Investigating the Effects of *Bacillus* Endophytes on Plant Growth

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Abstract

Synthetic chemicals such as pesticides and fertilizers are used in agriculture to enhance plant growth and productivity but often have detrimental impacts on the environment and human health. Endophytes, the beneficial microbes colonizing the internal tissues of plants, which have been shown to promote plant growth directly by mobilizing nutrients or indirectly by production of phytohormones and suppression of pathogens, offer a potential alternative. The objective of this study was to identify endophytes in the genus *Bacillus* that can promote plant growth. Four *Bacillus* strains were evaluated for their effects on growth of *Brassica rapa* (field mustard). Seeds were soaked in bacterial broth prior to planting in sterile soil. Plants were evaluated for root and stem length as well as wet and dry weight. While no statistically significant differences were evident between strains, this research provided useful insights for future studies. By identifying endophytes that can enhance growth of crop plants, sustainable agricultural practices can be promoted.

Introduction

- The use of fertilizers and pesticides may have negative consequences including algal blooms, greenhouse gas emissions, and reduction of soil fertility (1, 2).
- Endophytes (microbes living within healthy plant tissues) in the genus *Bacillus* are promising candidates for use as biofertilizers and bio-control agents (3).
- Several mechanisms are used by these bacteria to promote plant growth (Figure 1).

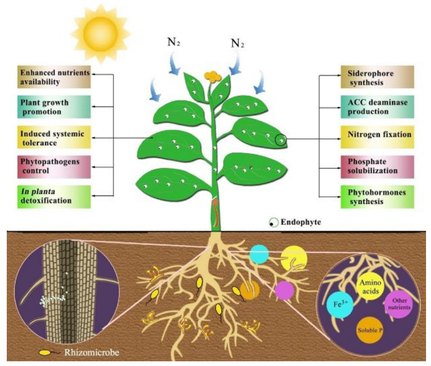

Figure 1. Plant growth-promotion mechanisms used by endophytes (4).

- The goal of this work is to identify endophytes in the genus *Bacillus* that promote plant growth. Future studies will further evaluate these species for activity against plant pathogens as well as effects on nutrient mobilization such as nitrogen fixation.

Materials and Methods

- The *Bacillus* species investigated in this study include *B. thuringiensis*, *B. atrophaeus*, *B. pumilus* (American Type Culture Collection strain #700814 – this strain does not produce an antibacterial agent), and *B. pumilus* (environmental isolate – this strain is known to produce an antibacterial compound).
- Seeds of *Brassica rapa* were surface sterilized and soaked in each of the four bacterial broths prior to planting in sterile soil. Control, untreated seeds were also planted.
- Ten four-inch pots containing three seeds each were used for each group.
- Plants were watered daily, and insect repellants and fertilizers were not used. After 6 weeks, the plants were evaluated for growth characteristics including:
  - total wet length
  - wet and dry weight
- Analysis of variance (ANOVA) was conducted to compare bacterial treatments.

Results

While plants treated with *B. thuringiensis* did demonstrate greater length and higher weights compared with other treatments or controls, these differences were not statistically significant.

Table 1. Average plant growth characteristics. The power of ANOVA was 0.284 for wet weight and 0.05 for wet length.

Treatment	Wet weight (g)	Dry weight (g)	Wet length (cm)
B. atrophaeus	0.80	0.14	14.9
B. thuringiensis	1.77	0.31	17.95
B. pumilus ATCC	1.04	0.18	16.2
B. pumilus isolate	1.48	0.23	15.85
Control	1.12	0.17	15.68

Figure 2. Control treatment. Seeds were not exposed to any endophytes prior to planting.

Results

Figure 3. *Bacillus thuringiensis* treatment. This treatment had the largest and healthiest looking plants.

Figure 4. *Bacillus atrophaeus* treatment. This treatment had the lowest survival rate (four of ten plants) and was smaller than the other treatments.

Figure 5. *Bacillus pumilus* ATCC treatment.

Figure 6. *Bacillus pumilus* isolate treatment.

Discussion

- No statistically significant differences were found between treatment groups for wet weight or length. This may be attributed to the small sample sizes and high variability in the data.
- Visual differences were observed, however, for plants inoculated with *B. atrophaeus* and *B. thuringiensis*. *Bacillus atrophaeus* treatment group had the lowest survival of plants, and they were smaller with less flowering. Endophytes are often host specific, promoting growth in one plant but not another. This may be the case with *B. atrophaeus* and *Brassica rapa*. In contrast, all plants in the *B. thuringiensis* treatment group were larger and looked healthier than the other treatments.
- *Bacillus thuringiensis* has been well-documented for "biological control" by causing the death of harmful insects, although no insect pests caused damage to the plants in this study.
- Future studies will use *in vitro* microbiological techniques to assess possible mechanisms used by *B. atrophaeus* and *B. thuringiensis* to affect plant growth. The sample size for the greenhouse experiment will be larger and dry above-ground biomass will be used for statistics.

Literature Cited

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