Rhizobacteria for Improvement of Plant Growth and Establishment

George Lazarovits

Pest Management Research Centre, Agriculture and Agri-food Canada, 1391 Sandford St. London, Ontario N5V 4T3, Canada

Jerzy Nowak

Department of Plant Science, Nova Scotia Agricultural College, Truro, N.S. B2N 5E3, Canada

From the microbial point of view, soil is a nutritional desert. In contrast, soil adjacent to roots (the rhizosphere) is relatively nutrient rich because, depending on plant species, age, and environmental conditions, as much as 40% of the photosynthates moving into roots are lost to the soil in the form of soluble exudates, mucilage, shed cells, or cell-wall material (Lynch and Whipps, 1991). It is not surprising, therefore, that the root zone harbors great microbial activity. Populations of bacteria in the rhizosphere are enormous, ranging from 10^{10} to 10^{12} cells per gram of soil. These rhizosphere residents, once considered as passive bystanders of the root environment, are now known to affect plant health, development, and environmental adaptation, both beneficially and detrimentally (Curl, 1982; Hornby, 1990). Biological and metabolic activities of this root-soil interface are among the most critical factors contributing to the support of life on our planet (Smucker, 1993).

Fourteen processes associated with rhizosphere competence have been identified to be under genetic control in root colonizing microorganisms (Lynch, 1990b, 1990c). Of these, seven can be considered as beneficial to plant growth; five as neutral or variable, depending on environmental or physical conditions; and only two (infection and phytotoxicity) as potentially harmful. The beneficial effects of microorganisms, such as *Rhizobium*, *Frankia*, and mycorrhizae, are clearly recognized and these organisms are extensively used as plant inoculants (Okon and Hadar, 1987). However, less well-known associations also illustrate the potential impact of plant-microbe associations on plant stands and plant performance. The association between pasture grasses, such as perennial ryegrass (*Lolium perenne* L.) and tall fescue (*Festuca arundinacea* Schreb.), and endophytic fungi of the *Acremonium* spp. is in some ways an ideal model system for where we wish microbial inoculant technology to be directed. These grasses abound in 12 to 14 million ha of grazing ranges in the United States and New Zealand, and represent the staple food source of millions of sheep and cattle (Siegel et al., 1985). Plants colonized by the endophyte grow taller and faster; have higher levels of resistance to microbial pests, insects, and herbivore foragers; and have much higher tolerance to drought and other environmental stresses (Clay, 1988; Siegel et al., 1985). As a result, infected plants rapidly displace noninfected plants in natural settings. As the fungus is seedborne, even the seeds are protected from predation (Clay, 1988). Unfortunately for livestock managers, the endophyte produces some tremoregenic mycotoxins that, when ingested by grazing animals, cause a disease known as ryegrass staggers or fescue toxicosis. Nevertheless, the fungus has become commercially used for improving the resistance of turf grasses to attack by soil insects as well as for enhancing survival of turf during drought-like conditions.

Although our understanding of how root-associated bacteria influence plant growth and development has advanced greatly in the last decade (Bowen and Rovira, 1991; Glick, 1995; Kapulnik, 1991; Lynch, 1990a; Okon and Hadar, 1987; Rovira, 1990), the work in this field is still rudimentary. In a review of more than 1200 papers dealing with microbial crop protection, Powell and Rhodes (1994) found that the majority of studies involved screening of newly isolated strains against one disease, in one environment, and on one crop. Few went beyond laboratory tests and even fewer described how to deliver and establish effective populations of the agents for the environment that they were destined to inhabit. Biological control in the soil remains an insignificant component of pest management systems (Harman and Lumsden, 1990; Powell and Rhodes, 1994). Despite a level of frustration about the slow pace of development of biological control products, there is an unequivocal belief that rhizosphere management can become an important arsenal in our crop protection strategy. This optimism is derived from both a better understanding of how rhizosphere microorganisms influence plant health and by the decline in approval of chemicals for pest control.

Beneficial effects of rhizosphere bacteria have most often been based on increased plant growth, faster seed germination, and better seedling emergence. As a result, these bacteria are now commonly called plant-growth-promoting rhizobacteria (PGPR) and emergence-promoting rhizobacteria (EPR) (Kloepper et al., 1986, 1988b). Taken at face value, these terms can be misleading. For instance, PGPR that act through biological control enhance germination or growth only in cases where disease pressure exists, but have no impact at disease-free sites. Similarly, bacteria that stimulate growth of one plant species or cultivar may retard the growth of another. Bacteria that enhanced emergence of forest seedlings were found to inhibit their growth (O'Neill et al., 1992). Are these beneficial or detrimental microorganisms? The terminology describing such microorganisms will likely become more exact as we obtain a greater understanding of the modes of action involved in growth promotion.

PGPR can have great impact on plant yields. Kloepper et al. (1989) analyzed the effects on yield of three bacterial inoculants applied to a variety of field crops. Compared to controls, increases in yields as high as 160 % were observed, but reductions, although smaller in magnitude, were also common. Erratic performance of bacterial inoculants is a typical result under field conditions (Schroth and Becker, 1990). This inconsistency should not be surprising, however, *Rhizobia* inoculants, among the most widely used biological agents in agriculture, were similarly substandard in their development. *Rhizobia* performance was greatly improved by selecting strains for increased survival in specific soil types, greater compatibility with crop species or cultivars, superior functioning under diverse climates, improved compatibility and competitiveness with other soil microorganisms and higher nitrogen fixing efficiency. These improvements took 30 to 40 years to achieve and are still ongoing (van Elsas and Heijnen, 1990). Improving PGPR strains may require an even longer time, since we are dealing with more diverse mechanisms and dissimilar populations inhabiting rhizosphere, phyllosphere, and internal plant tissues.

The greatest barrier to the effective use of PGPR is our lack of understanding of the mechanisms of biological interactions in the rhizosphere. PGPR may induce growth promotion directly or indirectly. Direct influences include production of phytohormones, liberation of phosphates and micronutrients, nonsymbiotic nitrogen fixation and stimulation of disease-resistance mechanisms. Indirect effects

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arise from PGPR altering the root environment and ecology (Bowen and Rovira, 1991; Glick, 1995; Hornby, 1990; Kapulnik, 1991; Lynch, 1990a, 1990b, 1990c; Okon and Hadar, 1987), for example, acting as biocontrol agents and reducing diseases, liberation of antibiotic substances that kill noxious bacteria, competition with deleterious agents, metabolism of toxic products, etc. Since the mode of action of a PGPR is rarely known when it is field tested, factors that can modify growth-promoting activity, such as environment, plant genotype, and native microflora, are not generally considered in interpreting the results. Very likely our expectations of such agents are also too high and misplaced. For example, fungicides are expected to improve plant growth only in instances where disease occurs. Even with these chemicals, efficacy is often limited by disease pressure. Microorganisms should be tested with the same expectations. In addition, in most studies, the microorganisms are screened under highly controlled laboratory conditions, and expecting them to perform as well in the field is probably inappropriate (Schroth and Becker, 1990). There have been only a few attempts to develop systematic screening methods that identify bacteria suitable to the rhizosphere of a host plant throughout its life cycle (Nijhuis et al., 1993). Every aspect of development and production of microbial inoculants will likely prove to influence efficacy, as even the growth media used were found to influence the competency of a bacterium for biological control (Dickie and Bell, 1995; Zaady et al., 1993).

There is an enormous volume of literature on the application of bacteria for improvement of plant performance (Whipps and McQuilken, 1993), but few bacteria have been developed as commercial products. The organisms under most scrutiny for potential use in agriculture are bacteria belonging to the genera *Pseudomonas* and *Bacillus* species (Powell and Rhodes, 1994). Fluorescent pseudomonads enhance plant growth and crop yield under greenhouse and field conditions and protect wheat (*Triticum aestivum* L.) plants from soil parasites (Bowen and Rovira, 1991; Cook, 1992; Okon and Hadar, 1987; Thomashow and Weller, 1990). Several mechanisms are likely operating in this growth enhancement, including site competition, synthesis of plant-growth-promoting substances, antagonism through production of antibiotics and siderophores, and, most commonly, biological control (Kloepper et al., 1988b). Cocktails of various *Pseudomonas* strains provided superior protection than a single organism (Thomashow and Weller, 1990). Bacilli are also very attractive as potential inoculants in agriculture, as they produce very hardy spores that can survive for prolonged periods in soil and in storage containers. Various formulations of *B. cereus* increased soybean (*Glycine max* L. Merr) yields at two Wisconsin sites over a 5-year period in the presence and absence of root-rot caused by *Phytophthora sojae* Kaufmann & Gerdemann (Osburn et al., 1995).

There is a good potential for exploiting bacterial inoculants for Canadian climates. The most consistent benefits of PGPR, seen as enhanced seed emergence and plant performance, have been obtained during cool, wet springs and short growing seasons and when plants are placed under stress (Kloepper et al., 1986; Okon and Hadar, 1987; Schroth and Becker, 1990). Reducing transplant shock for high-value crops, particularly those that are transplanted, also became one of our research objectives (Nowak et al., 1995). Numerous studies have incorporated microbial inoculants into soilless mixes as a means of delivering the microorganisms to the plant roots (Whipps and McQuilken, 1993). This practice is considered to have the best potential for establishing introduced organisms in the rhizosphere since under greenhouse conditions physical, environmental, and microbial factors are more readily controlled (Okon and Hadar, 1987; Schroth and Becker, 1990).

Our search targeted organisms that directly stimulated plant growth. To improve the chances for establishment we looked for bacteria that could colonize the rhizosphere and internal vascular tissues. There is mounting evidence that the vascular system of plants is extensively colonized by bacteria (endophytes) and that these organisms form populations distinct from those that colonize the rhizosphere (Bell et al., 1995; Chen et al., 1995; Fahey et al., 1991; van Peer et al., 1990). Endophytic bacteria have been reported to have beneficial and detrimental effects on red clover (*Trifolium pratense* L.) (Sturz and Christie, 1995). Whether the bacteria promoted or inhibited growth

depended on the cropping history or prior exposure of the plants to the natural microflora in the peat-based medium (Sturz and Christie, 1995). Utilization of endophytes, normally considered contaminants by those involved in plant tissue culture (Leifert et al., 1994), may become an integral part of mass clonal multiplication of plants such as potato (*Solanum tuberosum* L.).

There is a need for simple model systems that permit testing of plant-rhizosphere microbe interactions. In light of the numerous and highly complex interactions that may be involved in plant growth promotion, surprisingly few attempts have been made to determine the effect of PGPR on plants under sterile conditions. Several investigators have used test tube assays that included a soilborne pathogen in sterile soil or soil-like media for assessing biological control and growth promotion (Défago et al., 1990; Kloepper et al., 1988a, 1991; Voisard et al., 1994). Lifshitz et al. (1987) found that a growth pouch assay allowed them to identify growth-promoting strains of bacteria on canola (*Brassica campestris* L.) in the absence of either pathogens or deleterious microorganisms, and this assay has become widely used. Our assay procedure relied on treating potato nodal explants with the test bacteria under tissue-culture conditions. The nodal cuttings were dipped into a bacterial suspension, blotted dry, and inserted into agar medium. Most bacteria do not grow to any extent on the culture medium itself, but many grow readily adjacent to the plant tissues. The assay is fairly rapid and simple, but not universally useful for all bacteria. The presence of sucrose in the culture medium allows some genera to overgrow the nodal explant and kill it. We have screened many bacteria strains, many of which were previously identified as growth promoters by other assay procedures (Kloepper et al., 1986, 1991). Using the nodal explant assay only, a few strains were found to benefit potato. Most bacteria, particularly of the *Serratia* group, in fact inhibited growth. An autotrophic culture system presently being investigated may be more suitable for studying in vitro interactions between plants, pathogens, and beneficial bacteria.

Potato and other vegetable nodal explants in dual cultures with a nonfluorescent *Pseudomonas* spp. (PsJN) isolated from sterilized onion (*Allium cepa* L.) roots showed significant growth stimulation under sterile tissue-culture conditions (Fig. 1A) and during early growth after transplanting (Fig. 1B) (Nowak et al., 1995). Inoculated plantlets were taller, had more nodes, higher dry matter content, better developed root systems (with more branching and more root hairs), more leaf hairs, increased amounts of chlorophyll and starch, and were more lignified (Frommel et al., 1991). Noninoculated plantlets desiccated rapidly when removed from tissue-culture conditions, whereas bacterized plants remained turgid because they had functional stomata and could regulate water loss (Frommel et al., 1991). Survival of bacterized potato plantlets transplanted directly from tissue-culture conditions to the field was higher than that of nonbacterized controls (Table 1). Thus, co-culturing of potato nodal explants with PsJN could eliminate the 2- to 3-week-long greenhouse hardening step presently used in some pre-elite seed potato production systems. Under tissue-culture conditions, bacteria multiply inside the vascular bundles of stems and roots and are transferred with cuttings taken from plantlets (in perpetuity), making the system ideal for mass multiplication via nodal explant.

PsJN also promotes growth and development of potatoes under greenhouse and field conditions. Earlier stolon formation and tuber set and production of more tubers were observed in growth chamber (Table 2), greenhouse (Nowak et al., 1995), and field (Frommel et al., 1993) experiments. Yield increases were also obtained in some trials conducted in Ontario and Nova Scotia where the bacterium was applied to cut or whole seed tubers as a spray or dip (Table 3). As with other studies with PGPR, the extent of the response was cultivar-dependent and varied from site to site.

A factor that promises to favor use of PsJN as PGPR inoculant is its ability to colonize the internal root structures (endorrhizosphere) and vascular system in shoots (endophyllosphere), where competition is less intense and environmental factors more consistent than in the exorhizosphere. Increasing the establishment and survival of inoculants by novel application techniques, for example, seed priming (Harman and Taylor, 1988; Whipps and McQuilken, 1993) or encapsulation (van Elsas and Heijnen, 1990), is required for improving

Fig. 1. Bacterization effect (*Pseudomonas* sp., strain PsJN) on in vitro growth of potato plantlets [(A) 4-week-old plantlets cv. Red Pontiac; nodal explant inoculated as described by Frommel et al. (1991)] and greenhouse grown sweet pepper transplants [(B) 7-day-old seedlings (obtained from surface sterilized seeds) bacterized by root dip in the bacterium suspension of $\approx 2.5 \times 10^8$ cfu/mL, grown in vitro for 3 weeks, followed by 5 weeks in cell packs (in ProMix) in the greenhouse].

Table 1. Effect of bacterization on survival of potato plantlets cv. Kennebec.^z

Location	Year	Plant survival (%)	
		Control	Bacterized
Nova Scotia	1987	59.9	98.5**
	1990	64.2	98.3**
London	1990	71.0	97.0*
Alliston	1989	32.5	73.8**
	1990	35.5	93.0**
New Liskeard	1990	15.8	78.5**

^zThe plantlets were directly transplanted from tissue culture vessels to the field.

* **Significant difference in the mean between the control and bacterized treatment, respectively, at $P \leq 0.05$ and 0.01 by Duncan's multiple range test.

Table 2. Bacterization effect on tuberization responses in two potato cultivars.^z

Cultivar	Bacterization ^y	Stolon dry wt (mg)		Tubers (6 wk)	
		4 wk	6 wk	#	Dry wt (g)
Kennebec	—	11	443	1	0.04
	+	55*	574*	3*	0.78**
Shepody	—	29	82	2	0.30
	+	43*	418**	3	1.38**

^zFour-week-old bacterized and nonbacterized tissue-culture plantlets transplanted to a peat-based growth medium (promix; ASB Greenworld, Point Spain, N.B., Canada) in pots (10 cm top $\times$ 7 cm bottom $\times$ 9 cm height), one plant per pot, and grown in growth chamber under 16-h photoperiod and 22/17 °C day/night temperature;

^yExplant inoculation as described by Frommel et al. (1991).

* **Significant difference from the mean (six plants per treatment) at $P \leq 0.5$ and 0.01 , respectively, by Duncan's multiple range test.

Table 3. Effect of seed piece bacterization on weight and size of potato tubers cv. Kennebec.

Year	Treatments	Kg/plot		Tubers/plot	
		<55 mm	>55 mm	<55 mm	>55 mm
1989 ^z	WS ^y control	1.1 a	6.9 b	23.4 a	41.0 b
	Bacterized	0.6 a	11.9 a	18.6 a	60.2 a
	CS ^x control	0.7 a	5.9 b	21.6 a	33.0 b
	Bacterized	0.9 a	6.6 b	18.2 a	37.2 b
1990 ^w	WS control	2.8 b	42.9 b	70.0 ab	171.4 a
	Bacterized	2.5 b	51.5 a	56.0 bc	188.4 a
	CS control	2.2 b	35.9 b	48.0 c	141.6 a
	Bacterized	3.4 a	42.3 a	89.2 a	162.8 a

^zGrowth cycle 119 days, n: 20; blocks 5; cv: 17.5% and 12.8% for weight and number of tubers, respectively.

^yWhole seed.

^xCut seed.

^wGrowth cycle 133 days, n: 31; blocks 5; cv: 13.9% and 7% for weight and number of tubers, respectively.

Mean separation within a column for a specific year followed by the same letters do not differ significantly at $P \leq 0.05$ by Duncan's multiple range test.

inoculant performance (Kumar and Dube, 1992; Schroth and Becker, 1990). Multi-strain inoculants can also be applied either as co-inoculants or sequentially.

Currently, we are investigating the regulation of growth promotion by PsJN using molecular biology techniques. Mutants were created by random insertion of the Tn5 plasmid into the bacterial genome. Mutated bacteria were identified by resistance to antibiotics conferred by the Tn5 plasmid. The mutants were then screened for loss of growth promotion activity. Several such mutants were identified and tested to ensure that they were capable of colonizing the plants in tissue culture. The DNA region associated with the Tn5 plasmid will be characterized and tested for its capacity to restore growth promotion. Once the smallest region of DNA that can restore growth-regulating activity is identified, then the functioning of the genes may be more readily manipulated. Although PsJN promotes growth of potatoes, tomatoes (*Lycopersicon esculentum* Mill.), peppers (*Capsicum annuum* L.), and other vegetables (Nowak et al., 1995), it is clearly cultivar specific. In our laboratories, under tissue-culture conditions, bacterial treatment increased the dry weight of roots of the potato cultivars Norchip by up to 600% to 1000% and Kennebec by 200% to 400%, whereas it inhibited the root weight of 'Chaleur' by 40%. The interactions between PsJN, potato cultivars, and environmental conditions are turning out to be more important than we imagined. When bacterized plantlets and nontreated plantlets (controls) of 'Kennebec', 'Shepody', and 'Norchip' were grown at 10, 20, and 25 °C, for 3 weeks, the benefits of bacterization were the most evident at 25 °C, where 'Kennebec' showed a 70% shoot growth stimulation over control, followed by 'Norchip' (30%), and 'Shepody' (11%). The differences were less pronounced at 20 °C and at 10 °C. Compared to controls, at these temperatures, bacterial inoculation inhibited shoot growth of 'Shepody' and 'Norchip' (46% and 23%, respectively), but there was no effect on 'Kennebec'. At 10 °C, roots were formed on bacterized plantlets of 'Norchip' and 'Kennebec', but not on 'Shepody'; none of the control plants had roots. At week 6, the best shoot-growth stimulation caused by the bacterium was recorded in 'Norchip' at 25 °C, followed by 'Shepody' at 20 °C, 'Norchip' at 20 °C, and 'Kennebec' at 25 °C. At 10 °C the bacterium inhibited growth of 'Norchip' and 'Shepody' plantlets by $\approx 50\%$ and stimulated growth of 'Kennebec' plantlets by 38% (Nowak et al., unpublished). The responses were not related to the extent of bacteria colonization of the plantlets. How this applies to field performance of PsJN is unknown, but it clearly indicates that environment is a very important component of bacterial performance. Plant genotype-specific responses to bacteria inoculants have also been found by others (Arsac et al., 1990; Becker et al., 1990; Chanway and Holl, 1994a). Quantitative differences in phytohormone production by bacteria and the degree of sensitivity of plants to phytohormones are being suggested as the main reasons for this phenomenon (Glick, 1995). The complexity of the plant genotype-bacterium-environment interactions creates a real challenge for the development of inoculants as a new agricultural practice. Good understanding of the interaction is also crucial for plant breeders. An

interesting dual-culture system of free-living nitrogen-fixing *Azotobacter* species and plants has been developed by Varga et al. (1994). The system allows establishment of artificial symbiosis, sustainable with respect to the nitrogen supply. The bacteria colonize the entire plant and in many cases are also transmitted through seeds (Sz.S. Varga, personal communication). Given the appropriate organism and strategy for establishing it in plants, rhizosphere, or both, PGPR can have tremendous impact on the way we will grow crops in the 21st century.

An understanding of the mechanism of action can be exploited to enhance the beneficial effects of PGPRs. For example, low amounts of phytohormone precursors added to a soil containing PGPR stimulated plant growth to a greater extent than that obtained in soil without the precursor or without PGPR (Arshad and Frankenberger, 1991; Nieto and Frankenberger, 1989, 1990). In our experiments with the pseudomonad strain PsJN, bacterization of in vitro-grown potato plantlets increased their content of zeatin, dihydrozeatin, and dihydrozeatin ribose. The bacteria also facilitated movement of these cytokinins from a growth medium amended with seaweed extract to shoots of potted plants (Table 4). When the bacteria were absent, cytokinin levels in shoots of plants grown in media alone or media amended with seaweed extract were virtually identical, but bacterized plants showed a 60% increase in cytokinin content. The concentrated seaweed extract contained only traces ($\mu\text{g}\cdot\text{mL}^{-1}$) of zeatine: 3.6 of dehydrozeatin and 0.11 of dehydrozeatin ribose. No cytokinins were produced by the bacterial cultures alone. This simple experiment demonstrates that PGPR can induce cytokinin synthesis in plants in early stages of growth and development and can facilitate uptake and/or translocation of these growth regulators in plants with fully developed roots. It also suggests that to take full advantage of PGPR we may need to include soil amendments that promote growth promotion efficacy. For example, strains of *Enterobacter cloacae* (Jordan) Hormaeche and Edwards, appear to protect plants from Pythium damping off by ammonia production (Howell et al., 1988) but the presence of certain sugars suppressed ammonia production. These organisms thus may be more effective if applied in conjunction with specific organic materials formulated into seed treatment or incorporated into transplant plugs.

One of the more interesting mechanisms of disease suppression of both bacterial and fungal rhizosphere agents may be through activation of the plants defense mechanisms (Alström, 1991; Chen et al., 1995; Duijff et al., 1993; Maurhofer et al., 1994; Meera et al., 1994; van Peer et al., 1991; Wei et al., 1991; Zdor and Anderson, 1992). This form of disease suppression is of particular interest because it promises control of not only soilborne pathogens, but also of a wide spectrum of foliar pests and parasites. There is still some question as to whether translocation of biocidal agents from the roots to the other plant tissues, which results in protection through a chemical control rather than by induced resistance, may be occurring (Alström, 1991).

Rhizosphere bacteria and fungi are an untapped source of antifungal, antibacterial, antiinsect, and antinematode agents. Lievens et al. (1989) screened more than 11,614 bacterial isolates from roots and leaves of various crops and found that 3910 produced antifungal metabolites that inhibited a specific pathogen of the crop in question. Ultimately, we envision that bacterial agents, such as PsJN, will carry genes that regulate not only plant growth, but also produce natural insecticidal and fungicidal compounds. Crop Genetics Intl. of Hanover, Md., has already used *Clavibacter xylii*, an endophytic bacterium, to deliver *Bacillus thuringiensis* toxin into corn (*Zea mays* L.) for control of corn borer (InCide) (Fahey et al., 1991).

Bacterization only marginally increased yields when tested under ideal climatic situations. The greatest benefits occurred when crops encountered stressful conditions for prolonged periods. Lodgepole pine seedlings treated with PGPR benefitted from bacterization only at the poorest site used for transplanting (Chanway and Holl, 1994b). Although inoculation of peanuts (*Arachis* L.) with *Bacillus subtilis* improved germination and emergence, increased nodulation by *Rhizobium*, enhanced plant nutrition, reduced disease effects, and increased root growth, the greatest yield responses were obtained when the peanuts were stressed by drought, cool soils, or some or other detrimental factor (Turner and Backman, 1991). Nontreated plants by

Table 4. Bacterization effect on cytokinin content (fresh weight basis) in 4-week-old 'Kennebec' plantlets grown in vitro (A) and in shoots and roots of transplants grown in the greenhouse on ASB medium amended and not amended with seaweed extract (B).

Treatment ^a	Cytokinin content ($\text{ng}\cdot\text{g}^{-1}$ fresh wt) ^b					Total
	Seasol ^c	Z	DZ	ZR	DZR	
A. Control	---	0	0	489	38	527
Bacterized	---	5	185	379	156	725
B. Control						
Shoots	---	246	21	342	31	640
Roots	---	294	0	5	0	300
Shoots	+	139	54	431	1	625
Roots	+	250	0	0	0	250
Bacterized						
Shoots	---	179	32	339	6	557
Roots	---	171	0	4	0	175
Shoots	+	294	191	383	37	905
Roots	+	45	0	0	0	45

^aFour-week-old plantlets transplanted to 14-cm pots and grown in the greenhouse for 4 weeks.

^bZeatin (Z), dihydrozeatin (DZ), Zeatin ribose (ZR), dihydrozeatin ribose (DZR); 2-g samples were used for analysis, extraction was done according to Guinn and Brummett (1990) and HPLC analysis according to Horgan and Kramers (1979).

^cKelp (*Durvillea potatorum*) extract (DM AG Co., Honolulu, Hawaii); 100 mL diluted in 4 L of water and mixed with 21 L of peat-based promix medium (ASB Greenworld, Point Spain, N.B., Canada).

comparison perform poorly under such conditions. Bacterial activity thus appears to lessen stress effects while maintaining productivity. In light of the fact that bacterial inoculants are inexpensive to produce, are environmentally rational, and can be readily incorporated into many current production practices, we envision that these organisms are destined to become an important component of crop production. Before this occurs, however, we will have to gain a much better understanding of the intricate associations that have evolved between plants and their rhizosphere residents.

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