Exploring the Expression of Green Fluorescent Protein (GFP) if selected by a Transformed *E.coli* in Bacteria

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Background

- The Green Fluorescent protein (GFP) is commonly used as a model in biotechnology due to its bioluminescent properties (Chalfie et al.,1995).
- GFP was first found in the *Aequorea Victoria* Jellyfish

Figure 1: Image of the *Aequorea Victoria* Jellyfish expressing GFP

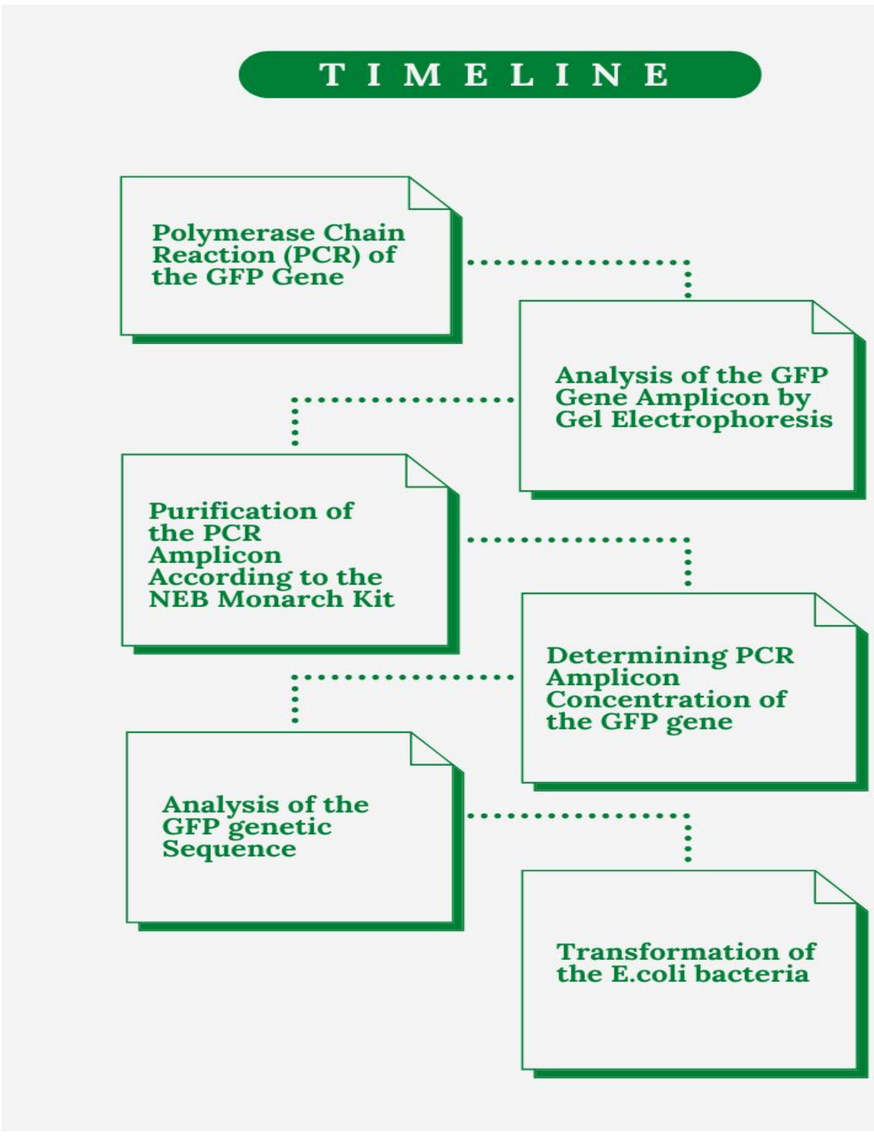
- Fluorescent properties of GFP may be used in order to identify the presence of proteins in organic structures (Zhou et al., 2014).
- In this experiment, the Green Fluorescent Protein (GFP) gene present in the pGLO plasmid was characterized
- pGLO plasmid was transformed into *E. coli* bacteria with ampicillin, Luria Broth, and arabinose, in order to lead to growth of *E. coli* colonies

Specific Aim

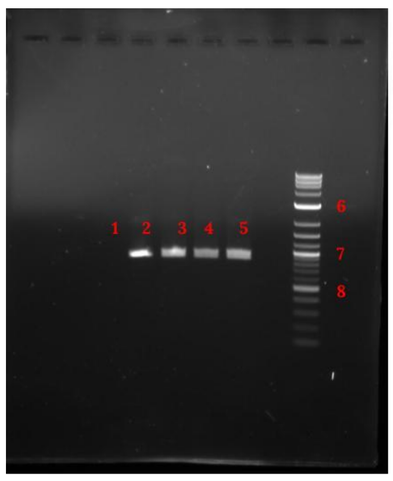
Research Question: Will the GFP protein be expressed when it is transformed into the E.coli bacteria.

Hypothesis: If the pGLO plasmid survives inside the E.coli bacteria then the GFP protein will be expressed in the presence of arabinose.

Methods

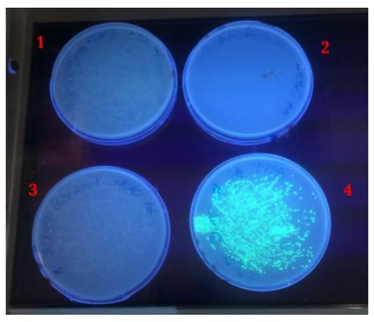


Results



1. Negative Control
2. Positive Control
3. GFP amplicon
4. GFP amplicon
5. GFP amplicon
6. Base pair ladder (3,000 bp)
7. Base pair ladder (1,000 bp)
8. Base pair ladder (500 bp)

Figure 2: Image of the Electrophoresis Gel marker 1 shows the negative control, marker 2 shows the positive control, markers 3 through 5 show GFP amplicons, and marker 6 shows the base-pair ladder used to find the approximate base-pair length.



1. -pGLO / LB
2. -pGLO / LB + AMP
3. +pGLO / LB + AMP
4. +pGLO / LB +ARA + AMP

Key:
LB - Luria Broth
ARA - Arabinose
AMP - Ampicillin Antibiotic
-pGLO - Not Containing the pGLO plasmid
+pGLO - Containing the pGLO plasmid

Figure 3: Image of the four agar plates. plate 1 shows the -pGLO plate with the Luria Broth, plate 2 shows the -pGLO plate with the Luria broth and Ampicillin antibiotic, plate 3 shows the +pGLO plate with the Luria broth and the ampicillin antibiotic, plate 4 shows the +pGLO plate with the Luria broth, arabinose, and ampicillin antibiotic.

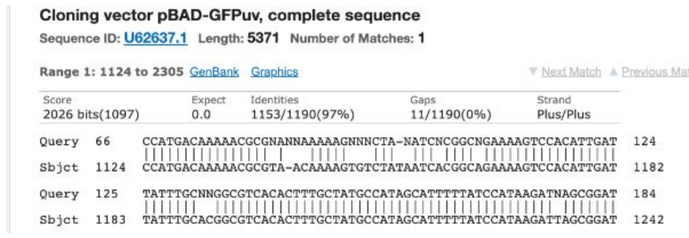
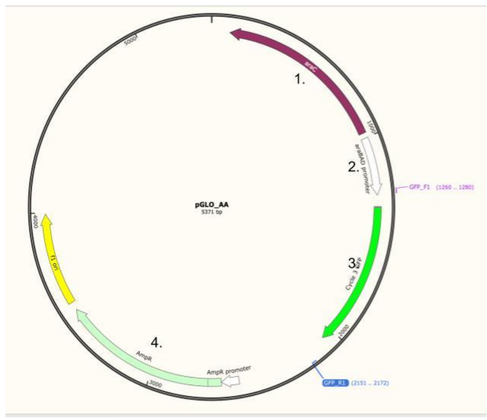


Figure 4: shows genetic sequence alignment of a pBAD-GFP clowning vector compared to the genetic sequence of the GFP amplicon.



1. Arabinose-C gene
2. pBAD promoter
3. GFP gene sequence
4. AmpR Gene sequence

Figure 5: shows the genetic map of the pGLO plasmid breaking down the the Arabinose-C gene, pBAD promoter, GFP gene, and the AmpR gene

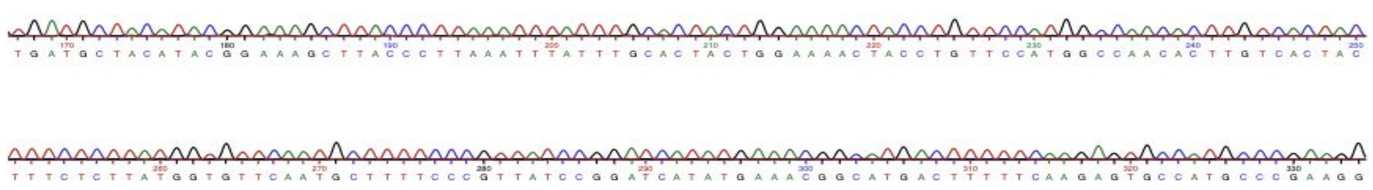


Figure 6: shows the chromatogram of the GFP Amplicon

Conclusions

- This study proved that under the correct circumstances and when transformed with the pGLO plasmid the *E.coli* bacteria will express GFP.
- some limitations for our study were:
  - The number of trials conducted
  - The PCR Amplicon could have been contaminated when sent to be sequenced by Eurofins Genomics
  - Experimental error may lead to imprecise results
- Future directions may be composed of using the GFP gene in pGLO to observe localization of proteins that lead to diseases.

References

Chalfie, M. 1995. Green fluorescent protein. *Photochemistry and Photobiology*, 62, 651 656.

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Tsien Y. Roger. 1988. THE GREEN FLUORESCENT PROTEIN. Howard Hughes Medical Institute; University of California, San Diego.

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