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Response of strawberry to inoculation with arbuscular mycorrhizal fungi under very high soil phosphorus conditions

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Abstract A field study was done to assess the potential benefit of arbuscular mycorrhizal (AM) inoculation of elite strawberry plants on plant multiplication, under typical strawberry nursery conditions and, in particular, high soil P fertility (Mehlich-3 extractible P=498 mg kg⁻¹). Commercially in vitro propagated elite plants of five cultivars ('Chambly,' 'Glooscap,' 'Joliette,' 'Kent,' and 'Sweet Charlie') were transplanted in noninoculated growth substrate or in substrate inoculated with Glomus intraradices or with a mixture of species (G. intraradices, Glomus *mosseae*, and *Glomus etunicatum*) at the acclimation stage and were grown for 6 weeks before transplantation in the field. We found that AM fungi can impact on plant productivity in a soil classified as excessively rich in P. Inoculated mother plants produced about 25% fewer daughter plants than the control in Chambly (P=0.03), and Glooscap produced about 50% more (P=0.008) daughter plants when inoculated with G. intraradices, while the productivity of other cultivars was not significantly decreased. Daughter plant shoot mass was not affected by treatments, but their

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roots had lower, higher, or similar mass, depending on the cultivar-inoculum combination. Root mass was unrelated to plant number. The average level of AM colonization of daughter plants produced by noninoculated mother plants did not exceed 2%, whereas plants produced from inoculated mothers had over 10% of their root length colonized 7 weeks after transplantation of mother plants and $\sim 6\%$ after 14 weeks (harvest), suggesting that the AM fungi brought into the field by inoculated mother plants had established and spread up to the daughter plants. The host or nonhost nature of the crop species preceding strawberry plant production (barley or buckwheat) had no effect on soil mycorrhizal potential, on mother plant productivity, or on daughter plant mycorrhizal development. Thus, in soil excessively rich in P, inoculation may be the only option for management of the symbiosis.

Keywords Arbuscular mycorrhizal fungi · Preceding crop · Strawberry · Soil phosphorus · Functional specificity

Introduction

Conventional agriculture practices for high-value crops in North America often include abundant fertilization leading to nutrient accumulation in the soils. In particular, P accumulates in soils with a P fertilization history (Zhang et al. 1995). Zhang et al. (2004) concluded that large amounts of residual fertilizer P were available in the soil in subsequent years following fertilization due to the slow conversion process of residual fertilizer P to stable P forms. In Quebec, the available P (Mehlich-3 extractible P; Sen Tran and Simard 1993) level of soils associated with strawberry nursery plant production ranged from 63 to 310 mg P kg⁻¹ (i.e., from medium to excessive; CPVQ 2000) in the spring of 1998 (unpublished), when this study was initiated.

Arbuscular mycorrhizal (AM) root systems are known to support stronger, healthier, higher-yielding plants through increased nutrient acquisition (Bolgiano et al. 1983; Johnson et al. 1992; Wacker et al. 1990; Miller 2000), reduced levels of water stress (Augé 2001), lower disease incidence (Dehne 1982; St-Arnaud et al. 1995), and decreased phytohormone production (Allen et al. 1980, 1982; Shaul-Keinan et al. 2002). However, the current perception is that these obligate symbionts play no role in soils where nutrients are highly available (Olsen et al. 1999). Improved plant growth in response to AM colonization is mostly achieved in soils when available soil P is limited (Bolgiano et al. 1983; Abbott et al. 1984; Thompson 1991). AM colonization, sporulation, and plant responses are inhibited by high soil P (Abbott and Robson 1984; Liu et al. 2000). The negative effects of soil P on plant response to AM fungi occur even when the condition of high available P is imposed on highly colonized plants (Hamel et al. 1996; Dekkers and van der Werff 2000). The level of mycotrophy of the crop explains the extent of the repression (Plenchette and Morel 1996). Fertilization was also found to select for less beneficial AM fungal species due to the prolific growth of these species and reduced crop reliance on AM fungi in nutrient-rich soil (Johnson 1993). Therefore, indigenous AM fungal populations in agricultural soils might not be the ones that best enhance plant growth. Thus, in high-P soils, the inoculation of mycorrhizae responsive crops with beneficial AM fungal isolates could provide economic benefits. Some AM fungi-plant combinations are more beneficial than others (Klironomos 2003). The selection of beneficial combinations would maximize AM-derived benefits. Thus, it may be profitable to identify the AM inoculant most appropriate for a given cultivar. Biodiversity is related to higher productivity; AM fungal species are functionally different and their impact on a host plant may be complementary (Hart and Klironomos 2002). A diversity of AM fungal species (indigenous or exogenous) may allow AM fungal populations to better adapt to fluctuating environmental conditions and achieve a higher consistency in the plant responses (Koomen et al. 1987). Thus, a multiple-species inoculum could be superior to a single-species inoculum.

A most effective AM inoculum would be of little use if the AM fungi it contains does not become established in the field due to the competitive pressure of indigenous AM fungal populations. The introduction of beneficial AM fungal strains through inoculation may be facilitated by reduction of the level of AM inoculum naturally present in a field which may be better adapted to the local condition and outcompete the introduced isolates. The reduction of the level of indigenous AM fungi can be achieved by practices such as fallow (Thompson 1987) or the inclusion of nonhost plant species in a crop rotation (Harinikumar and Bagyaraj 1988; Black and Tinker 1979).

Previous studies have shown that AM inoculation of strawberry plants have benefited vegetative growth such as runner formation, number of leaves, leaf area and shoot, and root dry matter (Holevas 1966; Daft and Okusanya 1973; Koomen et al. 1987; Hrselova et al. 1990; Niemi and Vestberg 1992; Vestberg 1992; de Silva et al. 1996). Taylor and Harrier (2001) and Khanizadeh et al. (1995) found that strawberry vegetative growth response to inoculation depended on cultivar–AM species combinations.

Strawberry nursery plant production is an industry that could be enhanced by AM inoculation techniques. In Canada, elite, or top-grade, strawberry plants are produced in vitro to insure that plants are free from any deleterious endophytes. The in vitro propagated plants are acclimatized in the greenhouse before being sent to specialized nurseries where elite plants will be multiplied vegetatively through aboveground runners, or stolons, from which new daughter plants are formed in fields that are pathogen-free or fumigated. The daughter plants produced are sold as foundation, or lower-grade, plants which are transplanted into strawberry production fields, where they can produce good fruit yields for 1 to 4 years, depending on the cultivar, climate, and pest pressure. In vitro propagated crops, such as strawberry elite plants, require much less AM inoculum than field crops and can be easily inoculated in the greenhouse, at the acclimatization stage of production, before transplantation into the field for multiplication. Once trans-

planted, the precolonized elite plants would establish a mycorrhizal hyphal network in the soil, which could serve as an inoculum source for the daughter plants. The objective of this experiment was to assess, under typical commercial production conditions and, in particular high soil P fertility, the impact of commercially avail-

typical commercial production conditions and, in particular, high soil P fertility, the impact of commercially available strawberry cultivars and AM inocula combinations on the productivity of elite strawberry plants and on the mycorrhizal colonization of the daughter plants produced. The effects on the effectiveness of inoculation of preceding host and nonhost crop species commonly used in rotation with strawberry production were also tested.

Materials and methods

Experimental design

A field experiment was conducted at the Horticultural Research Center of McGill University, Ste-Anne-de-Bellevue, OC, Canada. A site was selected in a Chicot sandy loam soil that was high in nutrient content, in particular, P. The experiment had three factors: rotation crop, strawberry cultivar, and AM inoculum. Two preceding crops were selected based on their susceptibility to AM colonization: barley is a host plant and buckwheat is a nonhost plant. In 1998, the two crops were randomized into blocks consisting of two 36×6 m plots. Barley and buckwheat were sown in early June 1998 at a seeding rate of ~50 and 108 kg ha^{-1} , respectively. Both crops were sown with a row width of 15 cm. Due to the adequate nutrient content of the soil, no additional fertilizer was applied. In the spring of 1999, five strawberry cultivars (Chambly, Glooscap, Joliette, Kent, and Sweet Charlie) were preinoculated in the greenhouse with one of three AM fungal treatments (control, Glomus intraradices Schenck & Smith, or a mixture of G. intraradices, Glomus mosseae Gerdemann & Trappe, and Glomus etunicatum Becker & Gerdemann; beneficial isolates selected by Premier Tech Inc., Rivière-du-Loup, QC) and transplanted in the field with the factorial combinations (cultivar×inoculum) randomized in subplots within the main plots. The strawberry plants were transplanted 0.5 m apart, as prescribed for strawberry plant production, into two 10-m rows 1.0 m apart, which constituted each sub-subplot. Each treatment combination was replicated four times. In the fall of 1998, straw was applied uniformly on the field soil and removed in the spring of 1999. To prevent fruit formation and stimulate runner and daughter plant production, the flower buds were removed from all strawberry plants as they formed, as it is done in commercial nurseries.

Inoculation of strawberry seedlings

Elite class strawberry (Fragaria×ananassa Duch.) seedlings were obtained from in vitro cultures prepared by Phytoclone, Inc. (St-Étienne-des-Grés, QC). At the acclimation stage, the seedlings were inoculated by transplanting into PRO MIX BX (Premier Tech), a peat-based plantgrowing mix (10% peat, 90% mycorrhizal spores; pH 5.5-6.5) to which a minimum of one effective AM fungal propagule per gram of growing mix was added. Noninoculated seedlings were transplanted into uninoculated PRO MIX BX. All plantlets were placed in a greenhouse and maintained for 2 weeks under a white plastic hood providing 45% shade. The plantlets were misted two to four times per day depending on sunlight intensity. Plantlets did not receive fertilizer during this period. Over the 2-week period, the hood was gradually removed and the misting was reduced until the plantlets became completely exposed to the greenhouse environment. The temperature in the greenhouse fluctuated between 25/18°C for the daytime/nighttime periods. The photoperiod was gradually increased to 16 h using HPS 400 W (30W/M2) lamps. Following the 2 weeks under the hood, acclimation to fertilization began with 150 ppm of N in the form of calcium nitrate, potassium nitrate, and magnesium nitrate applied twice per week. Fertilizer applications continued for 6 weeks with formulations of 20-5-30 and 15-0-15 with the application of 40 ppm of N using 4–25–35 during the final week. Following 4 weeks under greenhouse conditions, the temperature was adjusted to 18/15°C daytime to nighttime periods, respectively. The plantlets were irrigated on an as needed basis.

Data collection

Prior to seeding of the rotation crops, a soil analysis was done. Soil available P, K, Ca, and Mg was measured using the Mehlich-3 extracting solution (Mehlich 1984). Buckwheat and barley were plowed in as green-manure crops before seed onset. Shoot biomass was measured by sampling the shoots of barley and buckwheat in three 1-m² quadrants and drying at 70°C for 24 h before weighing.

In the spring of 1998, the mycorrhizal potential of the soil was tested by bioassay. The mycorrhizal potential was determined by taking three undisturbed soil cylinder samples (458 cm³ of soil) per block. Two 1-week-old sorghum plants were transplanted into each cylinder, then placed in the greenhouse and maintained for 5 weeks. The cylinders were watered as necessary from the bottom to prevent crusting of the soil surface. At the end of the 5 weeks, the roots were harvested from the cylinders, washed, and cut into 1- to 1.5-cm pieces. The roots were cleared by autoclaving in 10% KOH for 12 min and by staining with 0.02% acid fuchsin (Brundett 1994) in lactic acid, glycerol, and water (1:1:1). Percent colonization was determined using the grid-line intersect method, as described by Giovannetti and Mosse (1980).

Root colonization of the strawberry transplants was determined prior to transplanting. Daughter plant colonization was determined at 7 and 14 weeks after planting by randomly sampling five daughter plants per subplot.

The total number of daughter plants was counted in 2-mlength rows to determine the average number of daughter plants per mother plant produced. From the 2-m subplot, 10 daughter plants were randomly selected for crown measurement and fresh and dry root and shoot masses measurements.

Statistical analyses

Analysis of variance (ANOVA) was done with Network JMP 3.2.6 using the following model:

Y =PC + bloc[PC] + cultivar + PC * cultivar + bloc * cultivar[PC] + inoculum + PC * inoculum + cultivar * inoculum + PC * cultivar * inoculum,

where PC stands for preceding crop. When the analysis indicated differences among the means, means were compared using contrasts. The Shapiro-Wilk W test for normality was conducted on the data prior to analysis. The distribution was not normal for the variable daughter shoot dry mass. To meet the requirement of the ANOVA test, a natural log transformation was performed on the data set prior to analysis. The distribution was not normal for the percentage of colonization of daughter plants 7 and 14 weeks after transplantation of the mother plants and no transformation could bring normality. The distribution was normal when the data related to the control inoculum treatment were removed from the colonization at 7 weeks data set. Thus, an ANOVA was conducted on this partial data. The percentage of colonization at 14 weeks could not be analyzed by ANOVA; standard errors were computed.

Results

Soil analysis

The soil analysis revealed that the soil had previously been overfertilized. This was expected as the field had a history of repeated compost applications in the years preceding this study. The near-neutral pH (7.17) Chicot sandy loam was found to have 498 mg P kg⁻¹, 324 mg K kg⁻¹, 2,731 mg Ca kg⁻¹, and 377 mg Mg kg⁻¹. As expected in a high-P soil, the indigenous mycorrhizal population had a low mycorrhizal potential of 3.32%.

Impact of treatments on AM colonization

The root colonization rate of the inoculated strawberry mother plants, prior to transplantation, never exceeded 4% and noninoculated plants had no trace of mycorrhizal structure. The percentage of daughter plant root colonization at 7 weeks following transplantation was affected by a cultivar-inoculum interaction (Table 1). While G. intraradices produced higher percentage of root colonization in 'Glooscap' (P=0.002) and 'Joliette' (P=0.04) 7 weeks after transplantation, the two infective inocula produced similar colonization levels in 'Chambly,' 'Kent,' and 'Sweet Charlie' (Fig. 1a). Less than 2% of AM colonization was found in the roots of control plants at that time. Fourteen weeks after transplantation, AM colonization had declined in all treatments except in the uninoculated controls (Fig. 1b). G. intraradices produced the highest level of colonization in Chambly, Glooscap, and Sweet Charlie roots (Fig. 1b). AM colonization levels were similar in Joliette and Kent. The significance of these effects could not be tested by ANOVA as data distribution was not normal and could not be normalized by transformation. Colonization percentages were all below 12%. It is noteworthy that uninoculated plants developed only trace levels of colonization (less than 3%) even after 14 weeks in the field. The preceding crops did not influence AM development (Table 1).

Impact of treatments on plant multiplication

Strawberry plant productivity was measured as the number of daughter plants produced per mother plant. There was a cultivar×inoculum interaction for the number of daughter



Fig. 1 Daughter plant root colonization rates of five strawberry cultivars inoculated with *G. intraradices* (*G. intra*), a mix of *G. intraradices*, *G. mosseae*, and *G. etunicatum*, or uninoculated **a** 7 weeks and **b** 14 weeks after transplantation of their mother plants in the field. Note that the 7-week data related to control plot was excluded from the ANOVA to meet the normalized and consequently was not analyzed by ANOVA. Means (n=4) ±standard error followed by similar letters are not significantly different within a cultivar, according to contrasts (P=0.05)

Source	df	Root mass	Shoot mass	Daughter plants produced	AM root colonization	
					7 weeks ^a	14 weeks ^b
Preceding crop (PC)	1	ns	ns	ns	ns	_
Cultivar	4	0.01	ns	0.008	< 0.0001	_
Inoculum	2	ns	ns	ns	< 0.0001	_
Cultivar×inoculum	8	0.05	ns	0.03	0.0003	_
PC×cultivar	4	0.002	ns	ns	ns	_
PC×inocula	2	ns	ns	ns	ns	_
PC×cultivar×inoculum	8	ns	ns	ns	ns	_

 Table 1 P values taken from ANOVAs conducted on root and shoot dry mass, number of daughter plants per mother plant, and AM colonization of daughter plants at 7 and 14 weeks from transplantation

These mother plants were different in vitro propagated elite strawberry cultivars inoculated or not with a single-species or a three-species mix inoculum, at the acclimation stage in the greenhouse

^aData related to control plots were excluded from the ANOVA to meet the normality requirement of the test

^bNormality could not be obtained for this data and thus was not analyzed by ANOVA

plants produced per mother plant (Fig. 2). Inoculation generally reduced the number of daughter plants produced per mother plant, although these reductions were significant only in Chambly (P=0.03). Glooscap, in contrast, produced ~50% more daughter plants when preinoculated with *G. intraradices* (P=0.004). Marketable plants are all healthy plants with a crown diameter of 8–13 mm (R. Hogue, personal communication), and all plants randomly chosen from all treatments produced crowns within this range.

The mean shoot dry mass of daughter plants was not affected by treatments (Table 1). A cultivar by inoculum interaction (Table 1) revealed that while the root dry biomass of Kent daughter plants was reduced (P=0.003) by both AM inocula, the mixed inoculum increased root biomass production of Joliette daughter plants (Fig. 3).

Impact of preceding crop

Barley and buckwheat crops were planted into the field to increase or decrease the size of the indigenous AM population prior to introducing selected AM fungal species through inoculation of the subsequent strawberry crop. In the spring following these preceding crops, no difference in the soil mycorrhizal potential was found. After growing sorghum plants for 5 weeks in undisturbed soil cores taken from the barley and buckwheat plots, only 3.8 and 3.9% of AM colonization was found in the roots of the trap plants. Shoot dry mass produced by the two crops in 1998 was also not significantly different (barley 1.88 tons ha⁻¹, buckwheat 1.39 tons ha⁻¹). The only impact of preceding crop



Strawberry cultivar

Fig. 2 Number of strawberry daughter plants produced per mother plant for five strawberry cultivars inoculated with *G. intraradices* (*G. intra*), a mix of *G. intraradices*, *G. mosseae*, and *G. etunicatum*, or uninoculated. Means (n=4) ±standard error followed by similar letters are not significantly different within a cultivar, according to contrasts (P=0.05)



Fig. 3 Root dry weight of strawberry daughter plant for five cultivars. Means (n=4)±standard error followed by similar letters are not significantly different within a cultivar, according to contrasts (P=0.05)

was seen on Joliette daughter plant root mass, which was higher (P=0.00003) after barley than after buckwheat.

Discussion

Impact of inoculation on AM colonization

The percent colonization rates of the strawberry roots of <12% were much lower than those reported in previous greenhouse studies where lower concentrations of P were used. For example, Chávez and Ferrera-Cerrato (1990) obtained colonization levels of 25–75% in inoculated strawberry plants. Kiernan et al. (1984) observed colonization rates of at least 26.6% when strawberry plants were inoculated with *G. mosseae*.

In spite of the low colonization levels obtained in this study, it appears that a functional mycorrhizal network was present and operating. This is evident from the higher percent root colonization rates of the inoculated vs uninoculated AM treatments at 14 weeks. The AM fungi carried in colonized mother plants appear to have spread in the soil to reach the daughter plants and increase the level of colonization of daughter plants produced by the latter. The decline in percent root colonization between 7 and 14 weeks may be due to the fact that daughter plants are produced at greater distance from the point of AM fungi introduction, the mother plants, as the season progresses (Fig. 4). The low level of colonization of daughter plants of later cohorts may be reducing the plot mycorrhizal colonization average. A decline in root colonization with time may also be a function of the natural biological decline of

Fig. 4 Diagramatic representation of strawberry vegetative reproduction. *M* Mother plant, *d* daughter plant, *S* stolon



the fungi after 14 weeks in a high-P fertile soil. High soil P level reduces both intra- and extraradical AM development (Abbott and Robson 1984; Liu et al. 2000). The higher daughter plant mycorrhizal colonization rates sometimes seen in *G. intraradices* inoculated plots at 7 and 14 weeks suggest that *G. intraradices* extraradical mycelium tends to spread over longer distance than that produced upon inoculation with the mix of *G. intraradices*, *G. mosseae*, and *G. etunicatum*, or to better resist high soil P fertility than other species.

Daughter plants well colonized by effective AM symbionts may survive transplantation better. Colonization of transplants was shown to improve plant survival after transplantation (Carpio et al. 2003; Colozzi-Filho et al. 1994; Khaliel and Elkhider 1987). Thus, it seems that inoculation of elite plants may result in the production of better-quality foundation plants, upon improvement of plant production and inoculation strategies. The pretransplant colonization rate of <4% was lower than what is expected from 5 weeks of growth in the greenhouse after inoculation. It might be possible to modify nursery inoculation practices to allow for better colonization rates of the nursery plants. For example, higher dose of inoculant could be used. The dose used in this study, although generally adequate for the production of transplants as per Premier Tech Inc. experimental tests, might be suboptimal in the case of newly rooted vitroplants. Normal commercial production practices were used in this study; therefore, fertilization might also be optimized. Increasing the length of plant acclimation time would less likely be economical considering the high cost linked to greenhouse operations. The production of foundation plants well colonized by effective AM fungi should be more successful in soil with lower P levels than the one used in our study.

Impact of inoculation on cultivar productivity

Previous studies have generally reported a beneficial effect of mycorrhizal inoculation in strawberry productivity. Holevas (1966) and Chávez and Ferrero-Cerrato (1990) observed increases in fruit yield as a result of AM inoculation. *G. intraradices* and other single-species inocula have been shown to improve stolon production (de Silva et al. 1996; Khanizadeh et al. 1995; Kiernan et al. 1984; Chávez and Ferrera-Cerrato 1990). Khanizadeh et al. (1995) observed higher stolon production in inoculated strawberry plants and, in particular, in Chambly and Kent. Under the high soil P fertility condition of our study, the effect of inoculation on daughter plant production was positive only when Glooscap was inoculated with *G. intraradices*. In this case, however, productivity increased by ~50%. It appears that large economic benefit can result from the inoculation of Glooscap elite plants. The cultivar Chambly was negatively affected by AM inoculation. For cultivars under high soil P conditions, the AM fungi may be acting as an energy drain on the plants; the output of reduced C from the plant may exceed the benefit linked to the presence of the fungi (Graham et al. 1997).

The effect of multiple-AM fungal species colonizing a host plant is not necessarily additive. On the contrary, Davies et al. (2000) report that a mix of AM fungal species reduced growth of pepper plants as compared to inoculation with *G. fasciculatum* alone. The mixed inoculant, however, was more efficient at reducing drought stress (Davies et al. 2002), emphasizing that the response of a host plant to AM colonization is not only cultivar- or isolate-specific but also depends on the environment. Under conditions typical of commercial strawberry plant production, we found that the single-species inocula, *G. intraradices*, was not different in its effect on daughter plant production from that of the mixed *Glomus* species inocula, although only *G. intraradices* increased plant productivity significantly in Glooscap.

The lack of AM inoculation effect on shoot dry weight is a contrary finding to previous greenhouse studies, using lower P concentrations in the potting media, where AM inoculation increased shoot dry weight of strawberry daughter plants (Kiernan et al. 1984; Khanizadeh et al. 1995), and cultivar-AM interaction effects influenced shoot dry weights (Chávez and Ferrera-Cerrato 1990). Khanizadeh et al. (1995) reported that AM inoculation also had an effect at nutrient solution concentrations up to 1,000 µM P. The root dry weight being influenced by an inoculation-cultivar interaction is, however, consistent with the findings of Chávez and Ferrera-Cerrato (1990). Strawberry plants with high root dry mass may survive transplantation better. In this regard, it might be beneficial to inoculate elite plants of the cultivar Joliette, as it produced daughter plants with larger root mass and hence plant of better quality. In contrast, inoculation of the cultivar Kent resulted in daughter plants with lighter root mass.

Thus, it appears that if AM inoculation technology was to be implemented in strawberry plant multiplication nurseries with high P fertility soils, it should be used only with responsive cultivars. Furthermore, care would have to be taken to use the inoculum appropriate to given cultivars. In this study, only Glooscap and Joliette benefited from inoculation. Kent always responded negatively to inoculation and inoculated Chambly produced less daughter plants, whereas Sweet Charlie showed no positive or negative growth response. However, inoculation in Chambly and Sweet Charlie led to the production of daughter plants with the largest AM colonization levels.

Impact of preceding crop

The only impact of preceding crop was a higher root mass production in Joliette daughter plants after barley than after buckwheat. This effect was unrelated to AM development. The lack of response in AM development to barley and buckwheat as host and nonhost preceding crops is in contrast to previous studies, where host plants enhanced AM fungal development thereby increasing plant productivity of the subsequent crop (Dodd et al. 1990; Thompson 1991; Gavito and Miller 1998; Karasawa et al. 2001) and nonhost plants depleted the soil of indigenous AM species (Harinikumar and Bagyaraj 1988; Black and Tinker 1979). Management of AM fungal species through selective use of AM host preceding crops leading to net benefits in the subsequent crops has been effective in low-input agriculture (Panja and Chaudhuri 2004). The lack of a preceding crop effect in this study suggests that high-P soils, with a low indigenous AM background, requires the introduction of AM fungi through inoculation to achieve AM development and benefits in a given crop.

Conclusion

We found that AM inoculation may trigger a response on strawberry plant productivity even in excessively P-rich soil, a condition not uncommon in Quebec nursery fields. This response was cultivar×AM inoculum-specific and could be large and positive or negative. Thus, care should be taken to select the proper inoculum on responsive cultivars to improve rather than reduce the profitability of nurseries with high-P fertility soils. The single-species *G. intraradices* inoculant was more effective at increasing daughter plant production than a species mix in this study. The extremely low infective capabilities of the indigenous AM fungal population studied suggest that inoculation should be preferred over AM population management through crop rotation in excessively high-P soils.

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References

- Abbott LK, Robson AD (1984) The effect of VA mycorrhizae on plant growth. In: Powell CL, Bagyaraj DJ (eds) VA mycorrhizae. CRC Press, Boca Raton, FL, USA, pp 113–130
- Abbott LK, Robson AD, De Boer G (1984) The effects of phosphorus on the formation of hyphae in soil by the vesicular–arbuscular mycorrhizal fungus, *Glomus fasciculatum*. New Phytol 97:437– 446
- Allen MF, Moore TS Jr, Christensen M (1980) Phytohormone changes in *Bouteloua gracilis* infected by vesicular–arbuscular mycorrhizae: I. Cytokinin increases in the host plant. Can J Bot 58:371–374
- Allen MF, Moore TS Jr, Christensen M (1982) Phytohormone changes in *Bouteloua gracilis* infected by vesicular–arbuscular mycorrhizae. II. Altered levels of gibberellin-like substances and abscisic acid in the host plant. Can J Bot 60:468–471
- Augé RM (2001) Water relations, drought and vesicular–arbuscular mycorrhizal symbiosis. Mycorrhiza 11:3–42
- Black R, Tinker PB (1979) The development of endomycorrhizal root systems. II. Effect of agronomic factors and soil conditions on the development of vesicular–arbuscular mycorrhizal infection in barley and on the endophyte spore density. New Phytol 83:401–413
- Bolgiano NC, Safir GR, Warnicke DD (1983) Mycorrhizal infection and growth of onion in the field in relation to phosphorus and water availability. J Am Soc Hortic Sci 108:819–825
- Brundett M (1994) Estimation of root length and colonization by mycorrhizal fungi. In: Brundett M, Melville L, Peterson L (eds) Practical methods in mycorrhiza research. Mycologue Publications, Guelph, pp 51–59
- Carpio LA, Davies FT Jr, Arnold MA (2003) Effect of commercial arbuscular mycorrhizal fungi on growth, survivability, and subsequent landscape performance of selected container grown nursery crops. J Environ Hortic 21:190–195
- Chávez MCG, Ferrera-Cerrato R (1990) Effect of vesicular– arbuscular mycorrhizae on tissue culture-derived plantlets of strawberry. HortScience 25:903–905
- Colozzi-Filho A, Siqueira JO, Saggin Júnior O, Guimarães PTG, Oliveira E (1994) Effectiveness of different arbuscular–mycorrhizal fungi on initial and post-transplant growth and yield of coffee trees. Pesqui Agropecu Bras 29:1397–1406
- CPVQ (2000) Fertilization recommendations. Conseil des productions végétales du Québec, Inc.
- Daft MJ, Okusanya BO (1973) Effect of *Endogone mycorrhiza* on plant growth. VI. Influence of infection on the anatomy and reproductive development in four hosts. New Phytol 72:1333–1339
- Davies FT Jr, Olalde-Portugal V, Alvarado MJ, Ecamilla HM, Ferrera-Cerrato RC, Espinosa IJ (2000) Alleviating phosphorus stress of Chile ancho pepper (*Capsicum annuum* L. cv. San Luis) with arbuscular mycorrhizal inoculation. J Hortic Sci Biotechnol 75:661–665
- Davies FT Jr, Olalde-Portugal V, Aguilera-Gomez L, Alvarado MJ, Ferrera-Cerrato RC, Boutton TW (2002) Alleviation of drought stress of Chile ancho pepper (*Capsicum annuum* L. cv. San Luis) with arbuscular mycorrhiza indigenous to Mexico. Sci Hortic 347–359
- Dehne HW (1982) Interaction between vesicular–arbuscular mycorrhizal fungi and plant pathogens. Phytopathology 72:1115–1119
- Dekkers TBM, van der Werff PA (2000) Mutualistic functioning of indigenous arbuscular mycorrhizae in spring barley and winter wheat after cessation of long-term phosphate fertilization. Mycorrhiza 10:95–202
- de Silva A, Patterson K, Mitchell J (1996) Endomycorrhizae and growth of 'sweetheart' strawberry seedlings. HortScience 31: 951–954

- Dodd JC, Koomen IA, Hayman DS (1990) The management of populations of vesicular–arbuscular mycorrhizal fungi in acidinfertile soils of a savanna ecosystem. Plant Soil 122:229–240
- Gavito ME, Miller MH (1998) Changes in mycorrhiza development in maize induced by crop management practices. Plant Soil 198: 185–192
- Giovannetti M, Mosse B (1980) An evaluation of techniques for measuring vesicular–arbuscular infection in roots. New Phytol 84:489–500
- Graham JH, Duncan LW, Eissnestat DM (1997) Carbohydrate allocation patterns in citrus genotypes as affected by phosphorus nutrition, mycorrhizal colonization, and mycorrhizal dependency. New Phytol 135:335–343
- Hamel C, Dalpé Y, Lapierre C, Simard RR, Smith DL (1996) Endomycorrhizae in a newly cultivated acidic meadow: effects of three years of barley cropping, tillage, lime and phosphorus on root colonization and soil infectivity. Biol Fertil Soils 21:160– 165
- Harinikumar DM, Bagyaraj DJ (1988) Effect of crop rotation on native vesicular–arbuscular mycorrhizal propagules in soil. Plant Soil 110:77–80
- Hart MM, Klironomos JN (2002) Arbuscular mycorrhizal fungal diversity and ecosystem function. In: van der Heijden MGA, Sanders IR (eds) Ecological studies 157. Mycorrhizal ecology. Springer, Berlin Heidelberg New York, pp 225–242
- Holevas CD (1966) The effect of a vesucular–arbuscular mycorrhiza on the uptake of soil phosphorus by strawberry (*Fragaria* sp. var. Cambridge Favourite). J Hort Sci 41:57–64
- Hrselova H, Gryndler M, Vancura V (1990) Influence of inoculation with VA mycorrhizal fungus *Glomus* sp. on growth of strawberries and runner formation. Agric Ecosyst Environ 29:193– 197
- Johnson NC (1993) Can fertilization of soil select less mutualistic mycorrhizae? Ecol Appl 3:749–757
- Johnson NC, Copeland PJ, Crookston RK, Pfleger FL (1992) Mycorrhizae: possible explanation for yield decline with continuous corn and soybean. Agron J 84:387–390
- Karasawa T, Kasahara Y, Takebe M (2001) Variable response of growth and arbuscular mycorrhizal colonization of maize plants to preceding crops in various types of soil. Biol Fertil Soils 33:286–293
- Khaliel AS, Elkhider KA (1987) Response of tomato to inoculation with vesicular–arbuscular mycorrhiza. Nord J Bot 7:215–218
- Khanizadeh S, Hamel C, Kianmehr H, Buszard D, Smith DL (1995) Effect of three vesicular–arbuscular mycorrhizae species and phosphorus on reproductive and vegetative growth of three strawberry cultivars. J Plant Nutr 18:1073–1079
- Kiernan JM, Hendrix JW, Stolz LP, Maronek DM (1984) Characterization of strawberry plants produced by tissue culture and infected with specific mycorrhizal fungi. HortScience 19:883– 885
- Klironomos JN (2003) Variation in plant response to native and exotic arbuscular mycorrhizal fungi. Ecology 84:2292–2301
- Koomen I, Grace C, Hayman DS (1987) Effectiveness of single and multiple mycorrhizal inocula on growth of clover and strawberry plants at two soil pHs. Soil Biol Biochem 19:539–544
- Liu Å, Hamel C, Hamilton RI, Ma BL, Smith DL (2000) Acquisition of Cu, Zn, Mn, and Fe by mycorrhizal maize (*Zea mays* L.) grown in soil at different P and micronutrient levels. Mycorrhiza 9:331–336

- Mehlich A (1984) Mehlich-III soil test extractant: a modification of Mehlich-II extractant. Commun Soil Sci Plant Anal 15:1409– 1416
- Miller MH (2000) Arbuscular mycorrhizae and the phosphorus nutrition of maize: a review of Guelph studies. Can J Plant Sci 80:47–52
- Niemi M, Vestberg M (1992) Inoculation of commercially grown strawberry with VA mycorrhizal fungi. Plant Soil 144:133–142
- Olsen JK, Schaefer JT, Edwards DG, Hunter MN, Galea VJ, Muller LM (1999) Effects of a network of mycorrhizae on capsicum (*Capsicum annuum* L.) grown in the field with five rates of applied phosphorus. Aust J Agric Res 50:239–252
- Panja BN, Chaudhuri S (2004) Exploitation of soil arbuscular mycorrhizal potential for AM-dependant mandarin orange plants by pre-cropping with mycotrophic crops. Appl Soil Ecol 26:249–255
- Plenchette C, Morel C (1996) External phosphorus requirements of mycorrhizal and non-mycorrhizal barley and soybean plants. Biol Fert Soils 21:303–308
- Sen Tran T, Simard RR (1993) Mehlich III-extractable elements. In: Carter M (ed) Soil sampling and methods of analysis. Boca Raton, FL, USA, pp 39–42
- Shaul-Keinan O, Gadkar V, Ginzberg I, Grünzweig JM, Chet I, Elad Y, Wininger S, Belausov E, Eshed Y, Atzmon N, Ben-Tal Y, Kapulnik Y (2002) Hormone concentrations in tobacco roots change during arbuscular mycorrhizal colonization with *Glomus intraradices*. New Phytol 154:501–507
- St-Arnaud M, Hamel C, Caron M, Fortin JA (1995) Endomycorrhizes VA et sensibilité aux maladies: synthèse de la littérature et mécanismes d'interaction probables. In: Charest C, Bernier R (eds) Mycorrhizal symbiosis. Orbis, Frelighsburg, QC, pp 51–87
- Taylor J, Harrier L (2001) A comparison of development and mineral nutrition of micropropagated *Fragaria×ananassa* cv. Elvira (Strawberry) when colonised by nine species of arbuscular mycorrhizal fungi. Appl Soil Ecol 18:205–215
- Thompson JP (1987) Decline of vesicular–arbuscular mycorrhizae in a long fallow disorder of field crops and its expression in phosphorus deficiency of sunflower. Aust J Agric Res 38:847– 867
- Thompson JP (1991) Improving the mycorrhizal condition of the soil through cultural practices and effects on growth and phosphorus uptake by plants. In: Johansen C, Lee KK, Sahrawat KL (eds) Phosphorus nutrition of grain legumes in the semi-arid tropics. International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patencheru, India, pp 117–137
- Vestberg M (1992) Arbuscular mycorrhizal inoculation of micrpropagated strawberry and field observations in Finland. Agronomie 12:865–867
- Wacker TL, Safir GR, Stephens CT (1990) Effect of *Glomus fasciculatum* on the growth of asparagus and the incidence of *Fusarium* root rot. J Am Soc Hortic Sci 115:550–554
- Zhang TQ, MacKenzie AF, Liang BC (1995) Long-term changes in Mehlich-3 extractable P and K in a sandy loam soil under continuous corn (*Zea mays* L.). Can J Soil Sci 75:361–367
- Zhang TQ, MacKenzie AF, Liang BC, Drury CF (2004) Soil test phosphorus and phosphorus fractions with long-term phosphorus addition and depletion. Soil Sci Soc Am J 68:519–528