

MOTIVATION

- Some Bacteria have evolved resistance against all available drugs
- An estimated 35,000 Americans die of Antibiotic Resistant infections every year
- Due to misuse and overuse of antibiotics, bacteria have started evolving a defense mechanism against antibiotic drugs

ABSTRACT

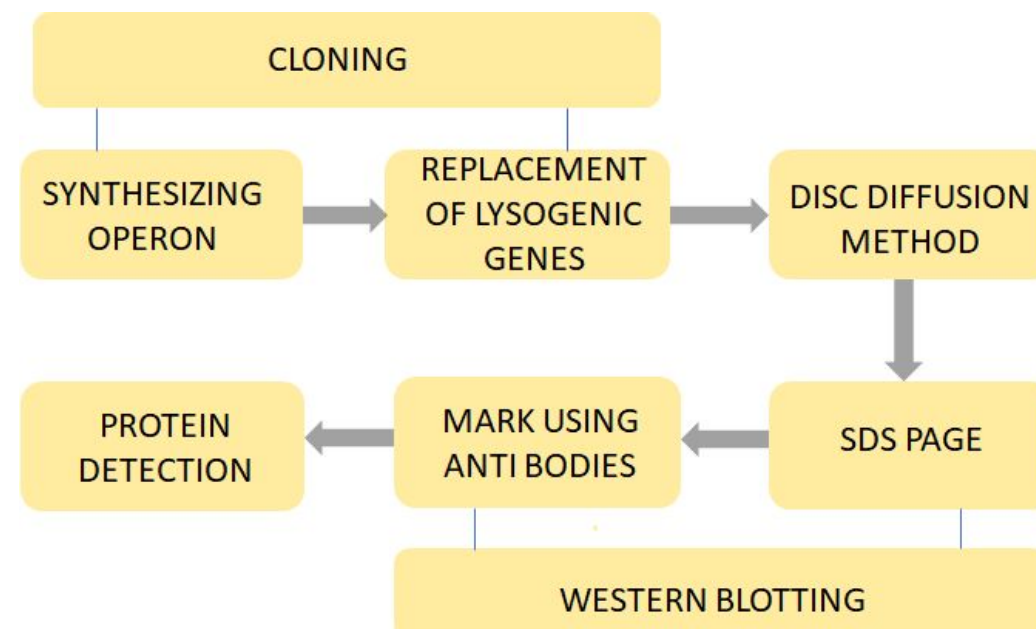
- Mutations have made bacteria Antibiotic-Resistant
- Bacteria also evolve to fight with Phage using CRISPR
- This project aims to make a stronger phage so that even if the bacteria get evolved, still it gets killed
- For this, in the Phage DNA, we are removing the section of gene sequence responsible for lysogeny and cloning a newly made operon in its place
- We check for the inhibiting activity by selected bacteriolytic enzymes by using Western Blotting method.

Phage vs. Antibiotic

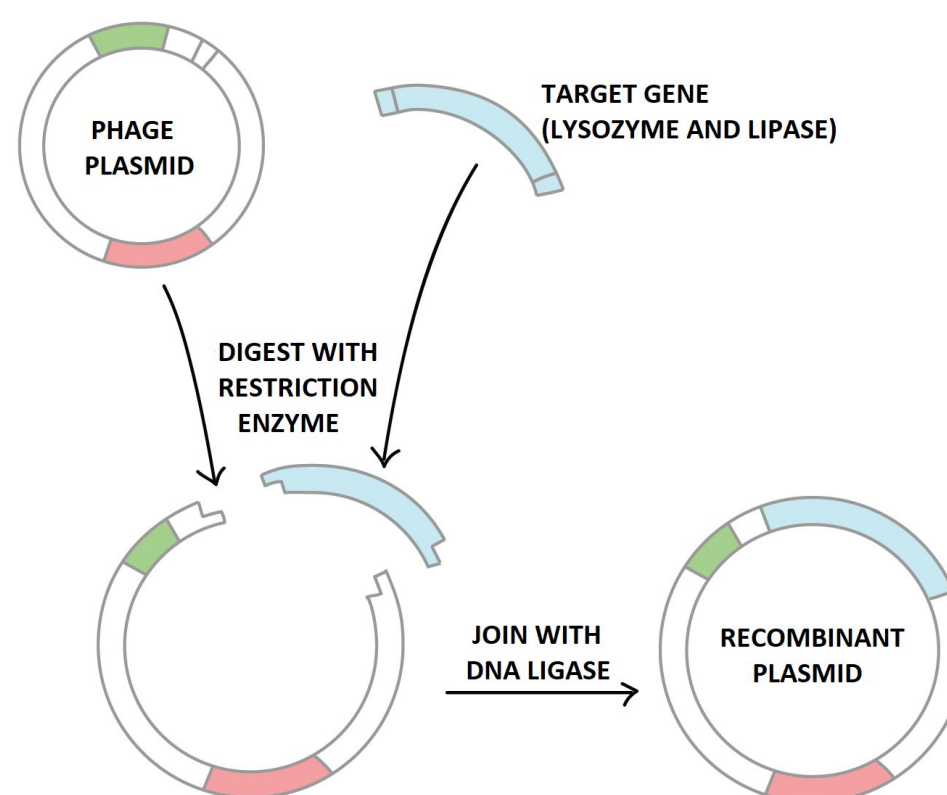
- Phage is **highly specific** towards particular bacteria
- Phage **coevolves** with bacteria to have advanced mechanisms for defense and attack

METHODOLOGY

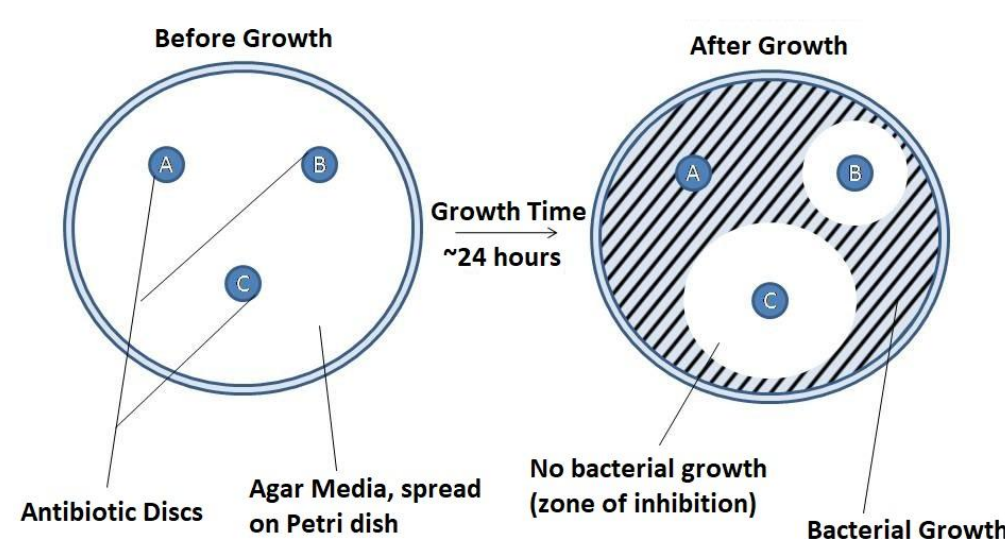
Cloning is the concept of introduction of foreign genetic material to some system. For example, introducing a non-native operon into a Bacteriophage.



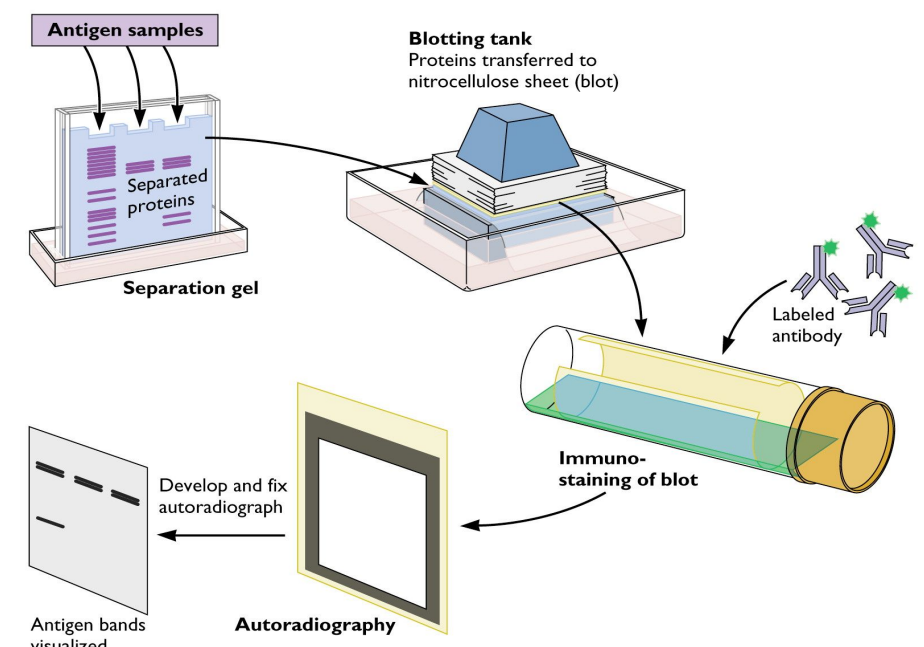
- Plasmid genes are separated, purified and ligated using Ligase Enzyme
- Section of Gene Sequence responsible for Lysogeny is replaced with newly made operon



- The solution is placed in a filter paper disc
- Disc Diffusion Method is used on a streaked Petri plate to attain Zone of Inhibitions



- Proteins are separated from the Zone of Inhibition using SDS-Page method
- Antibodies with specific markers are used for the identification of desired proteins



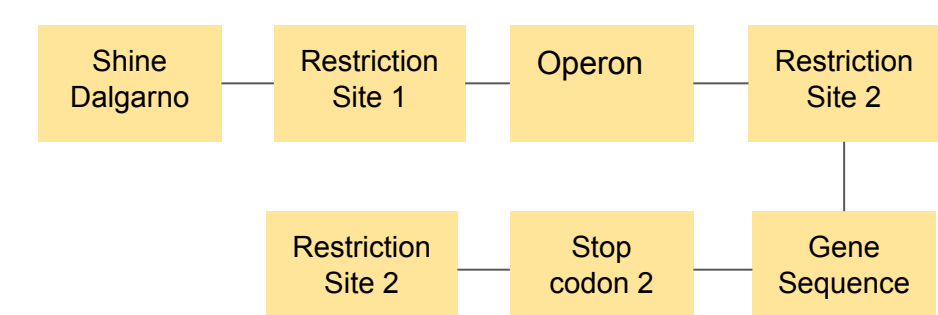
SELECTION OF ENZYME

We have selected the enzyme based on their activities

- Lysozyme - It shows antibacterial mechanisms
- Lipase - It performs lipolytic activity thus acts to be cidal for the pathogenic bacteria

C1 and Cro genes are responsible for switching between Lytic and Lysogenic states

FORMED OPERON:



SCOPE

Further, this can be checked on a disease-causing drug-resistant microbe to treat the patients with available permissions for tests. This technique can save the life of patients on a very large scale and can act as an ultimate solution to Antibiotic Resistance.

REFERENCES

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