

Annotation of Mycobacterium Phage Stagni through DNA Master

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Abstract

Last semester, our Bioinformatics class annotated the DNA Sequences belonging to two of our previously isolated bacteriophages, Stagni and Kady. It can be difficult to completely understand our process if it is not yet understood what exactly it means to “annotate” a gene. Gene annotation involves describing different fragments of a DNA sequence with the hopes of revealing which sections of that sequence is actually in fact a gene. In short, annotation ultimately reveals a DNA sequence’s entire genome arrangement. Our annotation procedure required us to identify where exactly individual genes were located and additionally required us to determine all of the coding areas on those genes. There are many different approaches that could be taken to do the following. Our process, however, included prediction based methods (which allowed us to find genes/gene structures based on nucleotide sequence) and sequence similarity methods (which allowed us to align our DNA with other sequences and proteins from different or similar species for comparison). We used a combination of software informers to do the following, including the freeware DNA sequence editor DNA Master and the computer program Phamerator. Synchronized use of the two programs allowed us to break down our phage’s DNA sequence into various components and generate genome maps that incorporated nucleotide and amino acid sequence relationships. As a result, we were able to assort our protein-coding genes into “phamilies” of related sequences using comparisons to generate a database of gene relationships.

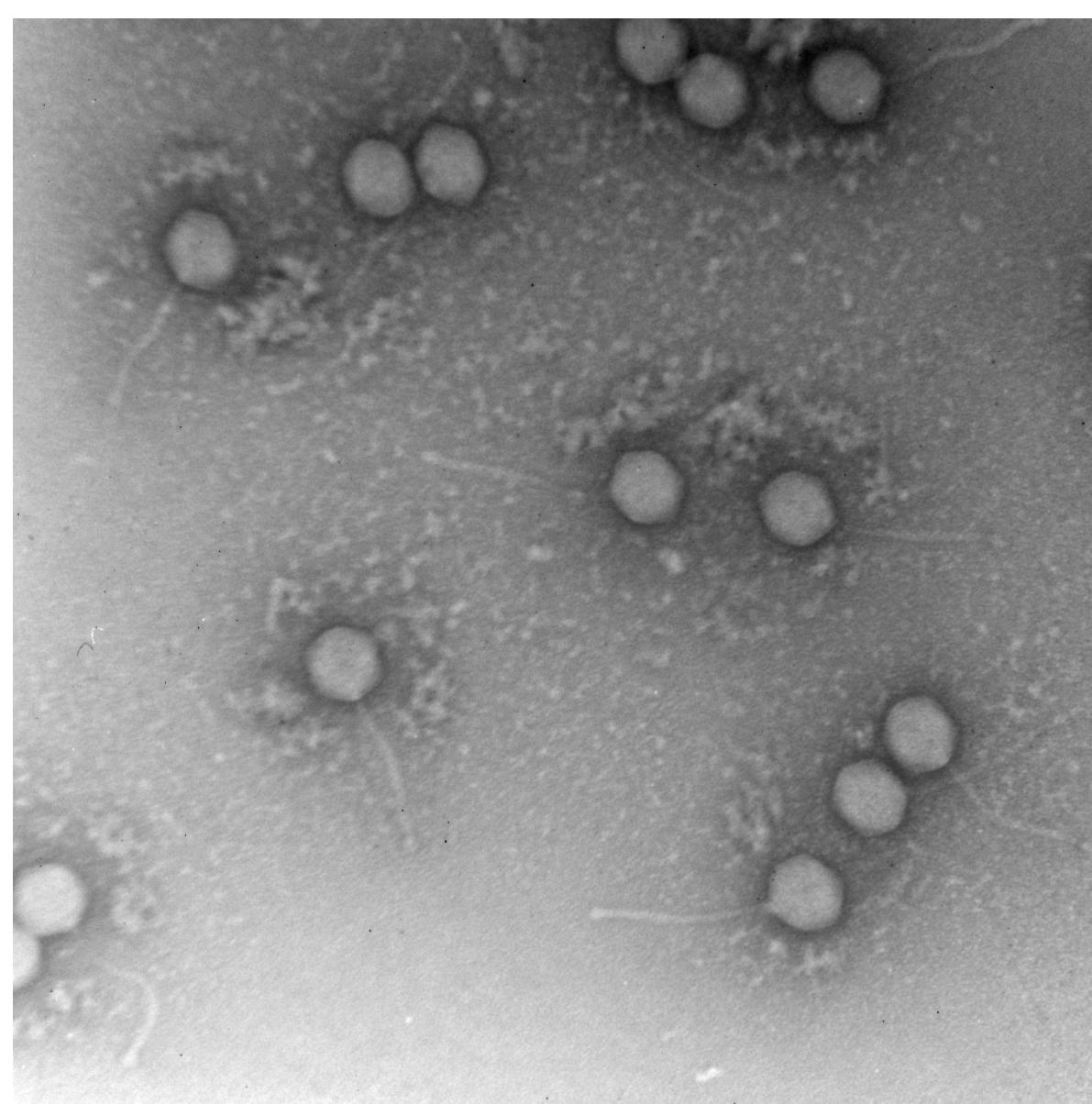


Fig. 1
Electron Microscopy image of Stagni

Introduction

- Bacteriophages, also known as phages, are viruses that destroy bacteria upon infection. Destruction of bacteria is achieved through the rapid replication and infection of phage. These phages require a bacterial host in order to replicate themselves.
- Our Bacteriophage, Stagni, was isolated from a lake in Livonia, Michigan
- *Mycobacterium smegmatis* was used as a surrogate host for our Bacteriophage
- Our phage’s DNA was analyzed using Gel Electrophoresis, a method used for separation analysis of macromolecules
- Pictures of our phage were obtained through Electron Microscopy.
- The genome of our phage was studied and revealed through DNA Master, a genome annotation and exploration program
- Stagni’s genes were annotated, revealing it’s entire genome arrangement.
- 34 of Stagni’s genes were annotated by different groups in our class. Data obtained from each group was compared with the data of other’s in separate groups
- Phamerator program was used to compare our bacteriophage’s genome to other well known bacteriophage genomes

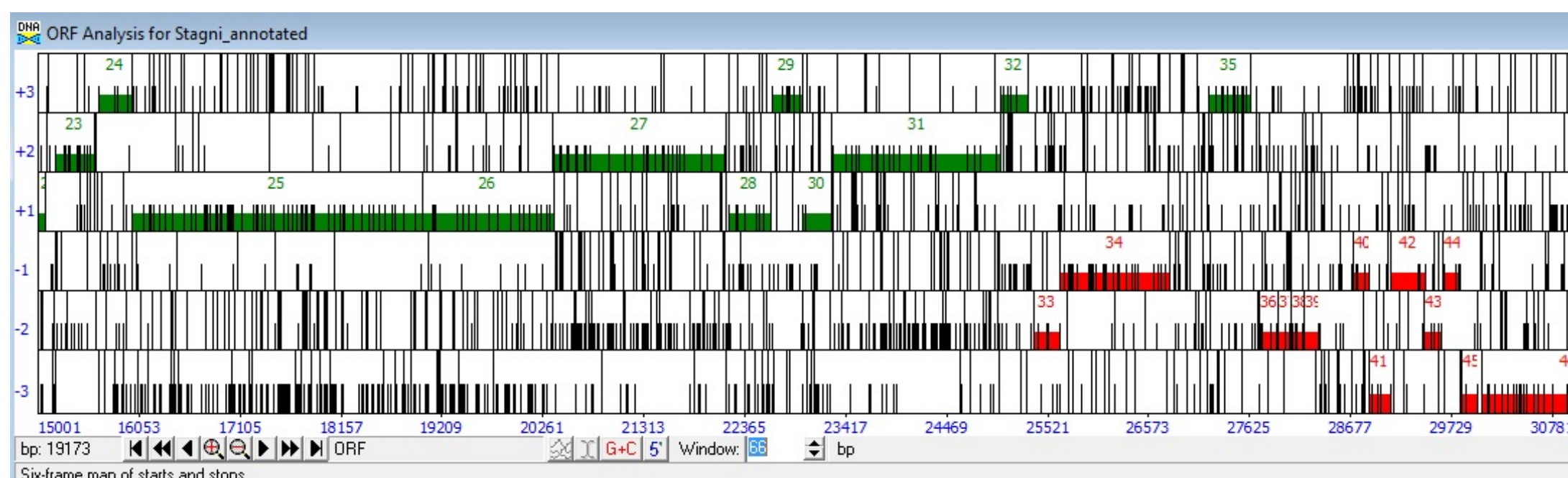
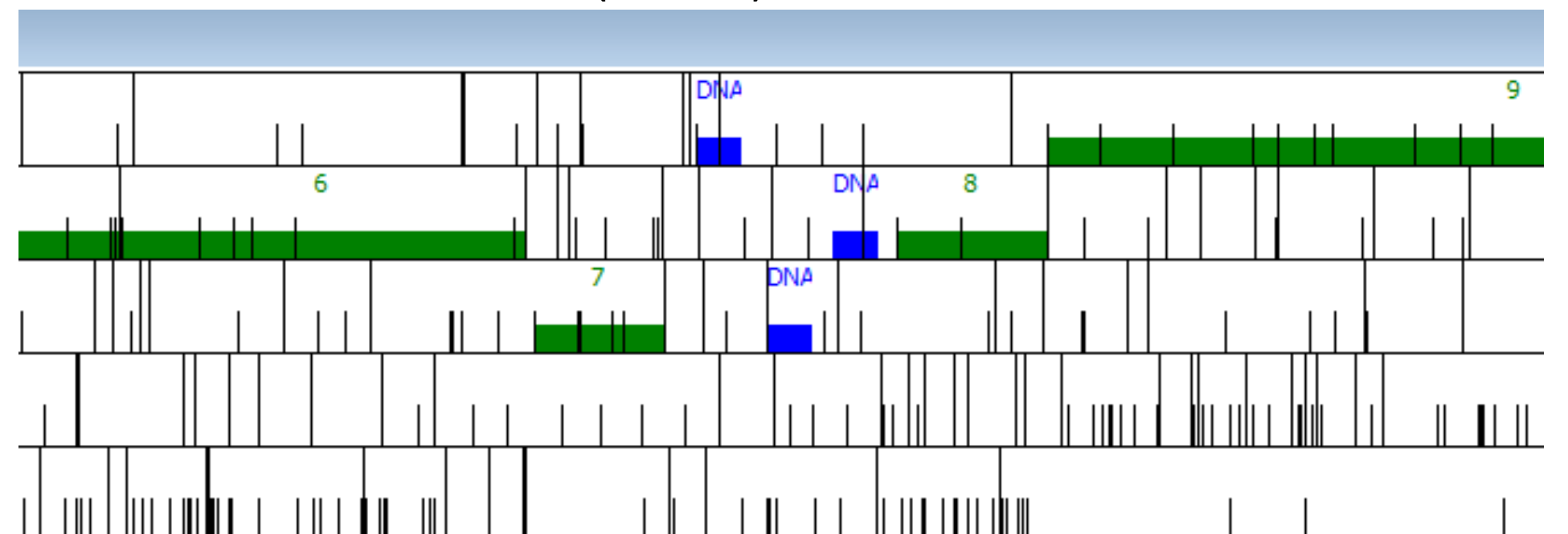


Fig. 2
Image of the DNA Master program shows genes that were annotated (forward genes are green and reverse genes are red)

Fig. 3
Image of the DNA Master program shows the tRNA genes that were discovered and annotated (in blue)



Conclusion

Therapeutic Phage therapy is the use of Bacteriophages to treat pathogenic infections. Phage Therapy has many potential applications in human medicine, agriculture, and dentistry. Since the genetic make-up of Stagni has been studied extensively, Stagni’s genome can be compared to the genomes of unfamiliar Bacteriophage. Additional research of Stagni’s genome can reveal characteristics of new, unique Bacteriophage that are capable of treating bacterial infections, thus creating advances in medicine.

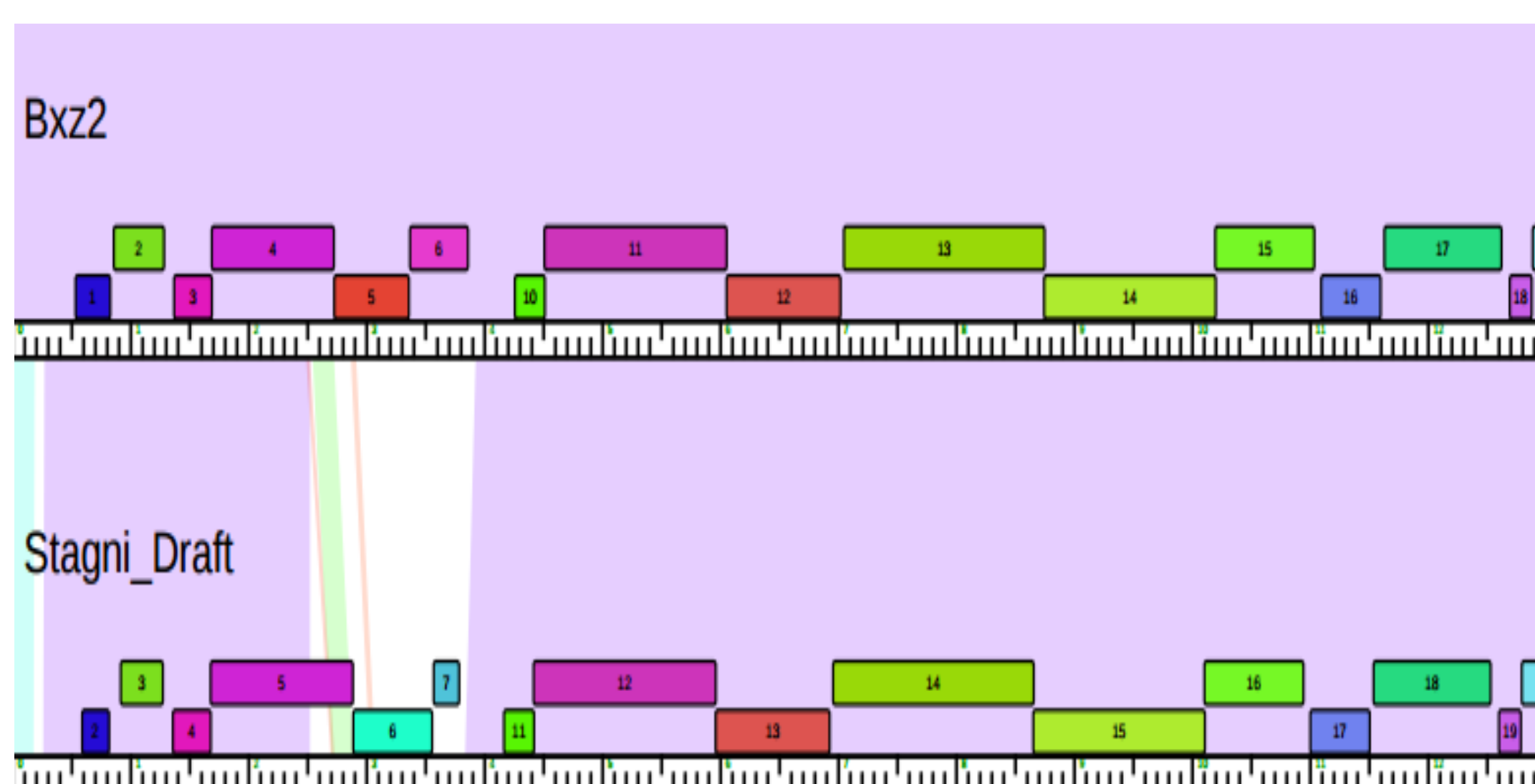


Fig. 4
Image of the Phamerator program compares the genomes of Stagni and a similar phage, Bxz2. The images shows that the genetic make-up of the two are very similar.