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4-14-2022

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Kayla Botto

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Botto, Kayla, "Genomic Similarity to Predict Infection in Bacillus Phages" (2022). *Research and Creativity Symposium*. 147.

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Genomic Similarity to Predict Infection in *Bacillus* Phages

By Kayla Botto
Faculty: Dr. Lynn Lewis
University of Mary Washington

Introduction

Bacteriophages

- Viruses that infect bacteria
- Commonly called phages
- Made of DNA/RNA and proteins
- Most diverse ‘organism’ on the planet
- Extremely specific to their host(s)

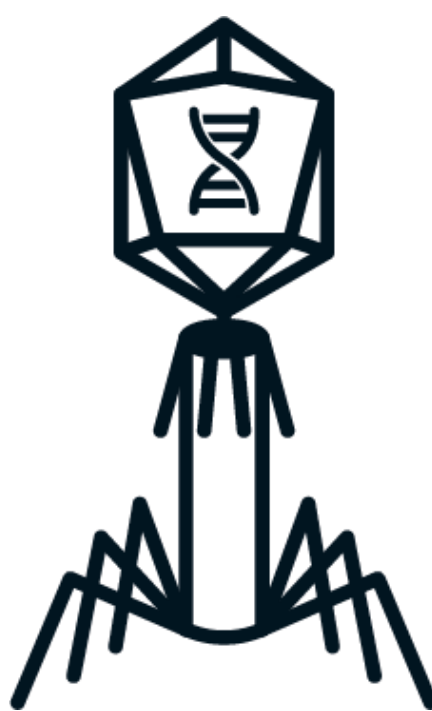


Figure 1: a T4 bacteriophage

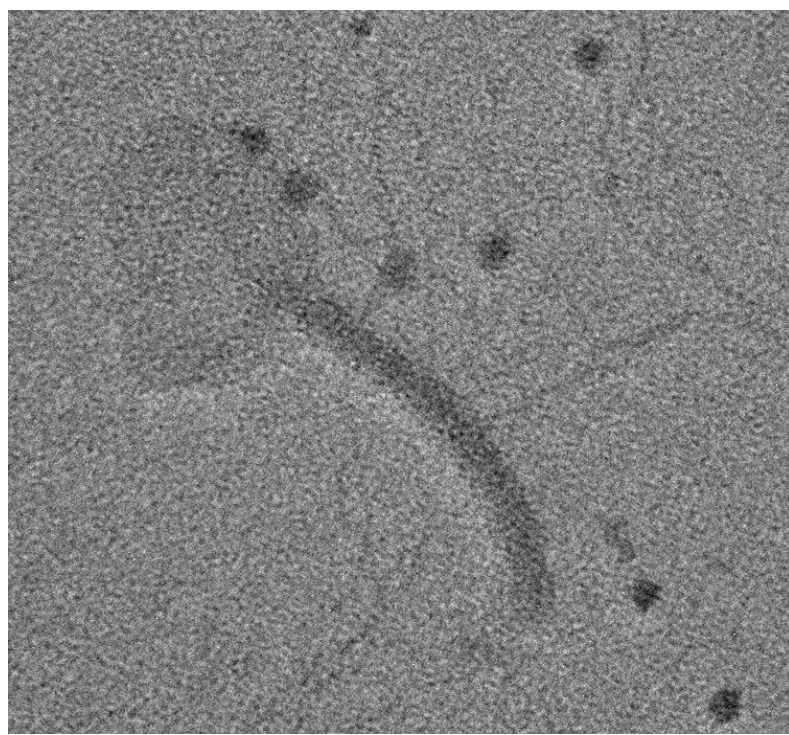


Figure 2: an electron micrograph of phage Grumio

Bacteria

- Host used was *Bacillus thuringiensis kurstaki* (Btk)
- Btk is closely related to *B. anthracis*
- Anthrax is caused by *B. anthracis*

Phage Therapy

- Uses phages to treat bacterial infections
- Currently experimental in the US
- Commonplace in some countries, such as Georgia
- Individualized treatment for each patient
- Must find phages that infect patient's specific bacteria
- Similar bacteria are more likely to be infected by the same phage

Problem: tedious to find phages that infect patient's specific bacteria and genes responsible for selectivity of hosts

Hypothetical solution:

A phage is more likely to infect a host that it is more genetically similar to and genes that control infection selection will be highly conserved

Materials

Sequenced Phages

1. Grumio
2. Freight Train
3. Cletus
4. Jack Rabbit
5. Hari

Bacteria

1. Btk
2. Btk 33679
3. B. t. Al Hakam
4. B. t. 350
5. B. cereus
6. B. cereus #4
7. B. cereus 14579
8. B. subtilis
9. B. subtilis 23857
10. B. subtilis B
11. B. anthracis delta Sterne
12. B. amyloliquefaciens
13. B. pumilus
14. B. sphaericus
15. B. megaterium
16. B. simplex

Host Range Study

Methods

- Grow many different strains of *Bacillus* bacteria
- Grow phages
- Spot phages on plates with different bacteria
- See which phages grow on which bacteria
- Perform many replicates

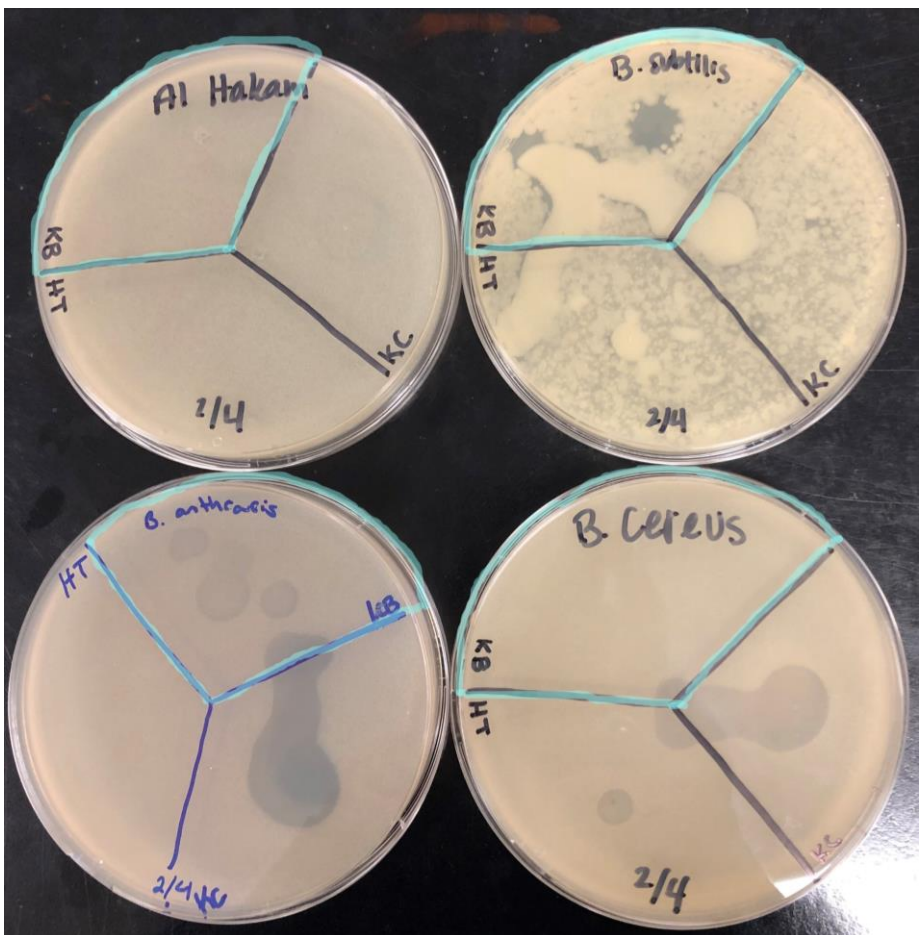


Figure 3: 4 plates each with a different bacterium on it and the 3 same phages on each

Results

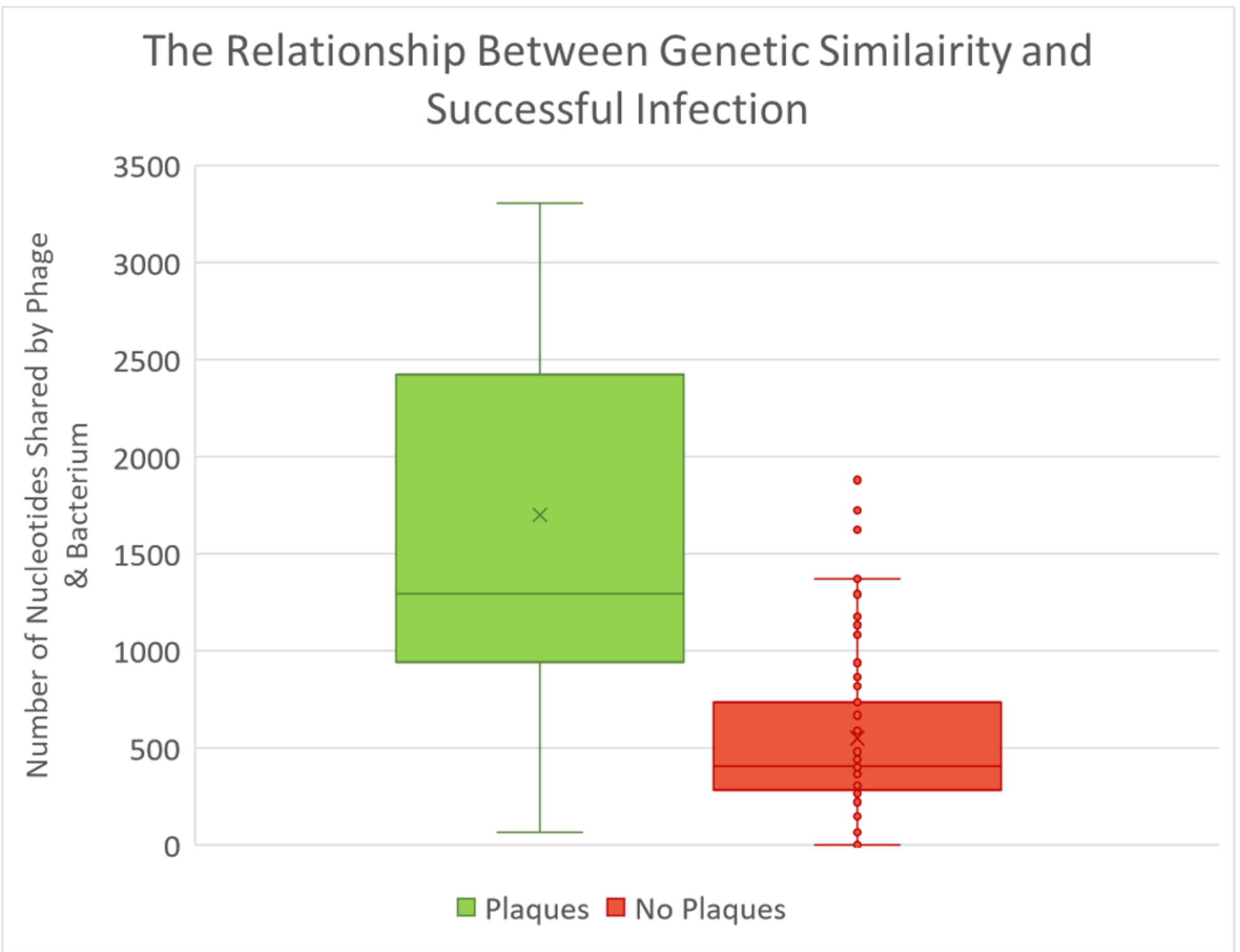


Figure 4: A box and whiskers plot where genetic similarity between a given phage and bacterium compared to successful infection (green) and unsuccessful infection (red). Success of infection and genetic similarity were significantly positive correlated (Spearman, p=0.06)

Genetic Comparison

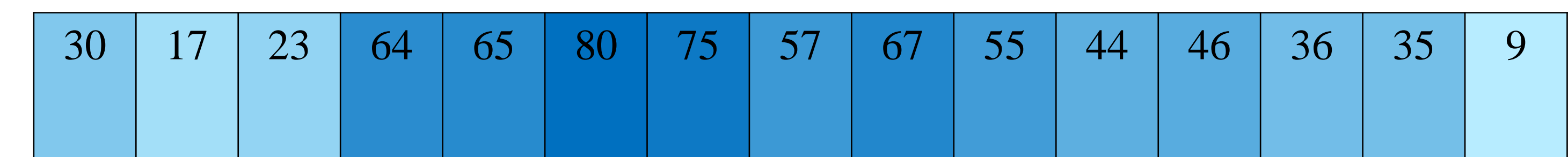
Methods

- BLASTn each phage genome against each bacterial genome for similarity in number of nucleotides
- BLAST each phage against each other to find similarity between phages
- Use DNA Master to identify genes in shared regions and note features of interest of genes
- Genes shared by all phages are predicted to be essential
- Compare phages that can infect the same bacteria to find possible genes that cause infection

Genetic Comparison (Cont.)

Results

Similarity Across the Phage Genome



Start less similar more similar End

Figure 5: Color graded graph showing amount of genetic similarity relative to position in the genome of the phage. Each section represents 20 genes.

- Genes shared by all phages and predicted to be essential were used for
 - replication of viral DNA such as DNA binding-protein & DNA polymerase II
 - and assembly of virions such as major capsid protein & baseplate assembly protein

Conclusions

1. There is a positive relationship between genomic similarity of bacteria and phage and the production of a successful infection
2. These 5 phages had high genetic similarity overall and genes that are predicted to be essential have putative functions related to replication and assembly

Future Work

1. Continue to analyze similarities between phage genomes
2. Perform another host range study with different phages
3. Confirm the putative function of genes that are predicted to be essential

References

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2. Phage Therapy : Current Research and Applications, *edited by Andrzej Gorski, et al., Caister Academic Press, 2014.* ProQuest Ebook Central, <http://ebookcentral.proquest.com/lib/umw/detail.action?docID=5897878>.
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Acknowledgements

Thank you to Dr. Lewis, Dr. Agrawal, and Dr. Morriss for being wonderful mentors.
Thank you to UMW and the NSF for giving me the opportunity to do my own research.