

Bacteria belonging to the *Legionella* genus are intracellular pathogens that in the environment reside inside the cells of aquatic protozoa, primarily free-living amoebae belonging to the species *Acanthamoeba*, *Naegleria*, *Echinamoeba* and *Vermamoeba*. Once in artificial water distribution systems, *Legionella* can pose a threat to human health and life, causing infections collectively referred to as legionellosis. The similarity between amoebae and alveolar macrophages is the reason why *Legionella* bacteria cause pneumonia, Pontiac fever and extrapulmonary infections. Within the *Legionella* genus, more than 60 species have been described to date. In 90% of legionellosis cases, the identified etiologic agent is the *L. pneumophila* species, and in another 4%, *L. longbeachae*. However, at least 24 other *Legionella* species have been isolated from people with atypical pneumonia. This disparity in the frequency of pathogen identification is reflected in the paucity of knowledge regarding the other *Legionella* species, whose ecology or virulence has so far been poorly understood.

*Legionella* genomes are characterized by a complex architecture, with a chromosome and numerous plasmids that can vary widely in size and content. Genes shared between species have been shown to account for only 15% of the genus' total gene pool. The variability of plasmids seems to be of particular interest because they are a vector for so-called horizontal gene transfer, by which a given bacterial population effectively acquires traits that allow it to achieve high fitness in the environment, which often also means achieving high virulence. Knowledge of plasmids found in *Legionella* bacteria is fragmentary. There is a lack of information regarding the processes responsible for their persistence and spread, as well as their role in the evolution of *Legionella* bacteria. An additional variable is that *Legionella* bacteria have a complex lifestyle in which the replicative phase takes place inside the eukaryotic host cell.

Within the framework of this project, we want to answer questions to what extent horizontal gene transfer and recombination are responsible for the variability of *Legionella* bacteria, which can lead to the emergence of pathogenic strains in the population. To achieve this, we plan, through molecular experiments and bioinformatics analyses of genomic sequences, to build a picture of the total plasmids of this group of bacteria (plasmidome). This picture will include information on the stability and spreading ability of individual plasmids, their content, co-occurrence and co-evolution of specific gene layouts, as well as the frequency of horizontal gene transfer involving plasmids, between different *Legionella* bacteria in the environment, as well as inside the eukaryotic host cell. This will significantly add to the knowledge of this group of bacteria and allow better prediction and prevention of potential legionellosis outbreaks in the future.