Introduction

Soil organisms, together with mycorrhizal fungi play a crucial role in the functioning of agricultural ecosystems. One type of mycorrhizal association, arbuscular mycorrhizal (AM) symbiosis, can contribute significantly to plant nutrition by promoting the uptake of phosphorus and nitrogen (George, 2000) resulting in improved plant growth and health. Colonization by AM fungi may also improve plant establishment, increase uptake of some ions and influence nutrient cycling; enhance plant tolerance to (biotic and abiotic) stress; improve the quality of soil structure; promote earlier flowering and fruiting; and increase crop uniformity.

However the effects of AM on the growth and development of horticultural crop plants have been described in many research papers (Azcón-Aguilar & Barea 1997; Lovato et al., 1995) but research focused on typical vegetable from Hungary is missing.

Hungary is known for spice pepper (Capsicum annuum L. var. longum), which is protected by EU as a national cultivar. It is traditionally cultivated in the Szeged and Kalocsa regions of Hungary (Somogyi et al., 2000) and about 28.6 thousand tons are annually produced from 1,500 ha (average 2000-2009). A growing awareness of sustainable agriculture, high-quality food, and more information on how food is produced has caused a demand for reduced chemical inputs in pepper cultivation also.

The aim of the present study was to evaluate a possible sustainable, ecological way of producing mycorrhizal inoculums Symbivit (Symbio-m, CZ) on growth and yield of field grown spice pepper.
Material and methods

Experimental design

The experiment was carried out at the Experimental Station of Szent István University Gödöllő, Hungary (longitude, 19°21’39”, latitude, 47°35’57”). The climate of the region is continental with a mean annual temperature and precipitation of 10.6°C and 539 mm, respectively. Seedlings of pepper (Capsicum annuum L. var. longum), cv. Szegedi and cv. Kalocskai, were propagated at the beginning of April in a greenhouse using special horticulture substrate (Klasmann TS3: 80% white sphagnum peat and 20% frozen black sphagnum peat, slow-release 14:16:18 (w/w/w) NPK fertilizer, pH 6.00) for 7 weeks and were bedded out on 26 May in a sandy soil with medium phosphorus supply. The major soil properties in the 0–20 cm layer were: pH (KCl): 6.67; humus content: 1.56; salt: 0.05%, AL-P2O5: 84.0 mg·kg⁻¹, AL-K2O: 113.0 mg·kg⁻¹, AL-Ca: 1347.0 mg·kg⁻¹, AL-Mg: 142.0 mg·kg⁻¹, NH4+-N: 0.59 mg/100 g, NO3--N: 2.19 mg/100 g, Cu: 3.63 mg·kg⁻¹, Fe: 1016.0 mg·kg⁻¹, Mn: 263.0 mg·kg⁻¹, Zn: 6.49 mg·kg⁻¹. A variety of cereals have been grown alternately on the site, however the last two years covered by grass/cover to restore soil fertility. A moldboard plow to 25 cm depth was used for soil tillage after each harvesting time and conventional seedbeds were prepared by chisel plowing followed by disking.

Before transplanting mycorrhizal fungi in a commercial product Symbivit® (mixture of G. intraradices BEG140, G. mosseae BEG95, G. etunicatum BEG92, G. claroideum BEG96, G. microaggregatum BEG56, G. geosporum BEG199, without bioadditives) produced by Symbiom Ltd. (Lanskroun, Czech Republic; www.symbiom.cz) was applied at 15 g of inoculum per pepper seedling into the planting hole and seedlings were planted immediately. In the experiment 50 plants of each pepper types inoculated with Symbivit (AM-treated) and 50 non-inoculated plants (Non-treated) were set up in three replicates using randomized block design. Five randomly chosen plants from each treatment were destructively harvested at the end of the experiment (on 3 September), shoots and roots, were dried at 60°C for 72 h and separately weighed. Harvesting was by hand twice and cumulative weight of yield from 100 plants per treatment evaluated.

Assessment of mycorrhizal colonization and hyphal length of AMF

Samples for estimating root colonization were collected on 26 June, 17 July, 27 July, 14 August and 3 September. At each time three randomly chosen from three repetitive plots of the same treatment plants were dug out with a soil core of 25 × 25 × 25 cm. The roots and the soil were stored in separate plastic bags at 4°C until processing within 24 h. Approximately, half of the root systems, at least 500 mg of fine roots from each plant were transferred to separate tubes and were subjected to Trypan Blue staining (Phillips & Hayman, 1970). Internal fungal structures (hypae, arbuscules, vesicles) were examined under a stereomicroscope at × 100 magnification and the percentage of root length colonized calculated using the gridline intersect method (Giovannetti & Mosse, 1980).

Determination of AM fungal hyphal length in the soil was based on the methods of Baath & Söderstrom (1979). A 1-g sample of dry rhizosphere soil from the last harvesting was dispersed in 100 mL deionized water in a blender for 1 min. Fungal hyphae were separated by wet-sieving and centrifugation. The separated fungal hyphae were placed in a Petri dish with 5 mL deionized water. Ten millilitres of agar solution (0.75%) containing trypan blue (0.05%) was added to each dish, and the mixture was then dried for 24 h at 70°C. The hyphal length was measured in the dried agar film by the intersection method (Tennant, 1975) under a binocular microscope (16 x magnification).

Data were analyzed by a one-way analysis of variance. Individual treatment differences were subjected to *t* test. Data were analyzed using Statistica 6.1 (StatSoft, Tulsa, OK, USA) software.

Results and discussion

Mycorrhizal technology is being used more frequently in horticultural vegetable production (Vosátk & Albrechtová, 2008) and an international mycorrhizal industry is developing (Vosátk et al., 2008). Under the threat of a depletion of world phosphate deposits, use of mycorrhizal inoculation in horticulture becomes even more desirable due to savings in chemical inputs and use of renewable materials in vegetable production.

Our results showed that the mycorrhizal AM-based commercial inoculant Symbivit was able to establish in roots and increased growth and yield of both spice pepper types. Mycorrhizal inoculation increased fresh (Figure 1.) and dry weights of shoots of spice pepper cv. Szegedi and only fresh weight of Kalocskai type (Figure 2., Table 1). There were no significant differences in the root weights due to treatment only in fresh weight of Kalocskai pepper type (Table 1.).

At each sampling date, except for that in July, the percent of mycorrhizal root colonization was greater for the AM-treated and non-treated plants (Figure 3A, 3B). However the relative high percentage of root colonization in the control non-inoculated treatment indicated the presence of indigenous populations of AMF in the soil. The root colonization showed seasonality by treated and non-treated plants. The lowest degree of colonization was observed in June in general and colonization percent increased during vegetative development and there was a slight decrease at harvesting.

Treated plants of both variants exhibited an increase in cumulative crop production compared with control non-treated plants. Inoculation with a commercial product containing a mixture of non-indigenous *Glomus* spp. could promote yield benefits of spice pepper cv. Szegedi by more...
Table 1. Effects of commercial mycorrhizal inoculants on growth, external hyphal length of AMF and cumulative crop production during cultivation of spice pepper cv. Szegedi and cv. Kalocsai

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Fresh weight</th>
<th>Dry weight</th>
<th>External hyphal length</th>
<th>Cumulative crop production</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>[g plant⁻¹]</td>
<td>[g plant⁻¹]</td>
<td>[m soil g⁻¹]</td>
<td>[g 100 plants⁻¹]</td>
</tr>
<tr>
<td>Szegedi</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AM-treated</td>
<td>1.66a</td>
<td>19.32b</td>
<td>1.30a</td>
<td>8.32b</td>
</tr>
<tr>
<td>Non-treated</td>
<td>1.77a</td>
<td>13.22a</td>
<td>0.97a</td>
<td>7.09a</td>
</tr>
<tr>
<td>Kalocsai</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AM-treated</td>
<td>3.38b</td>
<td>18.69b</td>
<td>2.18a</td>
<td>11.76a</td>
</tr>
<tr>
<td>Non-treated</td>
<td>2.16a</td>
<td>15.05a</td>
<td>1.48a</td>
<td>9.19a</td>
</tr>
</tbody>
</table>

Values followed by the same letter are not significantly different, P≤0.05, Tukey test. Values are means of five observations.

than 60% and by 10% of Kalocsai. The increase in pepper yield as a consequence of inoculation was observed by other authors (Gaur et al., 1998; Douds & Reider, 2003; Kaya et al., 2009; Russo & Perkins-Veazie, 2010) but not in spice pepper.

Mycorrhizal inoculation had a great positive effect on hyphal length also (Table 1) and there were differences between Kalocsai and Szegedi variants. According to the increased yield benefit, the same tendency was observed: spice pepper cv. Szeged showed higher values in hyphal length than Kalocsai pepper type. Extraradical hyphal length in the soil is directly related to root colonization, though colonization rates can be a poor predictor of extraradical hyphal length (Bingham & Biondini, 2009). As the plant allocates photosynthates to the fungus, it promotes hyphal growth inside and outside of the root, and the extension of

the extraradical hyphae results in greater nutrient transfer from soil to the plant (Lebron et al., 2012). In experimental chambers, the external hyphae of AM can deliver up to 80% of plant P, 25% of plant N, 10% of plant K, 25% of plant Zn and 60% of plant Cu (Marschner & Dell, 1994). Nutrient uptake by mycorrhizal inoculants was not measured in our experiments, but the increased growth of treated spice pepper together with increased root colonization and hyphal length really show their role in pepper cultivation.

Besides the activity of external hyphae of AMF, arbuscular mycorrhizal fungi directly or indirectly affect the soil ecosystem through multiple interactions with other soil organisms. Although they are not saprotrophs, AMF can enhance the rate of decomposition of organic material (Hodge et al., 2001), indirectly influencing decomposition.
measuring root colonization only the occurrence of AMF could be detected but the increased yield benefit together with increased hyphal length due to mycorrhizal inoculation show that AMF from inoculants were able to establish and affect vegetable plants.

In conclusion, it can be stated that inoculation with Symbivit containing mycorrhizal fungi could be an integral part of spice pepper production.

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References


Marschner & Dell


