Search and analysis of prophages related to phage AP-16-3 in genomes of *Sinorhizobium spp.*

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Motivation and Aim

Bacteriophages able to cause lytic infection of bacteria, which is accompanying by host cells metabolism reprogramming and their destruction in the result. Alternatively way is when bacteriophages initiate lysogenic infection of bacteria cell by integrating into chromosome, then prophages could be transmitted to subsequent generations. Rhizobiophage 16-3 [NCBI RefSeq NC_011103] was identified in the genome of the *Rhizobium (Sinorhizobium) meliloti* Rm41 in the 1960s. It is one of the first well studied bacteriophages of nodule bacteria of *S. meliloti* species. In this paper data on the AP-16-3 phage isolated by us in mountainous region of Dagestan, which genome showed high similarity to genome of the phage 16-3, is presented. So, it was a point of interest to evaluate an abundance of phage sequences similar to the AP-16-3 phage in strains from the genus *Sinorhizobium*.

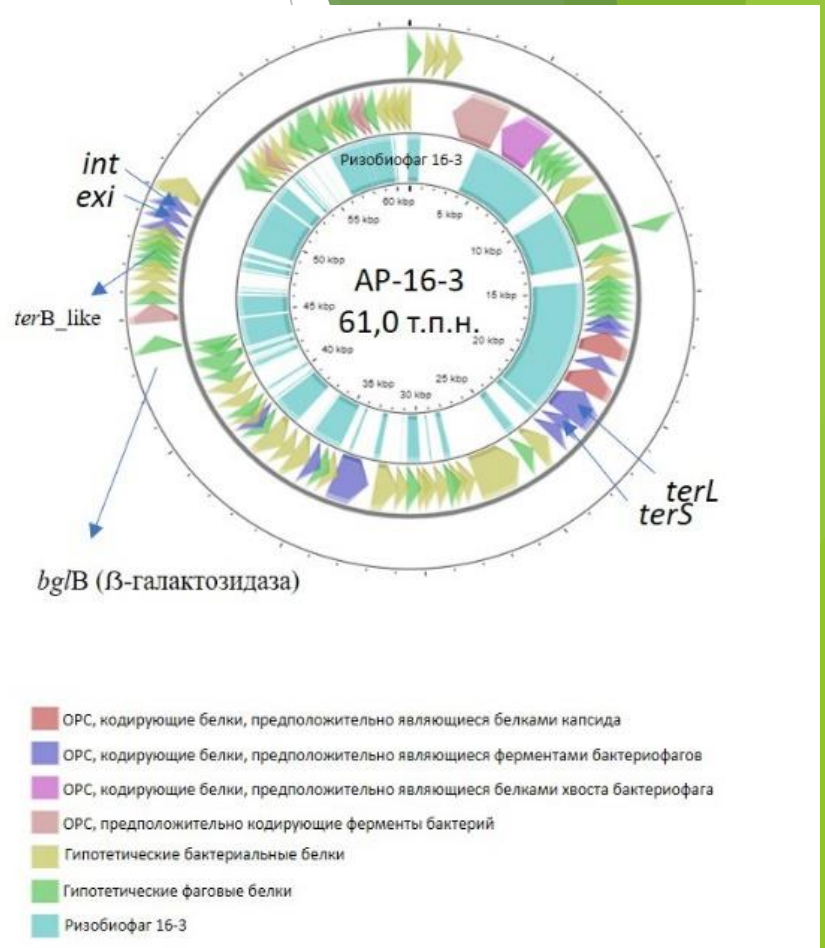
Methods and Algorithms

The AP-16-3 phage genome was sequenced using MiSeq, Illumina, assembled (SPAdes, Flye, Racon and Medaka modules, Pilon), and annotated by Prokka. The BLASTn and BLASTp tools were used for genome analysis. Prophage sequences were searched in 53 strains of *S. meliloti*, in 9 strains of *S. medicae*, and in 7 strains of *S. fredii* (NCBI database) using the PHASTER web server. The phylogenetic tree was constructed using the OPTSIL algorithm of VICTOR web tool.

Results

The object of the study was soil bacteriophage AP-16-3, which showed a high similarity with the temperate rhizobiophage 16-3. The phage was isolated from a soil sample collected at the northern mountainous region of Dagestan, Caucasus. The AP-16-3 phage genome was 61 kb, and 94 ORFs were identified, among which were those whose predicted products are necessary for phage infection cycle. Sequences similar to Rhizobium phage AP-16-3 were detected in genomes of 23% of tested strains of *S. meliloti* and in single strains of *S. medicae* and *S. fredii*. Nucleotide sequence analysis showed that 6 out of 8 prophages were intact, while other two were incomplete or defective prophages. The size of the intact prophage reached up to 63 kb, while the size of defective and incomplete prophages were at about 33 and 22 kb, respectively. Identity of prophage sequences revealed in *Sinorhizobium* strains across AP-16-3 sequence was ranged from 79.74 to 92.45 (up to 43% coverage). The number of ORFs encoding protein sequences in intact prophages were up to 144, while in defective and incomplete prophages were at about 30. It was revealed that all prophages contained ORFs encoded tail proteins, but prophages were distinct by ORFs determining synthesis of structural elements of virion.

A phylogenetic analysis of the identified prophage sequences was carried out. The application of the OPTSIL algorithm showed that a set of tested phage sequences similar to AP-16-3 phage were related to at least 7 genera of viruses (bootstrap 57%), presumably of *Siphoviridae* family Thus, sequences homologous to Rhizobium phage AP-16-3 are in each fifth strain of *S. meliloti*, but these sequences are significantly distinct.



Conclusion

The obtained data support that phages relative to Rhizobium phage 16-3 had a wide range of lytic activity and they are abundont in soil microbiome.

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