

## MYCORRHIZAL COLONISATION CAN BE ALTERED BY THE DIRECT AND INDIRECT EFFECT OF DROUGHT AND SALT IN A SPLIT ROOT EXPERIMENT

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### Introduction

Technical literature data about the effects of saline or sodic soil on the rates of arbuscular mycorrhizal fungal (AMF) colonisation are rather controversial. Salt was reported to inhibit mycorrhizal colonisation, spore germination, hyphal elongation and spore-forming activity (Juniper and Abbott 1993), while on the contrary several halophytes are reported to be heavily colonized by AMF at salt affected sites (Hildebrandt et al. 2000). This relative paradoxon can be partly explained by the manner of the plant-endomycorrhiza symbiosis: a halophyte macrosymbiont host with the microsymbiont endomycorrhiza partner can better tolerate the suboptimal or extreme conditions. The stressbuffer effect of some beneficial microbes in this respect was reported by Biró et al. (2000). To maintain the symbiosis by the macrosymbiont host can be a „profitable investment”, where the assimilates, carbohydrates are generally refunded with macro- and micronutrients by the fungi. The increased plant acquisition capacity or the limitation of salt uptake are all important conditions of the symbiosis in poor quality salt affected soils (Ruiz-Lozano and Azcon 2000; Marschner 1998). After these findings, our hypothesis is that the enhancement of mycorrhizal colonisation in the saline soils can be the result of a communication process between the macro- and microsymbiont partners. As a consequence of this the salt adaptation mechanisms of the halophytes can be related by the increased mycorrhizal colonisation (Füzy et al. 2006). Our aim in this study was to demonstrate this hypothesis in a split-root experiment, where the direct effects of the rhizosphere towards the mycorrhiza fungi can be well separated from the plant-mediated physiological actions. Stress-factors, such as the salt and the drought were examined, which are known to be the most prominent plant-growth-retardation reasons at the salt affected sites.

### Material and methods

A pot experiment was carried out with split-roots in the pots (Figure 1) according to the suggestion of Vierheilig et al. (2000). The pots were filled with steam-sterilized calcareous chernozem soil (Nagyhörcsök) and 5g *Glomus geosporum* inoculum was layered (at 2-3 cm depth) below the white-clover (*Trifolium repens*) seedlings. After the emergence, the plants were grown in a light chamber with 16h light (20000 lx, 25 °C) and 8h dark (16 °C) period. After 8 weeks of growth different treatments were given to the pots: i) *control*, irrigation with 60-60 ml tap water each half of pots, three times a week; ii) *salt-treated*: irrigation with 60-60 ml saline (1% NaCl) water, three times a week; iii) *drought-treated*: irrigation with 60-60 ml tap water, once a week. In this experiment an underneath irrigation was used, therefore the saline-water and the tap-water could not be mixed with each other (see Fig. 1). Four weeks later the mycorrhizal colonisation of

the plant roots was assessed, after staining them with aniline-blue. Among the colonisation parameters the infection intensity (M%) and the arbusculum richness (A%) were examined by the method of Trouvelot et al. (1986). Beside this, a laboratory conductometer was used for measuring the salt concentration in the soil. Data were subjected to one-way ANOVA and  $LSD_{5\%}$  values are shown.

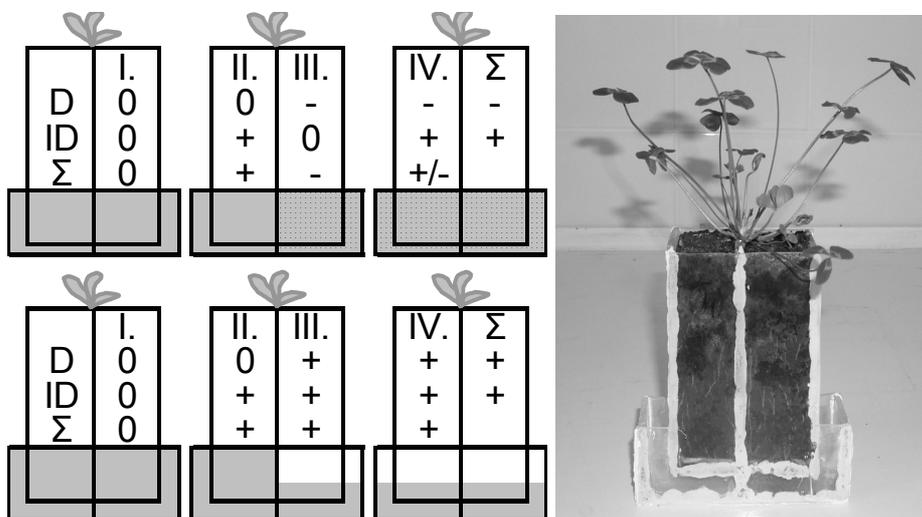


Figure 1. Pots of the split root experiment. The schematic figure and hypothetic effect of salinity (above) and drought (below). D: direct effect of stress to mycorrhiza colonisation, ID: indirect, plant-mediated effects, Σ: summarised effects. More details in the text.

## Results and discussions

### *Clover mycorrhiza interaction affected by the salt stress*

Visible effect of the salt on the plant performance was found at the end of 4<sup>th</sup> week. There were a wilting symptom developed on the whole plant, when both compartment were salted, and only on some leaves at the half-compartment-salted plants. After 4 weeks of saline treatment the EC of the soil increased from  $0,72 \text{ mS cm}^{-1}$  to  $3,52 \text{ mS cm}^{-1}$  (1:2,5 suspension). Colonisation intensity (M%) of AM fungi was found to be above than 60% on the non-salted control (I) and the roots were also containing high level of arbusculum structures (A%). There were no structural obstacle of the nutrient transport between the symbionts at this treatment. No significant difference in the M% or A% values was found between the non-salted “negative” control (I) and the non-stressed split compartment (II). This means, that the role of the host plant in the formulation or the stimulation of the symbiosis could not be demonstrated, when the half or the whole plant had non-stressed normal conditions. Significant differences on the AMF colonisation values was found, between the salt-stressed “positive” control (IV) treatments and at the half-stressed split experiment (III). In those pots the colonisation intensity (M%) of the AM fungi was reduced from 76% to 45%. Regarding the fact, that the treatment is the same for both

rhizosphere, the differences can be related only to the physiological status of the host-plant. The arbusculum richness of the roots at the III pots was especially high (42%), which show that the colonised root segments were heavily arbusculated and functioning in the symbiosis, developed. It is also assumed, that salt-affect could be developed only if the root segments were having a low arbusculum abundance (A%), so the symbiosis was relatively non-functioning.

Table 1. The effect of two environmental stress factors (salinity and drought) on the mycorrhizal colonisation abundance (M%) and functioning (A%) at various treatments in a split root experiment (n=4). Treatments: I – “negative” control, zero stress; II – non-stressed half of a split root; III – stressed half of a split-root; IV – “positive” control, full-stressed treatment (more detail about the level of stress in the text)

<b>Mycorrhizal colonisation at increasing salt stress</b>					
AMF values	I.	II.	III.	IV.	LSD <sub>5%</sub>
M%	63	65	45	76	10,1
A%	46	47	42	47	9,0
<b>Mycorrhizal colonisation at increasing drought stress</b>					
AMF values	I.	II.	III.	IV.	LSD <sub>5%</sub>
M%	39	37	48	31	22,5
A%	32	32	43	30	21,9

#### ***Clover mycorrhiza interaction affected by the drought stress***

No visible differences among plants at the different treatments were found at the end of the 4<sup>th</sup> week. Testplants soon recover without irreversible damage at watering after the drought-stressed periods. There were a continous high water content in the soil at the watered treatment, while it was almost zero in the drought-stressed treatments by the end of each week. Mycorrhizal colonisation intensity (M%) was found to be low both at the non-stressed and stressed (“negative” or “positive”) control, which means that the success of the inoculation in this experiment was limited, only the half of the whole root system was colonised. As the result of one-way variance analyses shows the differences among treatments are not significant, but comparing it with the salt-stress experiment the tendency of the changes in colonisation according to the treatments show the same negative effects. The highest values of AMF colonisation were found at the treatment III, where the drought stress affected the half-part rhizosphere of the hostplant, which was able to aquire enough water throught the other half of the roots (Table 1). The adverse effect in this experiment is reasonable, and it was predictable, as well (see the hypothesis on the Figure 1.). Drought is reported to initiate a direct mycorrhiza-stimulation effect while it is not known at the salt stress (Füzy et al. 2006, Stevens and Peterson 1996).

#### **Conclusions**

It was experimentally demonstrated by a split-root system, that the arbuscular mycorrhizal fungi can have a positive stress-buffer effect at the large environmental stress, like salt and drought. The way of control and function of the plant endomycorrhiza symbiosis is up till now an inadequately explained mechanism. Role of the host plant is

rather neglected in the process, the environmental factors, like the nutrient availability, however is said to be critical (Takács et al. 2006, Villányi et al. 2006). Beside those crucial factors the plant-mediated consequences of the enhanced mycorrhiza formation and functioning at the severe environmental stress could be also concluded from the study.

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