

Highlight

Life of microbes that interact with plants

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Microorganisms are key players in many biogeochemical processes and they strongly affect the health of soils and plants in many ways. Microbes are responsible for many soil activities, including decomposing dead plant tissues as saprophytes, but they can also attack living plants and cause plant diseases with great economic losses. On the other hand, some microbes can also protect crop plants against their pathogens and can improve the nutritional status of the plant (Bloemberg and Lugtenberg, 2001). This special issue of *Microbial Biotechnology* addresses the diverse ways in which knowledge of plant-associated microorganisms can be applied to benefit agriculture, industry and the environment. Manipulation of microbial properties and modifications in microbial populations in the plant rhizosphere may lead to new interesting and exciting approaches for disease control and plant growth promotion. Biochemical characterization of the molecular mechanisms involved in plant–microbe interactions is uncovering chemicals and enzymes with a wide range of industrial applications. Finally, plant-associated microorganisms can have ecosystem-level effects on plants and soil, which can be exploited to improve soil quality, enhance carbon sequestration and to achieve bioremediation of pollutants, such as hydrocarbons and toxic metals. Many of the fields, in which knowledge about plant–microbe interactions have been applied, are reviewed in this issue by Wu and co-workers (2009), including applications in biofertilization, plant disease control, production of bioactive compounds, remediation and carbon sequestration.

Because of its enormous economic importance, one aspect of plant–microbe interactions that has been extensively studied in the last 30 years is plant–pathogen interactions. Although most of the studies in this field were initially directed towards a better understanding of the molecular mechanisms of the induction of defence responses in plants, more recently many studies address the mechanisms that pathogens employ to invade and successfully colonize their host plants. Rodríguez-Moreno and colleagues (2009) presented excellent images of the real-time monitoring of disease development in olive knots mediated by *Pseudomonas savastanoi* pv. *savastanoi*. They examined the infection process by bright-field, epifluorescence microscopy and scanning and transmission electron microscopy, illustrating the ultrastructure of pathogen-induced knots for the first time. Among different interesting observations, the authors highlight the release of outer membrane vesicles from the pathogen surface, a phenomenon not described before for bacterial phytopathogens, opening discussion about the possibility of the release of virulence-associated proteins through outer membrane vesicles. As part of their active defence mechanisms, plants synthesize a wide range of antimicrobial compounds, and pathogens have evolved mechanisms to withstand or avoid them. Since the first report by Palumbo and colleagues (1998) about the involvement of efflux pumps in the successful colonization of alfalfa roots by *Agrobacterium tumefaciens*, many other reports have shown the importance of multidrug efflux pumps for successful root colonization and infection. In *Erwinia amylovora*, the causal agent of fire blight, the AcrAB efflux pump has shown to play a key role in resistance to antimicrobial chemicals (Burse *et al.*, 2004). The AcrAB efflux pump belongs to the RND (Resistance Nodulation cell Division) family, which carries tripartite structures that traverse the cellular envelope of Gram-negative bacteria and confer high resistance towards a range of structurally unrelated compounds. In this special issue Al-Karablieh and co-workers (2009) present new insights into the molecular mechanisms involved in AcrAB-mediated resistance to antimicrobials. It has been described that the outer membrane proteins (OMPs) of different efflux pumps can be exchanged between efflux pumps (Gotoh *et al.*, 1998; Yoneyama *et al.*, 1998), but in this article the authors

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demonstrate that the third component of the *E. amylovora* AcrAB efflux system, involved in resistance to phytoalexin and virulence, is dependent on TolC ([OMP]). Previous research has shown that some plants, such as *Berberis*, synthesize efflux pump inhibitors that act in concert with antimicrobials to inhibit microbial growth (Stermitz *et al.*, 2000). Knowledge of the molecular basis of pathogen resistance to antimicrobials, and of plant mechanisms to counteract antimicrobial resistance, can be used to identify inhibitors of efflux systems, such as the AcrAB efflux system, and thereby improving the yield of crops, as well as facilitating the discovery of antimicrobial chemicals that can be used in a clinical setting to inhibit multidrug-resistant human pathogens.

Biological control of pathogens can also be a useful tool to substitute or at least reduce the use of toxic chemicals for pathogen control in agriculture. Prieto and co-workers (2009) have studied in detail the colonization of olive tissues by the pathogen *Verticillium dahliae* and the efficacy of the root endophyte *Pseudomonas fluorescens* PICF7 in suppressing the growth of this pathogen. Fluorescent-tagged microorganisms have been successfully used to perform *in vivo* studies in many areas of research (Ramos *et al.*, 2001). However, although fluorescent proteins have been used to visualize plant colonization by bacteria and fungi, this is one of the first examples in which they have been used to monitor fungus–bacteria interactions in a woody plant. By studying the colonization pattern of both microorganisms the authors conclude that early and localized root surface and root endophytic colonization by the bacteria is needed to impair full progress of the wilting symptoms produced by *V. dahliae*. New detailed insights in the interactions between different microorganisms will allow the scientific community to develop new and better biotechnological strategies in biocontrol of plant pathogens.

In addition to suppressing the growth of pathogenic organisms through direct bacterium–pathogen interactions, many rhizospheric bacteria also promote plant growth through a wide range of different mechanisms (Bloemberg and Lugtenberg, 2001). Nitrogen-fixing bacteria (Dobbelaere *et al.*, 2003) and bacteria able to solubilize phosphates (Naik *et al.*, 2008) improve the nutritional status of the plant; bacteria can also produce hormone-like compounds that stimulate plant growth (Costacurta and Vanderleyden, 1995), and by eliciting systemic induced resistance and systemic acquired resistance they can increase resistance of plants to pathogens (Bakker *et al.*, 2007; van Wees *et al.*, 2008). Less well-studied are the beneficial effects that nitrilase-producing bacteria can have on plants. Nitrilases are enzymes that catalyse the hydrolysis of nitrile compounds to the corresponding carboxylic acids and ammonia, which have been shown to play key roles in the synthesis of plant

hormones and antimicrobial chemicals in plants, and in the detoxification of cyanides, herbicides and plant-derived nitriles. In this issue, Howden and Preston (2009) provide a comprehensive overview of current knowledge of nitrile compounds in plants, and the role of bacterial and plant nitrilases in nitrile metabolism, including recent research showing that increased expression of a β -cyanoalanine-degrading nitrilase results in enhanced root growth (Howden *et al.*, 2009). They discuss how knowledge of the regulation and function of plant and bacterial nitrilases can be used to promote plant growth, enhance bioremediation and to help industrial researchers to identify novel nitrilases for use in chemical synthesis.

For both pathogenic and beneficial plant–microbe interactions, root colonization by microbes is a crucial step towards the successful interaction between plant and microbes. Bacterial motility is a key trait in root colonization by pseudomonads and it has been shown that inoculation of low-motility strains, such as *P. fluorescens* F113 into the rhizosphere, can ‘select’ for mutants with high motility (Martínez-Granero *et al.*, 2006). Navazo and co-workers (2009) have identified three different signalling pathways that repress motility in *P. fluorescens* F113 pointing out the complexity of the signal transduction pathways that regulate motility in this strain. Such complexity might reflect the importance of the different processes in which motility is involved, such as chemotaxis, biofilm formation or root colonization. Finetuning of this trait might eventually differentiate between successful and poor colonizers, and knowledge of motility regulation mechanisms could be exploited to engineer and culture strains used for biocontrol and plant growth promotion such that they have optimal motility at the time of inoculation.

Rhizoremediation is the use of rhizosphere-associated microorganisms to remove toxic chemicals from the environment. Successful establishment of bacteria in the rhizosphere, together with efficient expression of catabolic pathways is fundamental in rhizoremediation processes as detailed in a review article by Segura and co-workers (2009). Plants produce many aromatic compounds that can act as inducers of bacterial catabolic pathways. The differences in secondary plant metabolism between different plants can affect the stimulation of the biodegradative capacities of the rhizobacteria, and the authors discuss current directions for biotechnological research to enhance the effectiveness of rhizoremediation, ranging from selection of the right bacterium–plant combination and the inoculation method to the use of microorganisms that produce surfactants that increase the bioavailability of pollutants, and genetic engineering of bacteria to improve their degradative capabilities.

In a biotechnological journal, like this one, special attention has been paid to the bacteria that made possible one

of the most powerful (although controversial) technologies in agriculture, the so-called *A. tumefaciens*-mediated transformation of plants. This bacterium causes crown gall as a consequence of the insertion into plant DNA of T-DNA coding for enzymes producing plant hormones that cause the uncontrolled proliferation of plant cells called tumours. This capacity to insert parts of its own plasmid DNA (T-DNA) into plant chromosomes has been used for the construction of transgenic plants expressing any gene of interest that was inserted between the left and right borders of the T-DNA. The molecular mechanisms involved in DNA transfer from *A. tumefaciens* to plant chromosomes have been extensively studied since the discovery of this process and, in this special issue, Ream (2009a) reviews and compares the DNA transfer mechanisms used by *A. tumefaciens* with those used by the closely related bacterium *Agrobacterium rhizogenes*. Although *A. rhizogenes* also transfers genes to the plant chromosomes, the transfer mechanisms are different from those used by *A. tumefaciens*, affecting their efficiency in biotechnological applications. Importantly, *A. rhizogenes* uses a DNA binding protein known as GALLS, instead of VirE2, to target T-strands to the plant nucleus. GALLS is structurally and functionally distinct from VirE2, and may interact with different host proteins, which means that it may provide an effective nuclear targeting mechanism in plant species that are recalcitrant to transformation by *A. tumefaciens*. Furthermore, the author speculates that as GALLS is less abundant than VirE2, it may transfer a partially coated T-strand, facilitating homologous recombination of plant DNA with heterologous DNA, and thereby enabling site-specific insertion and mutagenesis, and opening up new avenues for plant research and genetic engineering. The same author (Ream, 2009b), in an editorial article, discusses the utilization of transgenic plants. With an excellent sense of humour, the author considers the pros and cons of the utilization of transgenic crops by detailing several transgenic plant applications, ultimately concluding that the benefits outweigh the risks. Although his point of view is clear throughout the text, this article provides a useful overview of current progress in this area and could help students to form their own opinion about such a controversial topic.

Microbe–plant interactions are complex and are influenced by many factors that contribute to a successful interaction. Although, as demonstrated in this special issue, significant and relevant advances have been achieved in this field, microbe–plant interactions are far from being fully understood. Nevertheless, there is ample evidence to show that a better understanding of plant-associated microorganisms can have substantial benefits for agriculture, industry and the environment, which challenges present and future generations of scientists, to further pursue the goal of understanding the molecular

basis of plant–microbe interactions and of using this knowledge to biotechnologically exploit these interactions.

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