INFLUENCE OF NON-HOST PLANTS ON VESICULAR-ARBUSCULAR MYCORRHIZAL INFECTION OF HOST PLANTS AND ON SPORE GERMINATION

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Summary—Host (Medicago sativa) and non-host (Brassica oleracea) plants were grown in pots of sand-vermiculite or soil inoculated either with soil inoculum or with spores of Glomus mosseae. Cabbage only decreased the vesicular-arbuscular (VA) mycorrhizal infection of alfalfa plants, when both plants were grown together in sand-vermiculite pots inoculated with spores. The root exudates of the non-host plants did not affect the percentage spore germination. However, the percentage spore germination was decreased by volatile compounds of non-host plant roots.

The characteristics of the inhibiting factors of VA mycorrhizal infection in non-host plants are discussed.

INTRODUCTION

Several plants belonging to families such as Cruciferae and Chenopodiaceae do not develop VA mycorrhizal infection in their roots (Harley and Harley, 1987; Newman and Reddell, 1987). The lack of VA infection in these plants has been attributed to intrinsic characteristics of the cortex rather than to factors released in exudates (Ocampo et al., 1980; Glenn et al., 1985). Nevertheless, inhibition of the VA infection of host plants by some non-host plants has been observed (Hayman et al., 1975; Iqbal and Qureshi, 1976). These results indicated that the root exudates of the non-host plants can be implicated in the lack of infection of these plants and in the decrease of VA infection in the host plants, when they were grown together. However, from these experiments the possible negative effect of volatile compounds cannot be excluded. The inhibitory effects of non-host plants seem to depend on the conditions under which they were grown (Powell, 1980). The importance of volatile compounds of plant roots on VA mycorrhizal infection has been suggested (Koske, 1982). Indications of the negative effect of volatile compounds from Brassica napus roots on spore germination and VA hyphal extension have been observed (Tommerup, 1984). Therefore the effect of root exudates and volatile compounds of Cruciferae and their effect on VA infection of host plants with different type of inoculum and plant growth medium was studied.

MATERIALS AND METHODS

Experiment 1

The experiment was carried out in open pots of sterilized sand-vermiculite (1:1 v/v) or steamed soil (Barea et al., 1980), mixed with sterilized sand (1:1 v/v). Alfalfa (Medicago sativa cv. Aragon) and cabbage (Brassica oleracea cv. Brunswick) were used as test plants. Seeds were grown in moistened sand, and 2-week-old seedlings of alfalfa alone or together with cabbage were transplanted to the pots and grown under greenhouse conditions (supplementary light (Sylvania incandescent and cool-white lamps, 400 mmol m$^{-2}$ s$^{-1}$ 400–700 nm), 16–8 h light–dark cycle, 25–19°C and 50% R.H.).

The soil and sand-vermiculite pots were inoculated either with 30 sporocarps of Glomus mosseae or with 5 g of rhizosphere soil from maize plant pot cultures of the same G. mosseae isolate, which contained sporos, mycelium and infected root fragments. Plants were also inoculated with the strain 203 of Rhizobium meliloti isolated in this laboratory. Plants were watered from below, using a capillary system. Every week 5 ml of 1 mg Ashton nutrient solution (Hewitt, 1952), lacking nitrogen and phosphate, were added to the soil pots. The sand-vermiculite pots were given 1/2 strength plus 50 mg l$^{-1}$ of phosphate of the same nutrient solution. Ten replicates per treatment were used. After 10 weeks, root samples of each replicate pot were cleared and stained (Phillips and Hayman, 1970), and the root infection evaluated in the gridline intersect method (Giovannetti and Mosse, 1980). Shoot dry matter was recorded.

Experiment 2

Seeds of alfalfa and cabbage were surface-sterilized with HgCl$_2$ for 10 min and then thoroughly rinsed with sterile water. Seedlings were transferred to sterilized glass tubes. Five replicates, containing a strip of filter paper (Axcon-Aguilar and Barea, 1979) and 10 ml of 1/2 strength Long Ashton nutrient solution at pH 7.9, were used. Tubes without plants were used as nutrient solution controls. Plants were grown in a growth cabinet under the same ambient conditions as described for the greenhouse. Root exudates from plants were those accumulated in the nutrient solu-
Table 1. VA infections and shoot dry weights of alfalfa grown alone or together with cabbage and inoculated with G. mosseae

<table>
<thead>
<tr>
<th>Growth medium</th>
<th>VA inoculum</th>
<th>Plants</th>
<th>Shoot dry wt (mg)</th>
<th>% Root length infection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soil inoculum</td>
<td>Alfalfa</td>
<td>62 ± 10</td>
<td>60 ± 8</td>
<td></td>
</tr>
<tr>
<td>Soil</td>
<td>Alfalfa plus cabbage</td>
<td>74 ± 12</td>
<td>58 ± 7</td>
<td></td>
</tr>
<tr>
<td>Sporocarps</td>
<td>Alfalfa</td>
<td>71 ± 38</td>
<td>ND</td>
<td></td>
</tr>
<tr>
<td>Sporocarps</td>
<td>Alfalfa plus cabbage</td>
<td>57 ± 19</td>
<td>13 ± 7</td>
<td></td>
</tr>
<tr>
<td>Sporocarps</td>
<td>Alfalfa</td>
<td>36 ± 5</td>
<td>15 ± 2</td>
<td></td>
</tr>
<tr>
<td>Sporocarps</td>
<td>Alfalfa plus cabbage</td>
<td>161 ± 24</td>
<td>ND</td>
<td></td>
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<tr>
<td>Sporocarps</td>
<td>Alfalfa</td>
<td>39 ± 14</td>
<td>23 ± 6</td>
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</tr>
<tr>
<td>Sporocarps</td>
<td>Alfalfa plus cabbage</td>
<td>23 ± 4</td>
<td>25 ± 5</td>
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<tr>
<td>Sporocarps</td>
<td>Alfalfa</td>
<td>159 ± 31</td>
<td>ND</td>
<td></td>
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<tr>
<td>Sporocarps</td>
<td>Alfalfa</td>
<td>47 ± 15</td>
<td>29 ± 5</td>
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<td>Sporocarps</td>
<td>Alfalfa plus cabbage</td>
<td>21 ± 2</td>
<td>4 ± 0.5</td>
<td></td>
</tr>
<tr>
<td>Sporocarps</td>
<td>Alfalfa</td>
<td>213 ± 21</td>
<td>ND</td>
<td></td>
</tr>
</tbody>
</table>

Each figure is the mean for 10 pots. Standard errors of mean are given. ND = not determined.

Experiment 3

In order to see the effect on spore germination of gaseous substances from alfalfa and cabbage roots, a technique based on the method of Koske (1982) was used.

Seeds of alfalfa and cabbage were surface-sterilized and germinated in Petri dishes containing moistened filter paper. When roots of the seedlings were 2-3 cm long, plants were transferred to plastic Petri dishes with 3 pieces of filter paper moistened with 5 ml of Long Ashton nutrient solution. Surface sterilized spores of G. mosseae were placed on an agar plug (2 cm dia x 4 mm) which was attached to the inside of the Petri dish lid directly above the roots. Ten plates were used, containing 10-15 spores per plate. Petri dishes without plants, with nutrient solution or with water, were used as controls.

Then the Petri dishes were wrapped in aluminium foil covering spores and roots but not plant shoots and kept inclined on a tray with a 45° angle. Plants were grown in a growth cabinet for 15 days and spore germination percentage was determined after 1 and 2 weeks.

The three experiments were repeated twice under the experimental conditions described and similar results were obtained.

RESULTS

Table 1 shows that in sterilized soil alfalfa plants grown singly and inoculated with soil inoculum of G. mosseae, developed more VA infection than inoculated with sporocarps. VA infection was not decreased by the presence of the "non-host" cabbage. In contrast VA infection was reduced when two plants were grown together in sand-vermiculite pots, inoculated with sporocarps, though not when inoculated with soil inoculum.

Non-significant differences in the percentage spore germination (Table 2) in presence of root exudates from alfalfa or from cabbage were found. There was some indications that root exudates from alfalfa increased the percentage spore germination, but this was not statistically significant.

The production of volatile compounds by cabbage plants decreased the percentage of spore germination (Table 3).

DISCUSSION

The inhibitory effect of root extracts of non-host plants on VA mycorrhizal infection of host plants (Ocampo et al., 1986), indicated that the barriers to mycorrhizal infection in non-host plants are intrinsic and more probably related to physiological characteristics of the root cortex or epidermis than to any compounds that might be released by root exudates (Ocampo et al., 1980). From the results of our
experiments the absence of inhibitory effect of soluble root exudates of cabbage cannot be discarded because we do not know whether the concentration of exudates was similar to that in the soil, nor whether exudates from older plants are different. However, in other experiments, germination and hyphal growth of VA spores were unchanged either by the age of non-host seedlings at transplanting (Tommerup, 1984) or by the age of non-host plants grown in soil or in sand pots (Azcon and Ocampo, 1984). Thus the reduced rate of spore germination and the decrease of root infection of alfalfa plants, when grown together with cabbage, indicates that inhibitory volatile compounds were produced by cabbage. This effect was influenced by the medium in which the plants were grown. As could happen with the root extracts (Ocampo et al., 1986), the absence of effect of gaseous substances of non-host plants on VA infection of alfalfa grown in soil can be related to the more effective degradation of inhibiting substances by the soil rhizosphere microorganisms or by absorption sites on soil particles. The different species of spores and types of inoculum may also have been implicated in the different results obtained by different authors. No Gigaspora margarita entry points aborted or not, were observed on cabbage roots in presence of lettuce (Ocampo et al., 1980) and Gigaspora gigantea did not penetrate brassica roots (Glen et al., 1985); in contrast, Glomus fasciculatum “E3” (Ocampo et al., 1980) and G. mosseae (Ocampo et al., 1980; Glen et al., 1985) which penetrated the root of several Cruciferae tested. VA infection of host plants, grown in sand-vermiculite pots, were inhibited by non-host plants (Table 1), or by its root extracts (Ocampo et al., 1986), only when were inoculated with sporocarps of G. mosseae.

Future investigations are needed to elucidate the nature of these volatile inhibitory substances and their possible relationships to the spread of mycorrhizal infection in plant roots.

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REFERENCES


