Phage Resistance and tRNA expression in Marine Cyanobacteria

João Felipe Gueiros, Daniel Kurtz and Adi Rivkin; Mentor: Sophia Zborowsky, MSc.

This project was conducted at Prof. Debbie Lindell's lab, Faculty of Biology, Technion University

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Abstract

Unicellular marine cyanobacteria of the genera *Synechococcus* and *Prochlorococcus* account for 13% of global oxygen production.[1] Viruses which infect cyanobacteria, called cyanophages, are one of the major factors influencing cyanobacteria. To reproduce, phages must infect a host cell, and by hijacking cellular mechanisms, they multiply, leading to cell death. Many bacteria employ various strategies to resist infection. Little is known about defense mechanisms in cyanobacteria[2]. In a recent study conducted at the Lindell Lab at the Technion, a cyanobacterial strain, which has intracellular resistance to a phage, was proven to be resistant due to its lack of expression of specific tRNA molecules, which are used by the phage to reproduce inside its host. In this study, three different cyanobacterial strains were tested for expression of tRNAs in order to determine if the phenomenon is replicated in other strains. Lack of tRNA expression was observed in 1/3 and could be the mode of resistance in this strain.

Genetic Code and Translation

1. DNA is copied to messenger RNA (mRNA) which is used as a template to build proteins. A set of 3 nucleotides comprise a codon, which codes for a specific amino acid, which are the "building blocks" of proteins.
2. Transfer RNA molecules (tRNA) transport amino acids to the ribosome. Each tRNA molecule is bonded to a specific amino acid, and has an anticodon, which corresponds to a codon on the mRNA to which it bonds.
3. The movement of tRNA through the ribosome as it binds to the mRNA allows the ribosome to build a protein as dictated by the codons on the mRNA.

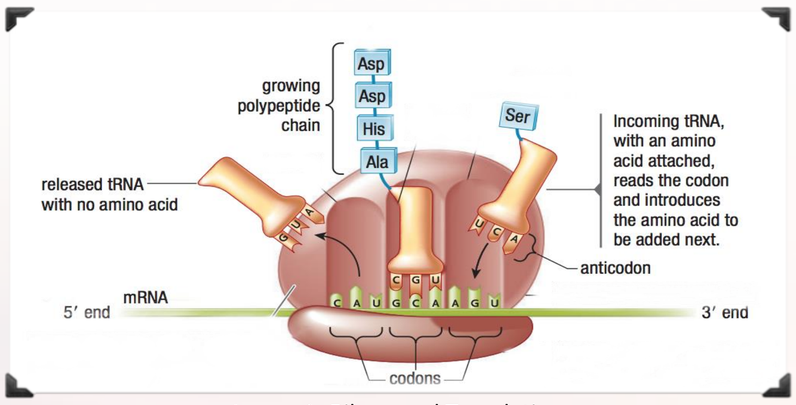

Image 1: Ribosomal Translation
Fraser et al, Nelson Education, 2012

Methods

Infection of cyanobacteria with Syn9 phage → Extraction of cyanobacteria and phage RNA → Reverse Transcription → Quantitative Real Time PCR → Calculation of tRNA expression levels

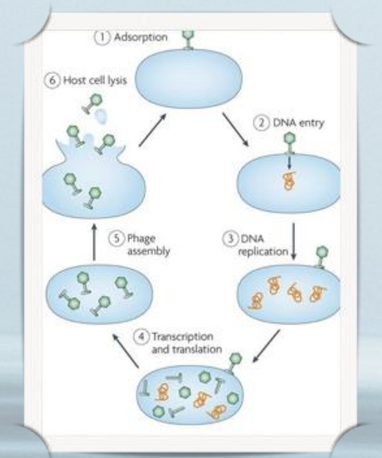

Image 2: Viral Replication Cycle
Labrie et al, 2010

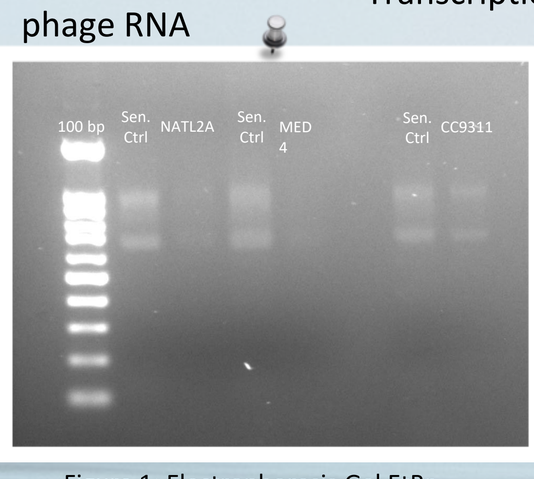

Figure 1: Electrophoresis Gel EtBr staining showing the extracted RNA

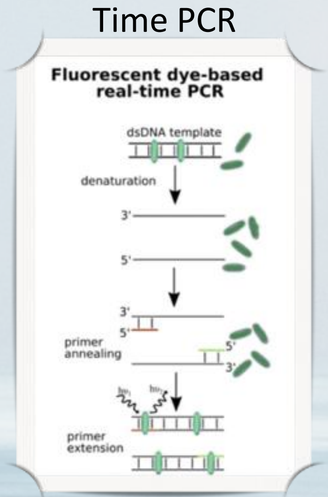

Image 3: Scheme of quantitative PCR procedure
Qiagen

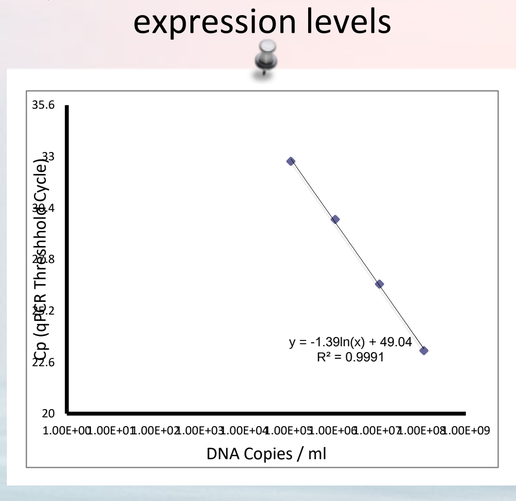

Figure 2: Example of a standard curve graph used to calculate DNA copies based on fluorescence

Lack of cyanobacteria and phage tRNA expression may prevent phage replication inside *Prochlorococcus* MED4

Results

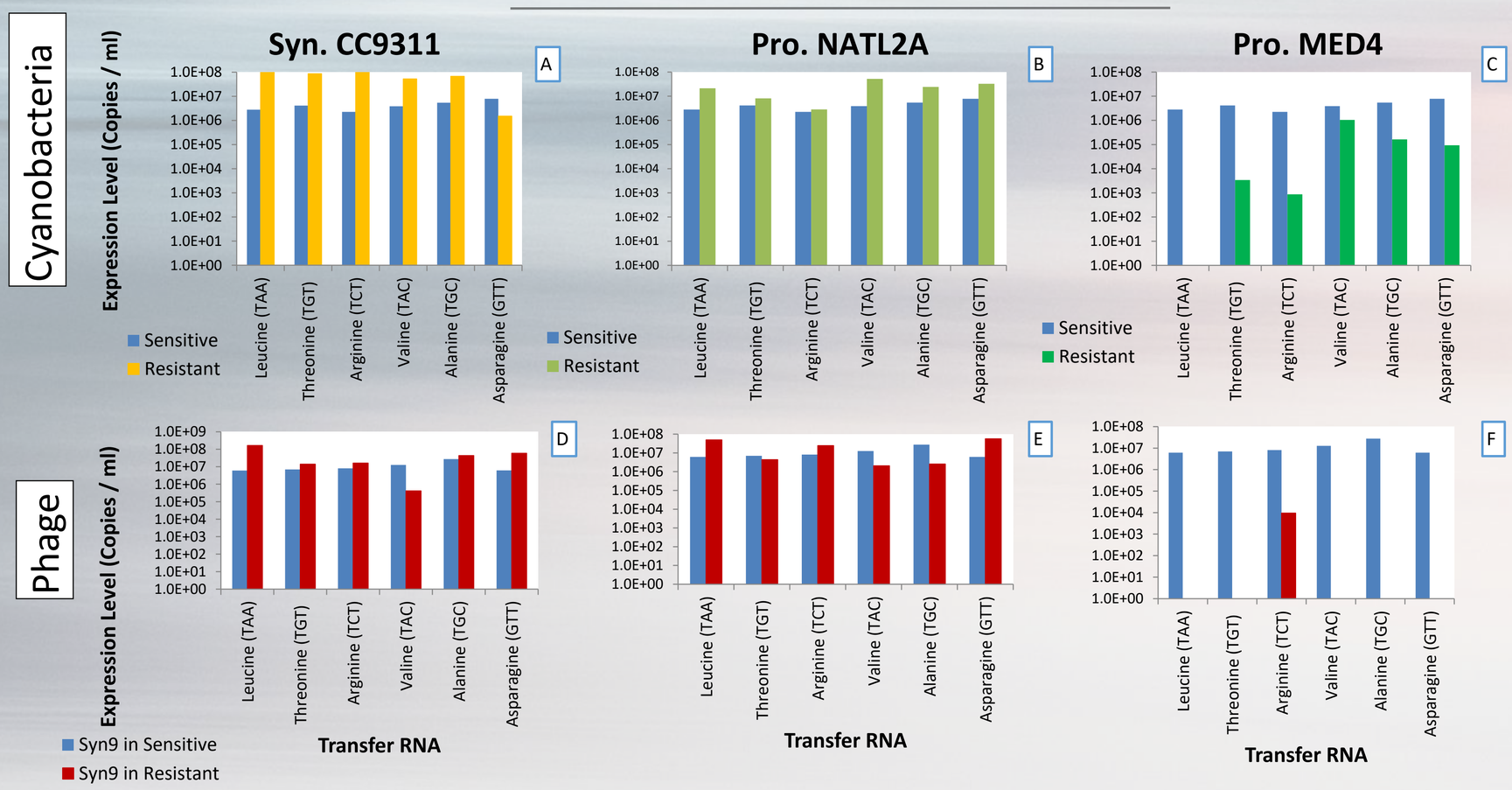

Figure 3: tRNA expression levels. Top row: cyanobacterial. Bottom row: phage

Conclusions

Cyanobacteria:

- In Syn. CC9311 (Fig.3A) and Pro. NATL2A (Fig.3B) the tRNA genes were expressed.
- In Pro. MED4 (Fig.3C) half of the tRNA genes investigated had little to no expression.

Phage:

- In Syn. CC9311 (Fig.3D) and Pro. NATL2A (Fig.3E) the phage tRNA genes were expressed.
- In Pro. MED4 (Fig.3F) the phage tRNAs tested had little to no expression.

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References

1. P. Flombaum et al., Present and future global distributions of the marine cyano-bacteria Prochlorococcus and Synechococcus. Proc. Natl. Acad. Sci. U.S.A. 110, 9824– 9829 (2013).
2. C. A. Suttle, A. M. Chan, Marine cyanophages infecting oceanic and coastal strains of Synechococcus–Abundance, morphology, cross-infectivity and growth-characteristics. Mar. Ecol. Prog. Ser. 92, 99–109 (1993).