May 14, 2015

Dear SMART member,

Please find attached the Hope College Fall 2014/Spring 2015 Mycobacteriophage Glass annotation file and author list. Glass is a Subcluster B2 phage with very high identity to previously sequenced B2 phages. Below is a series of notes highlighting things we believe you should be aware of regarding the annotation.

**Rearrangements from gp60 to gp64:** We deleted one ORF originally called as gp62 in one version of the auto annotation analysis by DNA Master that yielded a total of 91 potential ORFs. This forward strand ORF is found in the draft phamerator map for Glass, fully overlapping a reverse ORF (originally gp61; 54346-54194). In addition to the overlap of the opposite strand ORF, the original gp62 is called only in other draft phage genomes, has no coding potential, and was called only by Genemark. Other changes in the region include extension of the original gp61 start site (54496-54194, now gp62); the addition of gp61 that covers coding potential but does not have any BLAST hits; and the addition of gp63 which matches gp61 of Rosebush. We believe that the Qyrzula homolog of Bella96 gp63 should be recalled to extend its current start site based on a tblastx analysis. See the note for gp63. *This entire region should be carefully considered.*

**Additions:** We added four ORFs – gp61, gp63, gp71, gp82.

**Of Special Note:**

**gp5** – We annotated this ORF as QueU base on the PhagesDB Hedgerow blast hit, but the phamerator map of Rosebush and Ares annotate this as QueE. Please advise as to which annotation should be used.

**gp68** – This ORF has a 200 bp overlap with the previous gene; we feel this is justified. See the note for explanation.

**gp71** – This ORF was added and has a 151 bp overlap with the downstream gene. The overlapping ORFs are convergent, so the stop codons dictate the amount of overlap. See the note for explanation.

Thank you for your efforts on the QC process. Please let me know if you have any questions or concerns.

Sincerely,



Aaron A. Best, Ph.D.