

## Picking a colony, Complementation & Photosynthesis Pre-lab.

Name \_\_\_\_\_ GSI & Sect # \_\_\_\_\_ Station # \_\_\_\_\_

To answer these questions read all of the lab material on constructing recombinant DNA (pages 64-90) even though you are only doing pages 77 & 78 for lab 4. You also need to read about complementation (pages 111-118) even though you are only doing pages 111-113. You also need to read about photosynthesis on pages 119-133. Write legibly. You will NOT be admitted to lab unless you turn in this completed pre-lab at the start!

1. What color of colony should you select on your LB/AMP/X-gal plate if you had an insert?  
What color of colony should you select on your LB/AMP/X-gal plate if you had no insert?

2. What is histidine? What is the ploidy of the mutant yeast cells that you are using in the complementation experiment? Are they auxotrophs, or prototrophs? Explain your answer.

3. In your own words, very briefly describe how one performs pair-wise mating of yeast strains.

4. What is a complementation group?

5. State the color/or absence of color of the following solutions.

- a) a suspension of concentrated chloroplasts
- b) a 1M solution of DCPIP
- c) a 1M solution of DCPIP<sub>2</sub>

6. What is an absorption spectrum? How would you prepare an absorption spectrum for a purified solution of Beta carotene dissolved in acetone. You have all of the materials and equipment as outlined in the lab manual.

7. In this experiment you are measuring photosynthesis by examining electron transfer (reduction of DCPIP). List two other parameters you could measure as indicators of rates of photosynthesis (the experimental design can be different).

**Pre-lab continued on the backside.**

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8. In lab, you will prepare five treatments for the Hill reaction.

a) What is the final concentration of the standardized chloroplast suspension that you will use in the Hill reactions?

b) The final step in preparing a standardized 0.1 mg/ml chlorophyll solution is to add "X" amount of your enriched chloroplast suspension to 5 ml of cold sucrose phosphate buffer. Therefore you must determine how much of your concentrated chloroplast solution (p. 110, step 5i) that you need to add. Fill in the following table for the given O.D. values.

O.D. Value	Final Volume (V <sub>f</sub> )	X = Volume of enriched chloroplast suspension to 5 ml of cold sucrose phosphate buffer
1.0		
0.6		
0.4		

c) How would residual DCMU (as a result of failing to wash & rinse out your test tubes) affect rates of photosynthesis in your light reactions?