

Disease-causing microorganisms possess an arsenal of weapons, that target crucial elements of host defense systems. In many bacteria, a key role in this process is played by a specific structure resembling a syringe. It delivers proteins, called effectors, into cells of the organism attacked. The effectors are mainly involved in suppression of defense system. Since such effectors often show no similarity to the known proteins, possess unique enzymatic activities or novel protein folds, it is extremely challenging to unravel their detailed function. It is known however, that an effector set at disposal of a given bacterial strain determines its host range, and presence or absence of a single effector may affect successful colonization of a particular plant species.

The aim of this proposal is characterization of HopBF1 effector from *Pseudomonas syringae*, a bacterium that infects almost 200 various plant species including important crops. Using advanced bioinformatics analyses we have revealed that HopBF1 displays features reminiscent of proteins involved in phosphate group transfer to other proteins. Although the overall similarity to this protein family, referred to as kinases, is relatively low, our pilot experiments confirmed this enzymatic activity. Interestingly, our studies show that similar proteins are encrypted in DNA of other bacterial species. Predicted unique protein structure, together with its enzymatic activity proven in our preliminary assays, allow us to conclude that HopBF1 may represent a novel subfamily of the kinases.

In this proposal we would like to i) describe HopBF1 properties; ii) determine its subcellular localization; iii) identify genes that are transcribed in HopBF1-dependent manner; iv) find interacting host proteins, and iv) finally, test whether HopBF1 presence promotes bacterial virulence and how its ability to phosphorylate contributes to disease development. To address these questions we will adopt a multidisciplinary approach, that combines plant molecular biology, cutting edge biochemistry, bioinformatics and phytopathology studies.

Since HopBF1-like proteins are distributed among various microbes, human or plant pathogens, as well as free-living organisms, including species able to survive in toxic heavy metal concentrations, elucidation of HopBF1 mode of action may gain novel insight into virulence mechanisms of bacteria but additionally may provide understanding of strategies employed to thrive in extreme environments. Moreover, we expect that characterization of HopBF1 delivers a tool that precisely modifies the proteins involved in host defense response and thus enables tracking of signal transduction pathways, mediated by these proteins.