NAME _____

pGLO TRANSFORMATION LAB QUESTIONS

ANSWER THIS QUESTION BEFORE STARTING THE EXPERIMENT

IDENTIFY what the following abbreviations stand for:

LB = _____

amp = _____

ara = _____

FOLLOW DIRECTIONS TO INNOCULATE THE PLATES , STACK THEM, BOTTOMS FACING UP, TAPE THE STACK TOGETHER, LABEL WITH YOUR NAMES, AND PUT INTO INCUBATOR. ANSWER THESE QUESTIONS **NOW** BEFORE COLLECTING THE DATA AND ANALYZING THE RESULTS TOMORROW.

1. The pGLO plasmid contains the following **GENES**.
The transcription/translation of these genes produces what **PHENOTYPE**?

*ori*_____

*GFP*_____

bla _____

ara operon _____

2. What is the difference between pGLO and GFP?

_____

3. If the GFP is a "glowing protein", why did the tube containing just the pGLO plasmid not glow before it was added to the +pGLO bacterial tube?

_____

4. How did the **addition of $CaCl_2$** and **"heat shocking"** help facilitate the incorporation of the pGLO plasmids into the *E. coli* bacteria?

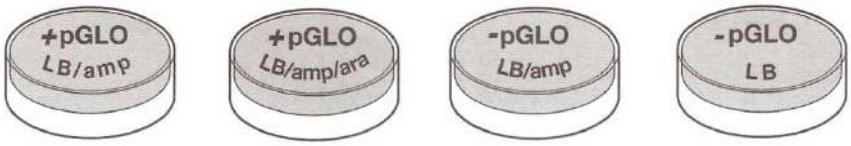
_____

_____

5. What are "reporter" genes and how are they used?

_____

_____



6. On which plates would you expect to find bacteria most like the original non-transformed *E.coli* colonies you initially observed? **EXPLAIN YOUR ANSWER**

_____

_____

7. If there are any genetically transformed bacterial cells, on which plate(s) would you expect to find them? **EXPLAIN YOUR ANSWER**

_____

_____

8. If there are any genetically transformed bacterial cells, on which plate(s) would you expect them to GLOW if exposed to UV light? **EXPLAIN YOUR ANSWER**

_____

_____

9. Which 2 plates should be compared to determine if any genetic transformation has occurred? **WHY?**

_____

_____

10. The GFP gene shares the same promoter as the *ara* operon, which codes for enzymes that break down arabinose sugar if it is present. EXPLAIN how this operon works and how the addition of arabinose to bacterial cells which have picked up the pGLO plasmid induces them to GLOW. DRAW A PICTURE IF THAT HELPS.

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_____

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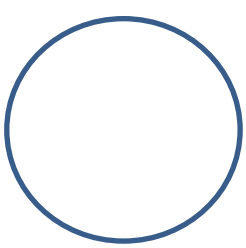
_____

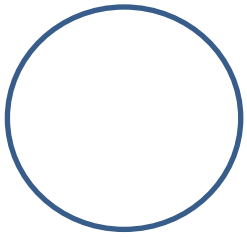
DAY 2:

REMOVE YOUR PLATES FROM THE INCUBATOR AND OBSERVE THE RESULTS

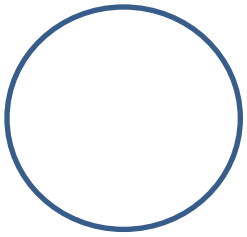
Draw pictures below showing any bacterial growth on the plates. Mark any growth on the chart.



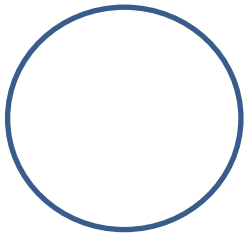
-pGLO, LB



-pGLO, LB/Amp



+ pGLO, LB/Amp



+ pGLO, LB/Amp/Ara

DESCRIBE what happened on each of the different plates and **EXPLAIN** your results. **Don't just say "bacteria grew on the plate". What characteristics did the bacteria have that allowed them to grow on the plates AND WHY?**

-pGLO, LB _____

_____

_____

-pGLO, LB/Amp _____

_____

_____

+ pGLO, LB/Amp _____

_____

_____

+lGLO/LB/amp/ara _____

_____

_____

QUESTIONS

1. What is a plasmid?

_____

_____

2. How are plasmids used in genetic engineering? Give some examples.
(Use your Campbell book/Recombinant plasmid handout to help you)

_____

_____

_____

_____

3. Why do bacteria on the +pGLO /LB/amp plate not "glow" if they picked up the pGLO plasmid?

_____

_____

4. The *ara* operon on the pGLO plasmid to which the GFP gene is attached is most like which operon you modeled with the pool noodles? **EXPLAIN YOUR ANSWER.**

_____

_____

5. What advantage would there be for an organism to be able to turn on or off particular genes in response to certain environmental conditions?

_____

_____

_____

6. Other than creating "cool glowing organisms" , how is the pGLO plasmid used as a DNA technology tool?

_____

_____