Exploiting CRISPR-Cas systems to study phage biology

Sylvain Moineau*[1]

[1]Department of Biochemistry, Microbiology and Bioinformatics – Faculty of Sciences and Engineering
Université Laval, Quebec city, Quebec, Canada

Résumé

The year 2017 marked the 100th anniversary of the publication by Félix d'Hérelle, who introduced for the first time the word "bacteriophage" (1). With it, the field of phage biology was born. Viruses are now recognized at the most abundant biological entities on the planet and display a remarkable genetic diversity. Not surprisingly, bacteria have a plethora of diverse defense mechanisms to combat their phages. Four decades after the discovery of one such defense mechanism, restriction-modification systems, another bacterial anti-phage system that cleaves foreign DNA was identified-one that acts as an adaptive immune system. Clustered regularly interspaced short palindromic repeats (CRISPR) and their associated *cas* genes protect microbial cells against infection by foreign nucleic acids, including phage genomes and plasmids. Bacterial CRISPR-Cas type II systems function by first incorporating short DNA 'spacers', derived from invading phage genomes or plasmid sequences, into a CRISPR array located in their genome. This step is known as adaptation or vaccination. The CRISPR array is then transcribed and matured into short RNAs (the maturation step), which, by recruiting a Cas endonuclease, act as surveillance complexes that recognize and cleave invading matching sequences (the interference step). For some systems, the cleavage requires a short motif, called the PAM, close to the sequence targeted by the spacer. Exploiting this system has also resulted in the development of the much-publicized CRISPR-Cas9 technology for precise genome manipulation of various organisms. This seminar will recall the roles played by phages in the discovery and understanding of CRISPR-Cas systems. I will also highlight the recent emergence of virulent phages capable of inactivating the CRISPR-Cas system through anti-CRISPR proteins (2) as well as the use of CRISPR-Cas9 technology for viral genome editing in order to better understand phage-host interactions (3). Finally, I will briefly present the use of CRISPR-Cas systems and phages as teaching tools (4).

1- Félix d'Hérelle, F. 1917. Sur un microbe invisible antagoniste des bacilles dysentériques. *Comptes Rendus de l'Académie des Sciences* 165:373-375.

2- Hynes, A.P., G.M Rousseau, M.-L. Lemay, P. Horvath, D. Romero, C. Fremaux, and S. Moineau. An anti-CRISPR from a virulent streptococcal phage inhibits SpCas9. Nature Microbiology. DOI: 10.1038/s41564-017-0004-7.

3- Lemay, M.-L., D. Tremblay, and S. Moineau. 2017. Genome engineering of virulent lactococcal phages using CRISPR-Cas9. ACS Synthetic Biology. 21:1351-1358.

4- Trudel, L., M. Frenette, and S. Moineau. 2017. CRISPR-Cas in the laboratory classroom. Nature Microbiology. 2:17018

*Intervenant