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# COLLEMBOLA DECREASE THE NITROGEN UPTAKE OF MAIZE THROUGH ARBUSCULAR MYCORRHIZA

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Abstract

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There is little information on how AMF (arbuscular mycorrhizal fungi) functions are influenced by Collembola. In this experiment the question was addressed whether the presence of Collembola may decrease the nitrogen uptake of maize plant through AM-fungi system. A microcosm experiment was set up on the field. The microcosms were separated into two compartments by a screen of 42 μm mesh size. A maize seedling was planted in one compartment of each microcosm. A mycorrhizal inoculum of *Glomus mosseae* and *Gigaspora rosea* was propagated into the soil. $^{15}N$ marked ammonium sulphate was applied at a distance of 0.15 m from the plant in the root-free compartment of the microcosms with or without Collembola (*Sinella coeca*). Hyphal length, N and $^{15}N$ content of the maize plants and the microbial biomass in the root-free compartment were significantly decreased in the presence of *S. coeca*. Presumably, Collembola destructed AMF hyphal network that led to decreased nitrogen uptake of plants.

*Key words*: Arbuscular mycorrhizal fungi, Collembola, N-uptake

## Introduction

Springtails have different effects on arbuscular mycorrhizal fungi (AMF). Many species of Collembola graze on AM fungi (Larsen, Jakobsen, 1996) and different Collembola species have a preference for different AMF species (Moore et al., 1985; Warnock et al.,

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1982). The effects of Collembola on the growth and function of AM symbiosis form a continuum from negative to positive influences. In particular, the effect may depend on the density of Collembola (Bakonyi et al., 2002; Gange, 2000). In case of low collembolan density stimulating effects may occur, while at high densities negative effects of AMF-consumption by Collembola will dominate. Bakonyi et al. (2002) found that the degree of AMF colonization was the highest at a density of 0.2–0.4 *Sinella coeca* g soil$^{-1}$. Higher or lower collembolan densities decreased the colonization rate. However, there is little information on how functions of AM fungi are influenced by Collembola (Gange, 2000). In this experiment the question was addressed whether high density of Collembola may decrease the nitrogen uptake of maize plant through the destruction of the AMF hyphal network.

## Materials and methods

A microcosm experiment was set up on the field. Each microcosm was separated by a screen of 42 μm mesh size into two compartments A and B (Fig. 1.) Microcosms were filled up with brown forest soil previously air-dried on the sun for a week in order to reduce the density of meso- and macrofauna. The soil was collected in the Botanical Garden of the Szent István University (Gödöllő, Hungary). The main chemical parameters of the soil were as follows: total C 1.6%, total N 0.15%, P 712 mg kg$^{-1}$, K 0.29%, Fe 0.78%, Zn, 43 mg kg$^{-1}$, Mn 480 mg kg$^{-1}$, $pH_{(H_2O)}$ 7.5. 500 g soil was filled into compartment A (length: 8 cm, diameter: 10.5 cm) and 1000 g soil into compartment B (length:15 cm, diameter:10.5 cm).

Pre-germinated maize seedlings were planted in the compartment B. Maize (*Zea mays* L.cv.P i o n e e r) seeds were surface sterilized using 30% $H_2O_2$ for 10 minutes and subsequently washed with sterile distilled water. Two seedlings were planted in each pot and after emergence thinned to one plant per pot. The soil surface of all pots was covered with a 2 cm layer of gravel sand (2 mm size) to minimaze evaporation.

The inoculum of *Glomus mosseae* (BEG 12*)* and *Gigaspora rosea* (BEG 9) were propagated on maize grown in a greenhouse for 7 weeks. For mycorrhiza treatments roots with adhering soil were used as inoculum. Each pot received an equal amount of inoculum (10% of soil volumes, respectivaly) into soil of compartment B. The roots could not penetrate grow into the root-free compartment A. The AMF were able to spread to both compartments of the microcosms. Microcosms were randomly placed near by near on a table in 1 m high. Plants were watered twice a week with 100 ml tap water.

Two different treatments were set up. $^{15}N$ was added at a distance of 0.15 m from the roots of the maize with (C+) or without (C-) Collembola. Number of replications were three. The total number of experimental pots were six. Mycorrhiza was allowed to grow for six weeks. Six weeks after the maize planting 200 mg $^{15}N$ marked ammonium sulphate (labelled with $^{15}N$ at 49.8 atom percent excess) solved in distilled water was put to each compartment A. Same time adult individuals of the Collembola *Sinella coeca* was put in the maize free compartment of the microcosms in a density of 0.6 animal g$^{-1}$ soil. Supposing that most Collembola individuals live in the upper 5 cm of the soil (Larink, 1997) an approximate calculation shows that $3x10^{-4}$ individuals m$^{-2}$ refers to 0.6 individuals g$^{-1}$ soil. Microcosms were destructively sampled after two weeks, the age of the plants was eight week. Total plant biomass, shoot biomass, root biomass, shoot/root ratio, plant water content, the total N content and $^{15}N$ content of the above ground plant parts, length of the AMF hyphae and microbial biomass were measured. N and $^{15}N$ concentrations were determined by VG Micromass 622 mass spectrometer. Determination of AM fungal hyphal length in the soil was based on methods of Baath and Söderström (1979). The hyphal length was measured in the dried agar film with the intersection method (Tennant, 1975) under a binocular microscope (16 x magnification). Microbial biomass was determined by the method of Amato and Ladd (1988). After checking for normality of the data, means were compared with two-sample two-tailed t-test. Ratio or percentage data were compared by Mann-Whitney u-test.

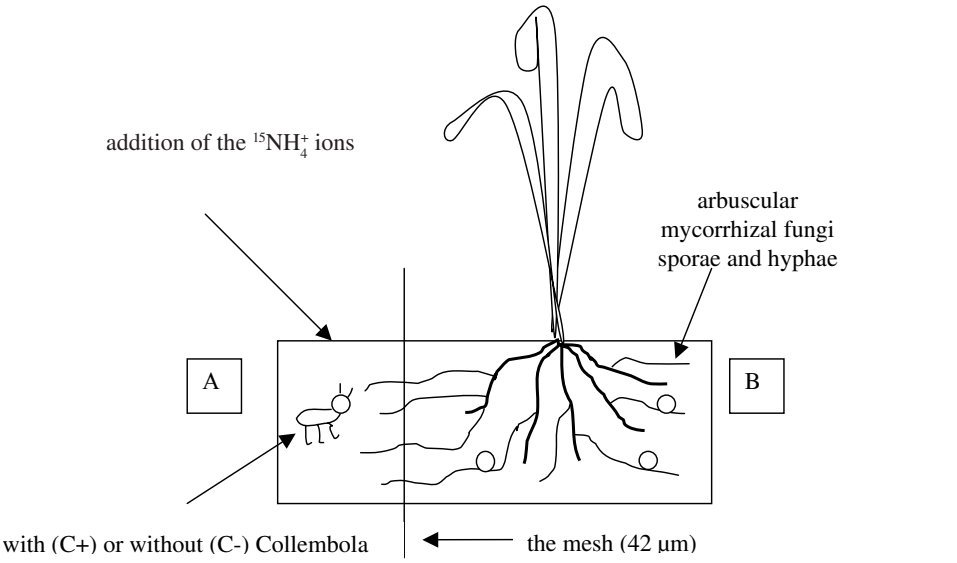


Fig. 1. Experimental setup. A: compartment A, B: compartment B.

## Results

No significant difference was found among the treatments in the total plant biomass, shoot biomass, root biomass, shoot/root ratio, plant water content. The presence of the Collembola decreased both the $^{15}N$ atom percent excess (Fig. 2) and the total N (Fig. 3) of the maize. Collembola activity decreased the hyphal length in compartment A compared to the other treatment (Fig. 4), while in compartment B the hyphal length increased. The presence of Collembola decreased the microbial biomass in compartment A while in compartment B there was no significance difference among the treatments in this respect (Fig. 5).

## Discussion

AMF hyphal length were between 4.25 and 6.85 m g soil$^{-1}$. Similar result were found in an other experiment with *G. intraradices*, *G. caledonium* and *G. invermaium* (Larsen, Jakobsen, 1996). These authors found that Collembola reduced the AMF hyphal length by 14, 37 and 39% depend on the AMF species. In our experiment with 20% lower hyphal length were found in the presence of Collembola. These results are in line with the observations that Collembola at a higher density had negative effect on the growth of AMF (Bakonyi

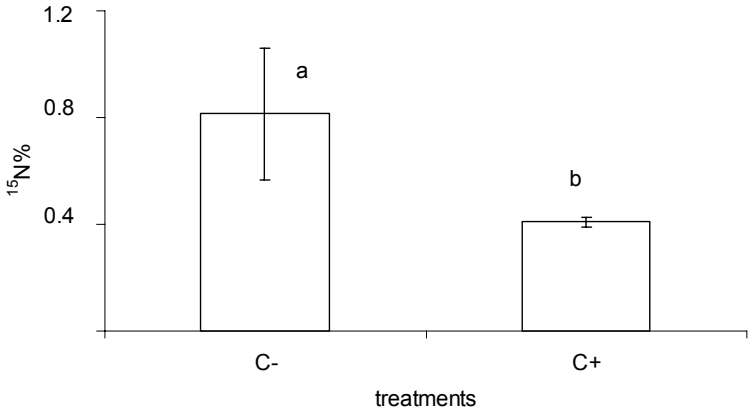


Fig. 2. Average (± SD; n = 3) $^{15}N$ atom percent excess (%) in the treatments. Different letters represent significant differences at $p < 0.05$.

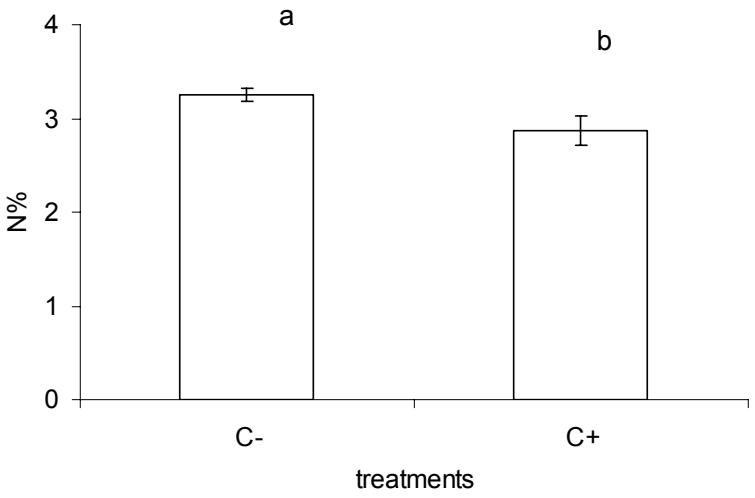


Fig. 3. Average (± SD; n = 3) N content of the maize (%) in the treatments. Different letters represent significant differences at $p < 0.05$.

et al., 2002; Warnock et al., 1982) and the microbial biomass (Kaneda, Kaneko, 2002). Other authors stress the role of saprophytic fungi because evidence shows that Collembola prefer conidial fungi over AMF (Klironomos, Kendrick, 1996). The $^{15}N$ uptake by maize plants was possible through AMF hyphal network under our experimental circumstances, because usually ammonium ions have low mobility in soil. Collembola destroyed hyphal

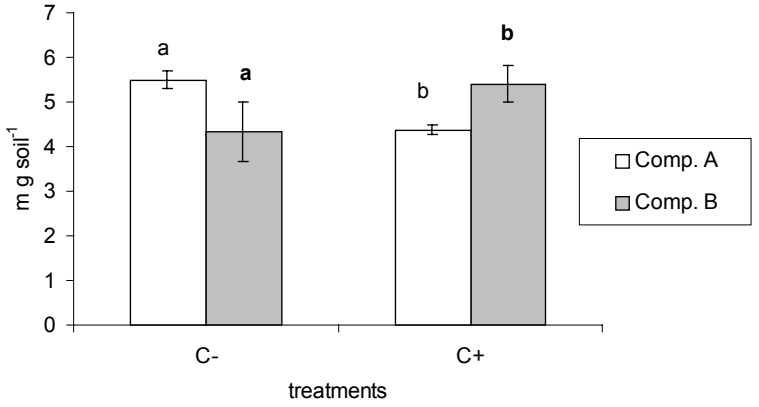


Fig. 4. Average (± SD; n = 3) hyphal length (meter g soil$^{-1}$) in the two compartments of the experimental box. Different letters represent significant differences at $p < 0.05$. (Normal letters refers to compartment A, bold letters to compartment B).

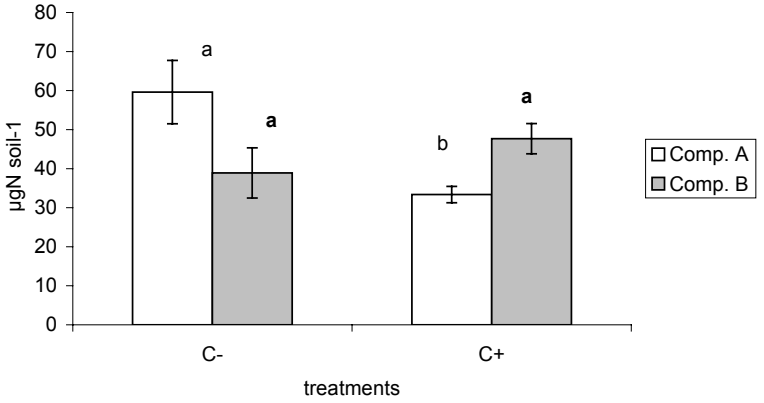


Fig. 5. Average (± SD; n = 3) microbial biomass N in the different treatments. Different letters represent significant differences at $p < 0.05$. (Normal letters refers to compartment A, bold letters to compartment B).

network as it is proved by decreased hyphal length. As a consequence, the uptake of the marked ammonium nitrogen by plants decreased significantly. Cole et al. (2004) found that microarthropods influence the microbial community, but had no effect on the microbial or plant uptake of N. However one species (*Ceratophysella denticulata*) reduced root $^{15}N$ capture in monoculture in this experiment too. Our dates support the view that Collembola can feed the AMF in the presence of saprophytic fungi and is able to influence the AMF hyphal length and function (Bakonyi et al., 2002).

*Translated by the authors*

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