LAB PRACTICAL EXAM I

1. Prepare a 50 ml 2% FBS/RPMI/Strep-Pen medium.
   a. Indicate each reagent, FBS, Antibiotic, RPMI? ______________ (2 pts)
   b. FBS volume? __________________________________________ (2 pts)
   c. Antibiotic volume? _____________________________________ (2 pts)
   d. RPMI volume? ________________________________________ (2 pts)

2. Pipetting technique assessment.
   a. Indicate the proper pipettes to be used for each volume?
      i. 1,000 μL ____________________________ (1 pt)
      ii. <100 μL ____________________________ (1 pt)
      iii. <20 μL ____________________________ (1 pt)
   b. Demonstrate pipetting technique for 8 mL? ________________ (2 pts)

3. Focus on the tissue culture flask that contains the cells? ____________ (2pts)

4. Demonstrate trypsinization technique.
   a. Outline steps (5 pts)
      i. ______________________________________________________
      ii. ______________________________________________________
      iii. ______________________________________________________
      iv. ______________________________________________________
      v. ______________________________________________________
   b. Show floating cells under the microscope. ________________ (3 pts)
   c. Spin cells down at appropriate speed/time. ________________ (2 pts)
   d. Discard medium and disperse cell pellet ________________ (2 pts)

5. Cell counting with a hemocytometer.
   a. Outline steps (5 pts)
      i. ______________________________________________________
      ii. ______________________________________________________
      iii. ______________________________________________________
      iv. ______________________________________________________
      v. ______________________________________________________
   b. Prepare a trypan blue suspension for 5 μL cell suspension ____(2 pts)
   c. Perform a x5 dilution ____________________________ (2 pts)
   d. Perform a cell count under the microscope ________________ (2 pts)
   e. Determine cell count as cells/mL __________________________ (2 pts)

6. Culture cells in a 25 cm² flask at 1x10⁵ cells/mL, 10 mL final volume.
   a. Show calculation based on your cell concentration (3 pts)
      _______________________________________________________
      _______________________________________________________
      _______________________________________________________
   b. Carry out the sub-culturing (3 pts)
      ________________________________

END