FINAL PUBLISHABLE SUMMARY REPORT

Microorganisms from the fluorescent pseudomonads group are valuable in agriculture for plant-growth promotion and biocontrol of fungal pathogens and have environmental applications in soil decontamination or bioremediation. The project entitled **"Mining the genome of *Pseudomonas fluorescens* F113 to improve agricultural and bioremediation applications. *In silico* genomics and functional genomics approaches to exploit Pseudomonas in biotechnology (MIGENOF113)"** aims to fully explore the potential of *P. fluorescens* F113 and to maximise its current uses with the following **objectives**:

- To understand F113 genomic characteristics and to identify genes to improve and expand its biotechnological applications by *in silico* analysis of the genome sequence.
- To study the properties conferring ecological fitness by the analysis of the virulence factors that F113 possesses against invertebrate predators, in particular insects.
- To study gene expression and regulation in order to devise in the future engineering of a strain better adapted to the environment and better suited for agricultural and environmental uses.

Main results

The bioinformatic genomic analyses led to the identification of genes potentially involved in the interactions with invertebrates, such as secretion systems. Some of these genes were unique to *P. fluorescens* F113 and not shared by other *Pseudomonas* species. Of particular interest are type 3 secretion systems, molecular syringes found in animal and plant pathogens. Strain F113 possesses a type 3 secretion system homologous to those found in human pathogens such as *Salmonella*. This system in F113, as well as additional type 6 secretion systems, could be important for the interaction with invertebrates. In addition, other genes encoding potential insecticidal toxins have been located in F113 genome.

We proved that *P. fluorescens* F113 was toxic for the larvae of the insect model *Galleria mellonella* (Figure 1). The toxicity did not depend on one single trait but the combination of several of them: production of secondary metabolites, motility…

A functional genomic screen revealed other genomic regions encoding genes responsible for the toxic phenotype, among them VgrG effectors acting through type 6 secretion systems (Figure 2).

Figure 1

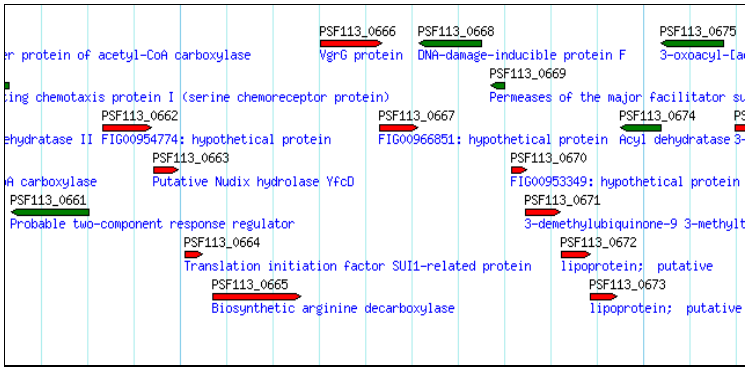

Figure 2

In addition, our experiments showed that strain F113 avoids predation by nematodes and protozoa, and the toxicity towards protozoa is due, in part, to the secretion of secondary metabolites.

Conclusions

-The genome secuence of *Pseudomonas fluorescens* F113 unveiled numerous virulence factors.

-*P. fluorescens* F113 is toxic towards the insect model *Galleria mellonella*, and repels other invertebrates (nematode and protozoa).

-The genes identified reveal that several traits are involved in toxicity: secondary metabolites, motility and secretion systems.

Impact

The outcome of this project has contributed to a better understanding of a group of bacteria that includes plant beneficial strains as well as human and plant pathogens. The identification of new genes in *P. fluorescens* F113 that contribute to toxicity towards invertebrates is a first step in the possible use of this bacterial strain for pest control. Further experiments with other insect species will be needed to assess toxicity towards specific insect pests for future uses. The ability of F113 to avoid predation by soil invertebrates is a positive indication of its survival capability in field conditions. The results and knowledge generated by this project is useful to devise new biotechnological applications for this strain and to improve current ones. In the long term, the use of microorganisms for crop fertilisation and pest controls will contribute to greener and more efficient agricultural practices.

This project has also contributed to create new collaborative links between Universities in the United Kingdom and Spain, reinforcing the exchange of knowledge, techniques and skills among scientific institutions in the European Union.