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Comparative Analysis of Cluster FF CI Repressor genes

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Temperate bacteriophages are capable of integrating their DNA into a host bacterium's genome by switching off their lytic life cycle in favor of the lysogenic cycle. Extensive research has been done on the enterobacteriophage lambda to understand the genes that control lysogeny and has identified three regulatory genes, the CI repressor, CII and CIII.   
  
Guinevere, a temperate actinobacteriophage, was isolated from soil on Stevenson University's campus using an enriched isolation procedure and was sequenced at the Pittsburgh Bacteriophage Institute using the Illumina MiSeq platform. Guinevere's genome is currently being annotated at Stevenson and is 42116 bp long with a GC content of 64.9%, 67 predicted genes, and 3 predicted tRNAs. Guinevere, an FF cluster phage, presents with clear, haloed, uniform plaques, which is not typical for temperate phages, however, some other FF phages such as Gusanita and Zaheer share this quality.   
  
Synteny is generally well conserved among FF phages, with most genes being on the forward strand. A ~6 kb region in the middle of the genome contains genes on the reverse strand assumed to play a role in lysogeny, such as integrases. FF phages Elesar and QuinnAvery are exceptions, in that they are missing some genes within this region that are present in the other FF phages. The last gene in this 6 kb section (gp 43) had a high-quality match to lambda's CI repressor on HHPred and contained its helix-turn-helix domain but interestingly lacked the C-terminal dimerization domain. A comparative analysis of this gene was done across all annotated FF phages to determine if this finding was consistent. The last gene (pham 220155) in this region in Cole, Elesar, Nandita, Guinevere, Gusanita, Zaheer, Ryan, Popper, and QuinnAvery were analyzed in HHPred and all returned with high quality matches (>90%) to the lambda CI repressor except for Ryan, which had a score of 84.67%. The C-terminal dimerization domain was missing in these FF phages, but the helix-turn-helix domain was present with varying degrees of amino acid similarity.   
   
The lack of a C-terminal dimerization domain suggests that a functional repressor may not be made, which could have consequences for lysogeny. A multiple sequence alignment was performed using Clustal Omega and used to generate a phylogenetic tree of these same FF protein sequences and the lambda CI repressor sequence. No correlation was seen between clade relationships and turbidity of plaques, suggesting that a different gene may be responsible for clear plaques in temperate phages. Additionally, the lack of a C-terminal dimerization domain in the genes analyzed suggests that a CI repressor may not be present in FF phages and instead, other genes that involve helix-turn-helix domains are responsible for maintaining lysogeny.