

Bacterial Transformation: the pGLO™ System

8-2

OBSERVATIONS AND INTERPRETATIONS

- Record the appearance (clear, violet, or fluorescent green) of the pGLO™ plasmid solution in natural light and in the dark with only UV illumination.
- Record your observations of the plates after incubation in the table below.

| Plate | Inoculum | Number of Colonies | Colony Appearance in Ambient Light | Colony Appearance with UV Light |
|------------|-----------------------------|---------------------|------------------------------------|---------------------------------|
| LB | <i>E. coli</i> (-pGLO™ DNA) | | | |
| LB/amp | <i>E. coli</i> (-pGLO™ DNA) | | | |
| LB/amp | <i>E. coli</i> (+pGLO™ DNA) | Able to grow on amp | Green | Green |
| LB/amp/ara | <i>E. coli</i> (+pGLO™ DNA) | | Green | Green |

QUESTIONS

- Fill in the genotype of *E. coli* prior to transformation and after transformation for the GFP and the *bla* genes.

| <i>E. coli</i> | GFP Gene (GFP ⁺ or GFP ⁻) | <i>bla</i> (<i>bla</i> ⁺ or <i>bla</i> ⁻) |
|-----------------------|--|---|
| Before transformation | | |
| After transformation | | |

- What was the purpose of examining the original pGLO™ solution with and without UV illumination?
- What was the purpose of transferring the + DNA and - DNA tubes from ice to hot water to ice again?
- Why were microtubes incubated for 10 minutes in LB broth rather than transferring their contents directly to the plates?

Pglo Transformation Lab Answer Key

J Ma



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