APPROACHES FOR ENHANCING PGPR-MEDIATED ISR ON VARIOUS VEGETABLE TRANSPLANT PLUGS

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Abstract

Plant growth-promoting rhizobacteria (PGPR) have been shown to increase plant growth of a number of agronomically important crops, and some PGPR strains have induced systemic resistance (ISR) against multiple pathogens. We have been investigating use of a biopreparation consisting of combinations of PGPR with ISR activity and the organic amendment chitosan in peat-based plant growth media for production of vegetable transplants. These biopreparations were designed to enhance seedling growth and develop disease-suppressive transplant plugs through activation of ISR. Several biopreparations (LS treatments) were tested, with each containing industrially formulated spores of two *Bacillus* strains and chitosan. Greenhouse experiments indicated that most of the LS treatments significantly enhanced growth and reduced disease severity, compared to controls, in the following systems: tomato against bacterial spot (*Xanthomonas axonopodis* pv. vesicatoria) and late blight (*Phytophthora infestans*), cucumber against angular leaf spot (*Pseudomonas syringae* pv. lachrymans) and tobacco against blue mold (*Peronospora tabacina*). In field trials conducted at an experimental farm in Florida, several LS treatments significantly reduced root-knot nematode damage on cucumber and tomato, and the treated plants exhibited more root mass at mid-season in these field trials. The same LS treatments, which reduced root-knot nematode damage, also induced reduction in severity of naturally occurring cucumber anthracnose and of inoculated bacterial spot on tomato. Subsequent field trials demonstrated that a mid-season booster application of PGPR, applied either by foliar spray or by injection into the drip irrigation system after the initial treatment of transplants, enhanced PGPR-mediated ISR activity compared to no booster application. Some LS treatments significantly improved the quality and quantity of fruit of tomato, pepper and cucumber.

Introduction

Plant growth-promoting rhizobacteria (PGPR) have been shown to increase plant growth of a number of agronomically important crops, and some PGPR strains have induced systemic resistance (ISR) against multiple pathogens, including fungi, bacteria, viruses, and, in a very few cases, nematodes (Kenney et al., 1999; Kloepper et al., 1999; Murphy et al., 2000., Reddy et al., 1999; Ryu et al., 1999; Shouan et al., 1999; Yan et al., 1999). An alternative approach for control of nematodes is the use of specific organic amendments, such as chitin and chitosan. We have been investigating the combination of PGPR-mediated ISR with chitosan amendment of peat-based plant growth media for production of vegetable transplants. The combination of PGPR with chitosan is designed to enhance seedling growth and develop disease-suppressive transplant plugs through the enhancement of PGPR-mediated ISR.

The purpose of this study was to accelerate development of vegetable transplant plugs and increase plant growth and health. More specifically to increase the rate of seedling growth thereby decreasing the time required to produce transplants in commercial greenhouse prior to transplanting into fields and to develop disease suppressive transplant plugs, which are protected for a time from multiple diseases.

Materials and Methods

The effects of several biological preparations (LS series, Table 1) on plant growth-promotion, ISR against various foliar pathogens, nematode control and fruit yield on various vegetable transplant plugs were evaluated under greenhouse and field conditions. Each biological preparation contained industrially formulated spores of two PGPR strains *Bacillus subtilis* strain GBO3 plus one additional bacilli and a flaked chitosan. A series of greenhouse experiments were conducted to test the efficacy of these preparations added to transplanting potting media for growth promotion effects and also ISR against foliar pathogens using the following pathosystems: tomato bacterial spot (*Xanthomonas axonopodis* pv. vesicatoria) and late blight (*Phytophthora infestans*), cucumber angular leaf spot (*Pseudomonas syringae* pv. lachrymans), and tobacco blue mold (*Peronospora tabacina*).

Table 1. List of biological preparations.

Treatment	Strain #	Strain identification
LS213	GBO3 + GB99 (IN937a)	*Bacillus amyloliquefaciens*
LS254	GBO3 + GB87 (SE34)	*Bacillus pumilus*
LS255	GBO3 + GB88 (IN937b)	*Bacillus subtilis*
LS256	GBO3 + GB34 (INR-7)	*Bacillus pumilus*
LS257	GBO3 + GB35 (T4)	*Bacillus pumilus*
LS258	GBO3 + GB52 (1PC11)	*Brevibacillus macerans*
LS259	GBO3 + GB47 (1PN19)	*Bacillus subtilis*
LS260	GBO3 + GB49 (3P114)	*Paenibacillus macerans*
LS261	GBO3 + GB64 (C4)	*Bacillus cereus*

Field experiments were conducted during 1998 and 1999 to evaluate several of these LS preparations as potting mix amendments at seeding, or at seeding plus a mid-season “booster” application for control of root-knot nematode, anthracnose on cucumber, and bacterial spot of tomato. Further, the effects of biological treatments on growth and yield of tomato, cucumber and pepper were evaluated in the field naturally infested with root-knot nematode and foliar

pathogens. Four-week-old transplants were planted on raised beds in the field. Each treatment was replicated six times and arranged in a randomized complete block design. Seedling growth parameters were assayed at 0, 30 and 60 days after transplanting. Fruit yields were harvested at several times during the growing season. The data were subjected to standard analysis of variance procedure.

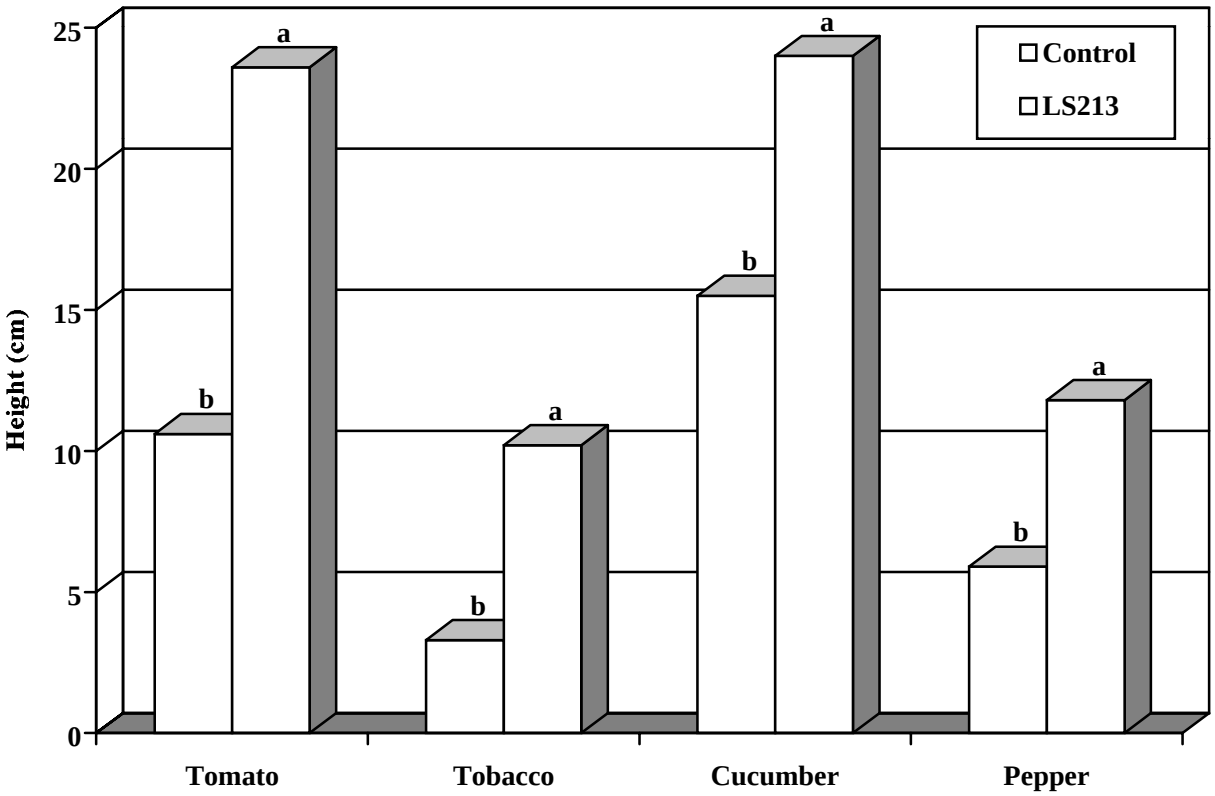


Fig. 1. Effect of LS213 on seedling shoot height of various vegetable transplant plugs 4 weeks after seeding. Mean values with different letters are significantly different at P = 0.05.

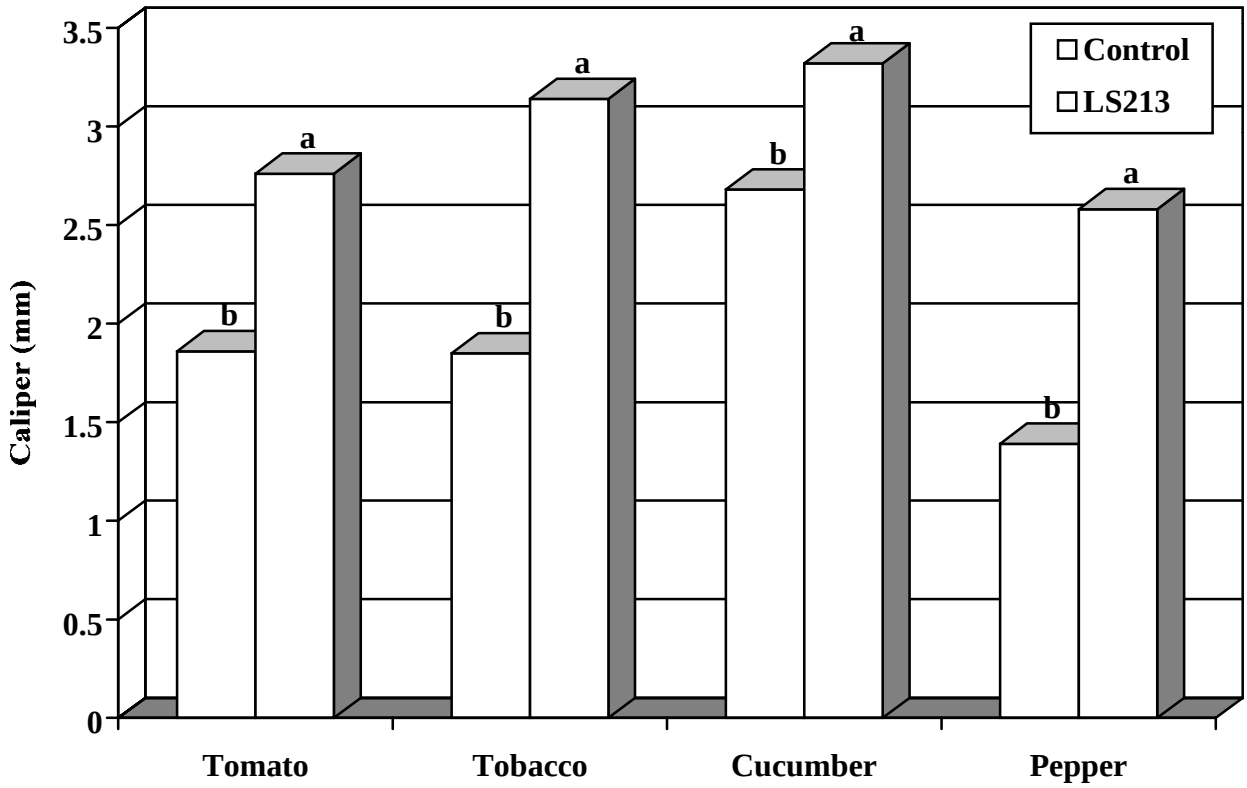


Fig. 2. Effect of LS213 on seedlings stem caliper of various vegetable transplant plugs 4 weeks after seeding. Mean values with different letters are significantly different at P = 0.05.

Results

Our greenhouse results showed that most of the LS series, particularly LS213, significantly increased seedling growth of all crops (measured by height, caliper, root and shoot fresh weight) compared to a untreated control (Figs. 1& 2). Most of the LS series, including LS213, provided significant levels of disease suppression for all diseases evaluated (data not shown here).

Table 2. Spectrum of disease protection by biological preparations under field conditions during 1998 at Sanford, Florida.

Treatment	Tomato root-knot[1]	Tomato bacterial spot[1]	Cucumber root-knot[1]	Cucumber anthracnose[1]
LS213	+	+	+	+
LS254	+	-	+	+
LS255	+	-	-	+
LS256	-	-	-	-
LS257	-	+	-	+
LS258	-	+	+	+
LS259	-	+	+	-
LS260	-	-	+	+
LS261	+	+	+	-

[1]Mean of 3 replications, 3-5 plants per replication. + = Significantly different from nontreated control at P = 0.05, and - = not significantly different from nontreated control.

During 1998, in Florida field trials, several of the LS treatments significantly reduced root-knot nematode damage on cucumber and tomato and also gave protection against cucumber anthracnose and bacterial spot on tomato (Table 2). Subsequent field trials demonstrated that a mid-season booster application of PGPR, after the initial treatment of transplants, enhanced PGPR-mediated ISR activity compared to no booster application (Tables 3 & 4).

During 1999, in Alabama field trials, significantly greater seedling growth occurred with all biological treatments compared to a nontreated control at 0 and 30 days after transplanting and with some treatments at 60 days after transplanting. Some of the LS treatments significantly improved the total marketable fruit of tomato (Fig. 3), pepper (Fig. 4) and cucumber (data not shown).

Table 3. Effects of biological preparations and delivery system on cucumber anthracnose under field conditions during 1998 at Sanford, Florida.

Treatment	In greenhouse mix[1]	Foliar spray[1]	In mix and foliar spray[1]
LS213	27	30	17*
LS254	27	20*	27*
LS255	20	20*	17*
LS256	47	27*	37
LS257	17*	20*	27*
LS258	37	30	27*
LS259	43	30	37
LS260	13*	20*	17*
LS261	40	27*	40
Nontreated control	43	50	50
Number significant	2	6	6

[1]Mean of 3 replications, 3-5 plants per replication. Anthracnose severity was assessed on a visual scale of 0-100% per replication, 0 = no lesions.
*Significantly different from nontreated control at P = 0.05.

Table 4. Effects of biological preparations and delivery system on root-knot nematode severity of tomato under field conditions during 1998 at Sanford, Florida.

Treatment	In greenhouse mix[1]	Foliar spray[1]	In mix and foliar spray[1]
LS213	4.5	4.2*	3.5*
LS254	3.8*	3.9*	2.8*
LS255	4.0*	3.0*	3.7*
LS256	4.2	6.6	4.6
LS257	3.9*	4.7*	3.4*
LS258	5.3	5.7	5.1
LS259	5.4	5.4	3.6*
LS260	4.5	5.2	2.9*
LS261	5.8	3.9*	3.5*
Nontreated control	5.8	5.9	4.6
Number significant	3	4	7

[1]Mean of 3 replications, 3-5 plants per replication. Root-knot severity measured on a 0-10 scale, 0 = no galls and 10 = 100% galls. *Significantly different from nontreated control at P = 0.05.

Fig. 3. Effect of biological preparations on tomato yield under field conditions during 1999 at Sand Mountain, Alabama. Asterisk indicates significant difference from control at P = 0.05.

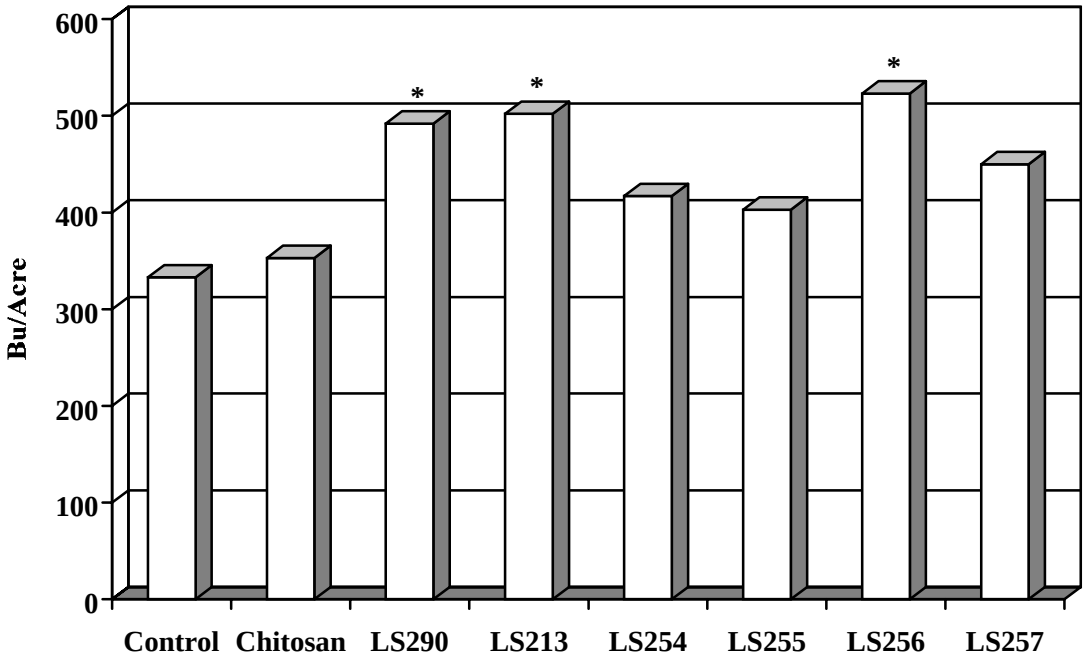
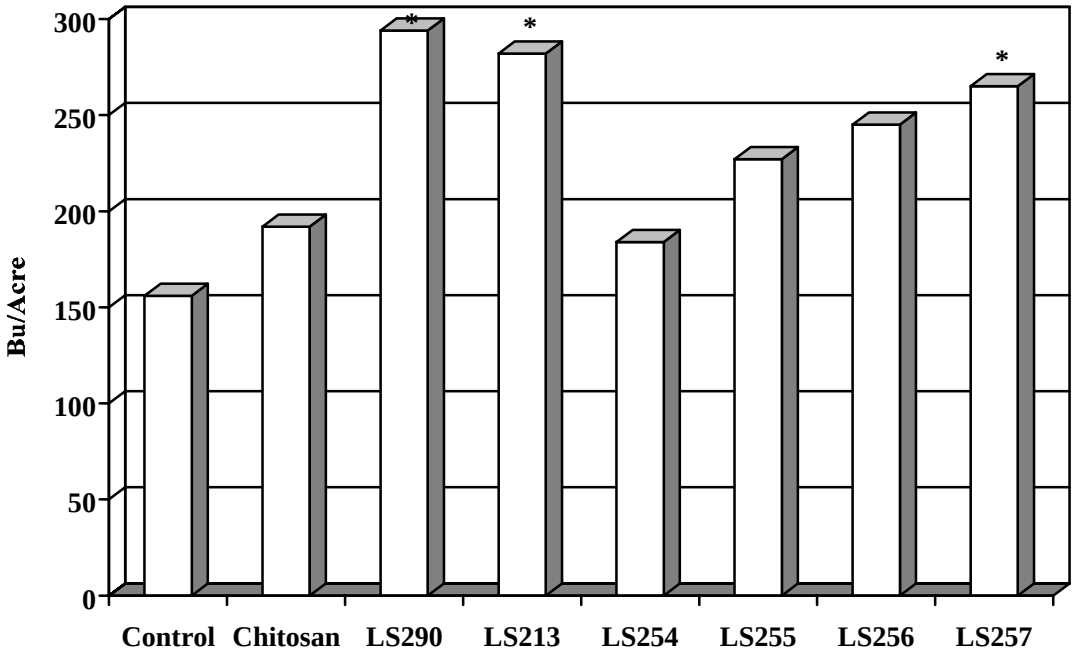


Fig. 4. Effect of biological preparations on pepper yield under field conditions during 1999 at Sand Mountain, Alabama. Asterisk indicates significant difference from control at P = 0.05



Discussion

Results presented here demonstrate that mixtures of two PGPR strains plus a formulation carrier chitosan, particularly LS213 exhibited a more consistent and significant level of growth promotion on all the crops tested. The transplant mix delivery system for the biological preparations has demonstrated potential to enhance transplant vigor, provide protection against root-knot nematodes and other foliar diseases in the field and enhance yield. Our studies suggest that enhanced efficacy of plant growth promotion and biological control may be achieved when supplementary applications of PGPR are applied in an aqueous formulation either as drench or as foliar spray. These results indicate a synergy in plant growth-promotion and ISR activity by the combination of chitosan and the mixtures of two bacterial strains.

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