student and place your properly labeled T25 flask in the incubator.

*I will be checking your flasks in 2-5 days. There should be **NO** growth if you performed proper aseptic technique. If your flask shows growth, I will be asking you to repeat this experiment Thursday during class time.*