

Natural mycorrhizal colonization of different apricot rootstocks

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Abstract

Mycorrhizae provide a permanent, reliable and natural way to reap the benefits of symbiosis. Since the susceptibility of different apricot rootstocks to mycorrhization varies considerably, tree conditions can be improved to varying degrees. To determine the degree of naturally occurring colonization (low, high and total), we tested two Hungarian scion cultivars ('Gönci magyar kajszai' and 'Ceglédi szilárd') with six rootstock cultivars from different origins. The results were obtained by microscopic analysis of dyed root samples. The rootstocks 'Apricot seedling' and 'Myrobalan 29C' are the most susceptible in our orchards. The insufficient colonization of the plum rootstocks 'Wavit' and 'Fehér besztercei' could be due to the water requirements of plum as a rootstock, which puts them at a disadvantage in non-irrigated conditions. However, no statistical differences were found between the rootstocks in the proportion of uncolonized roots, except that the roots of 'Gönci magyar kajszai' were more colonized than those of 'Ceglédi szilárd'.

Keywords: colonization; mycorrhizae; *Prunus armeniaca*; rootstock; symbiosis

Introduction

Cultivated apricot cultivars belong to the species *Prunus armeniaca* L., which belongs to the genus *Prunus* L. of the Rosaceae family. However, *P. mume* is also grown in China, where apricots originally came from (Vavilov, 1926; 1951). Apricots are the third most important stone fruit in the world, cultivated on 560,000 hectares worldwide, producing more than 4 million tons of fruit each year. Turkey, Iran, Uzbekistan, Italy and Pakistan account for 54% of apricot production (FAOSTAT, 2023). More than 50% of production comes from Asia, followed by Europe (27%) and Africa (14%) (Moustafa and Cross, 2019).

Rootstocks are essential for the budding or grafting process in fruit trees to maintain and propagate valuable genotypes. Rootstocks are crucial due to the spread of grafting, as noted by Taaren *et al.* (2016) and Pászti and Mendel (2018). The interaction between rootstock and scion benefits the grafted tree. Apricot rootstocks are evaluated primarily on the basis of their compatibility with scion cultivars and their adaptability to orchard soil conditions. Rootstock characteristics, including pest and disease tolerance and high soil lime content, are beneficial for apricot production in locations where scion cultivars may not thrive. Rootstock also

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influences growth vigor, tolerance to biotic and abiotic soil factors, flowering phenology, fruit quantity, and fruit quality (Beckman *et al.*, 1992; Boyhan *et al.*, 1995; Duval *et al.*, 2012). Roots are responsible for water and nutrient uptake and adaptation to ecological conditions (Darikova *et al.*, 2019). Adaptation of roots to edaphic conditions is essential for economic fruit growth. Several rootstock characteristics contribute to maintaining orchard health, and their suitability for mycorrhizal colonization is one factor (Mendelné *et al.*, 2023).

Arbuscular mycorrhizal fungi develop in roots and extend into the soil. These associations allow the hyphae to assimilate minerals and water for transport to host plants (Allen and Allen, 1980; Augé, 2001; Püschel *et al.*, 2008; Kaschuk *et al.*, 2009). The benefits of arbuscular mycorrhizal fungi include promoting faster plant growth, improving plant health, and mitigating the negative effects of external factors such as weather damage, poor soil conditions, pathogens, and pests on plants (Smith *et al.*, 2010; Cruz *et al.*, 2014; Bi *et al.*, 2021). In addition, these fungi are capable of forming symbiotic relationships with 90% of terrestrial plant species. Numerous fungal species can form associations with plant roots. However, the *Phylum glomeromycota* is the most important in agriculture in general, while *Entoloma clypeatum* is the most common in fruit trees (Szücs and Véghegyi, 1996; Schussler *et al.*, 2001). Artificial inoculation typically improves mineral uptake and overall fruit tree performance as well as stress tolerance (Mohammad *et al.*, 2003; Dutt *et al.*, 2013a; 2013b; Santoyo *et al.*, 2021). The symbiosis improves the supply of water and nutrients such as phosphate and nitrogen to the host plant. In return, some of the plant's fixed carbon (in the form of carbohydrates) is transferred to the fungus. This transfer occurs through symbiotic structures inside root cells called arbuscules. The development of AM is accompanied by an exchange of signalling molecules between the symbionts: strigolactones from the host and "Myc factors" from the fungus. Both are essential to maintain coexistence (Bago *et al.*, 2003; Parniske, 2008). Symbiosis is characterized by complex patterns of gene expression changes in plant roots. The up- and down-regulation of genes allows adaptation to the metabolism of each partner. This process is specific and tightly regulated at the molecular level (Liu *et al.*, 2007).

Previous studies during the dormant season have revealed significant differences in colonization rates among scion cultivars of several apricot species. These observations were mainly made during winter preparation and early spring (Bakos *et al.*, 2019).

Materials and Methods

The region of the trial (GPS coordinates: N 47°10'35.0, E 19°50'28.6) is characterized by a temperate continental climate with a semi-arid microclimate. The 665 trees/ha were planted in Chernozem soil. After planting, a living mulch was placed between the rows and drip irrigation was applied four times during the vegetative period, with 40 mm of irrigation each time. At least six replicates for each rootstock-cultivar combination were included in two randomly assigned blocks. The rootstocks analyzed include 'Apricot seedling' (*Prunus armeniaca* L.), which is widely used in cultivation, and 'Fehér besztercei' (*Prunus domestica* L.), which is registered in the national variety register of Hungary (Hrotkó, 1999). 'Wavit' (*Prunus domestica* L.), derived from clonal lines of the Austrian Wangenheim plum, has been observed to reduce growth vigor in many cases (Yordanov *et al.*, 2015). 'Wavit', similar to 'Fehér besztercei', is known for its uniform orchards and reduced vigor (Missere *et al.*, 2010). 'Myrobalan 29C' (*P. cerasifera myrobalana* L.) is particularly suitable for soils with a high lime content and offers a good adaptation (Foschi *et al.*, 2012). The experiment also includes 'Montclar' (*Prunus persica* L.), which has strong growth and good yields, but may have compatibility problems (Irisarri *et al.*, 2021). The Spanish myrobalan-almond hybrid 'Rootpac R' (*P. cerasifera* × *P. dulcis* Mill.) showed similar traits to 'Monclar' and 'Apricot seedling', with remarkable adaptability to suboptimal soil and climatic conditions and strong vigor (Surányi, 1999; Bassi *et al.*, 2006; Pinochet *et al.*, 2010; Agromillora, 2023).

The vigorous ‘Gönci magyar kajsz’ and the more moderate ‘Ceglédi szilárd’ were used as scion cultivars (Mendelné and Mendel, 2021; Mendelné *et al.*, 2022). The two scion varieties with different growth habits were grafted on six rootstocks as shown in Table 1.

Table 1. The six apricot rootstocks and their genetic background, grafted with ‘Gönci magyar kajsz’ and ‘Ceglédi szilárd’ scions, planted at the Cegléd Research Station, Hungary

Rootstock cultivar	Species
‘Wavit’ (Wv)	<i>Prunus domestica</i> L.
‘Fehér besztercei’ (Fb)	<i>Prunus domestica</i> L.
‘Montclar’ (Mc)	<i>Prunus persica</i> L.
‘Rootpac R’ (RR)	<i>P. cerasifera</i> myr. X <i>P. dulcis</i> Mill.
‘Myrobalan 29C’ (My)	<i>Prunus cerasifera myrobalana</i> Ehrh.
‘Apricot seedling’ (As)	<i>Prunus armeniaca</i> L.

Root samples were collected monthly, from March to October, at the MATE Research Station in Cegléd. Each tree was sampled on each occasion, and at least three root zones of each rootstock-rootstock combination were obtained. Mycorrhizal colonization was evaluated through microscopic analysis of root samples. Root samples were prepared and analyzed using the following technique: the samples were first washed with tap water and cleaned, then stained with aniline blue dye (Grace and Stribley, 1991). Next, KOH solution was used to remove the stain from the plant tissues (Phillips and Hayman, 1970). The samples were then mounted on slides and examined at five different points. Information on each root-root combination was obtained by analyzing 30 samples at 150 points. Hyphae, vesicles, and arbuscules were observed at each testing point to determine their presence and abundance. Three potential states were identified: no colonization (no visible mycorrhizal formations), weak colonization (few scattered formations visible), and strong colonization (numerous formations visible throughout the field of view). Following the examination of all samples, the degree of colonization was expressed as a percentage. Only natural mycorrhizal colonization was investigated; fungal species were not identified or isolated.

The statistical analysis involved conducting a one-way univariate analysis of variance (ANOVA) on the data, followed by separating the vegetative properties with the Games-Howell post-hoc test. For these procedures, the IBM SPSS v.27 software was utilized.

Results and Discussion

Since mycorrhizal colonization increases the surface area through which roots absorb water and nutrients, higher colonization values are more beneficial for plants. Arbuscular mycorrhizas form vesicles, or hyphae, which increase the surface area available for nutrient and water uptake during the symbiosis (Figure 1). The fibrillar hyphae are directly responsible for uptake, while the globular vesicles are persistence structures.

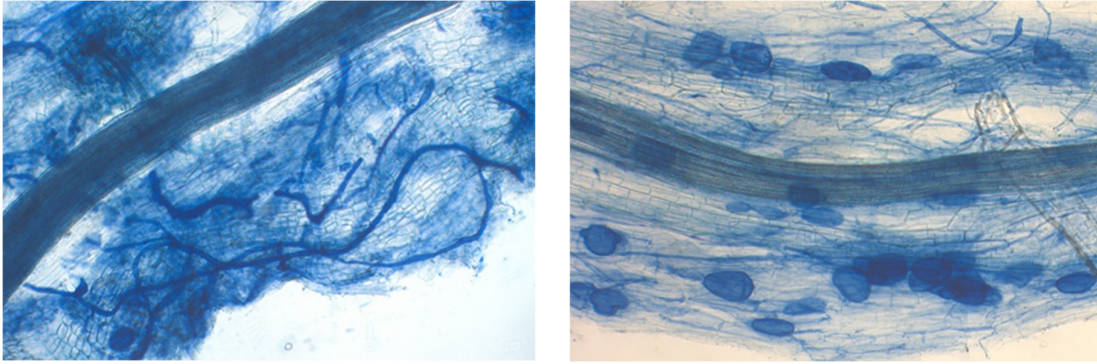


Figure 1. The fibrillar hyphae (left) and the globular vesicles (right) on roots of ‘Apricot seedling’. Magnification: $\times 10$

The study revealed remarkable differences between the rootstocks. Figure 2 displays the average of root samples from both cultivars analyzed.

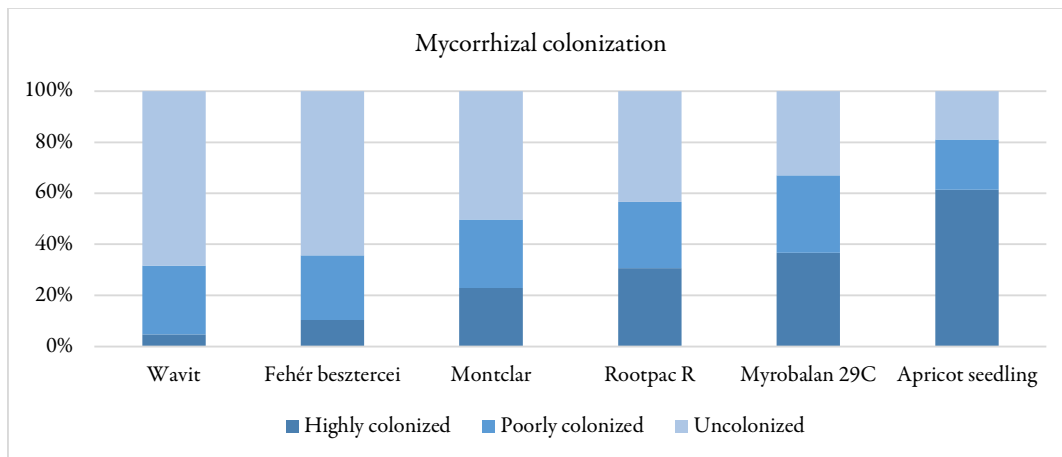


Figure 2. Percentage of mycorrhizal colonization of the six tested rootstock cultivars, given in the mean of the two investigated cultivars

The highest proportion of uncolonized roots was found on ‘Wavit’ (68%), which was not significantly different from ‘Fehér besztercei’ (64%). In 50% of the roots of ‘Montclar’ and in 43% of the roots of ‘Rootpac R’ no mycorrhiza was observed. Meanwhile, 33% of the roots of ‘Myrobalan 29C’ were recognized as uncolonized, while ‘Apricot seedling’ showed the best result in this aspect (19%). These values were significantly different at $p < 0.05$ level. The proportion of roots with low colonization was lowest on ‘Apricot seedling’ (20%) and highest on ‘Myrobalan 29C’ (30%). The other four rootstocks fell between the two extremes (25-27%) and did not differ significantly from each other. Greater differences were observed in the proportions of roots with high colonization. ‘Wavit’ and ‘Fehér besztercei’ belonged to the less colonized group (5% and 10%, respectively). ‘Montclar’ (23%), ‘Rootpac R’ (31%) and ‘Myrobalan 29C’ belong to the intermediate group of rootstocks. Trees on ‘Apricot Seedling’ had the highest percentage of well colonized roots, reaching 61%.

The plum-based rootstocks ‘Wavit’ and ‘Fehér besztercei’ do not support the growth of all apricot cultivars and have weak growth habits. They require the most hydrated soils compared to other tested rootstocks (Nagy and Lantos, 1996; Sitarek and Bartosiewicz, 2011), which makes them unsuitable for proper mycorrhizal symbiosis. Although ‘Montclar’ and ‘Rootpac R’ are known for their vigorous growth (Mendelné *et al.*, 2022) and ability to survive in poor soil conditions due to their pedigree (Irisarri *et al.*, 2021; Agromillora,

2023), they have lower colonization rates compared to the best performing 'Apricot seedling'. However, they have the best colonized and least uncolonized roots in this study. Although 'Myrobalan 29C' shows promising results, it falls considerably short of the best performing cultivar. Genetically, it is very similar to the apricot to which it is grafted, resulting in a closely matched metabolism (Mowrey and Werner, 1990; Badenes and Parfitt, 1995). The metabolic balance facilitates the symbiotic relationship with the roots.

There are differences between the colonization of the roots of the weak-growing 'C. szilárd' and the vigorous 'Gönci m. k.', as shown in Table 2. The values are very similar for the two scion cultivars, and the mean values of the rootstocks are also basically the same. There is a larger difference for two rootstocks: 'Wavit' has a higher colonization rate with 'Gönci m. k.', 'Rootpac R' with 'C. szilárd'. For the other four rootstocks, the scions do not differ significantly from each other.

Table 2. Percentage of mycorrhizal colonization of the two cultivars grafted on six rootstocks

Rootstock cultivar	Uncolonized		Poorly colonized		Highly colonized	
	C. szilárd	Gönci m. k.	C. szilárd	Gönci m. k.	C. szilárd	Gönci m. k.
'Wavit' (Wv)	73	64	25	29	2	7
'Fehér besztercei' (Fb)	63	65	25	25	11	9
'Montclar' (Mc)	53	48	28	25	19	27
'Rootpac R' (RR)	36	51	33	19	31	30
'Myrobalan 29C' (My)	37	29	41	19	22	51
'Apricot seedling' (As)	21	17	27	13	52	71
Mean	47	46	30	22	23	33

The average of the root samples shows that 'C. szilárd' is more poorly colonized, while 'Gönci m. k.' is more highly colonized in the average of the six rootstocks. 'Wavit' showed a higher total colonization with 'Gönci m. k.' in all aspects. 'Fehér besztercei' showed no significant differences. 'Montclar' showed less uncolonized and more highly colonized roots in combination with 'Gönci m. k.'. 'Rootpac R' was better in this comparison with 'C. szilárd'. The results of 'Myrobalan 29C' were similar to those of 'Apricot seedling'. Fewer uncolonized and poorly colonized roots and more highly colonized roots with 'Gönci m. k.'. 'Apricot seedling' showed better results of the two rootstocks, but 'Myrobalan 29C' is the closest of all tested.

Cultivar, root morphology, and phosphorus uptake have strong effects on mycorrhizal colonization (Tawaraya, 2003). The effect of cultivar has also been demonstrated for *Triticum aestivum*, *Lycopersicon esculentum*, *Allium fistulosum* and *Citrus* species (Menge *et al.*, 1978; Azcón and Ocampo 1981; Bryla and Koide 1990; Tawaraya *et al.*, 1999). The change in mycorrhizal colonization of the rootstock in response to the apple cultivar grafted onto it has recently been shown (Przybyłko *et al.*, 2021), but no data are available in relation to other fruit species.

Conclusions

Because mycorrhizal colonization is a highly symbiotic relationship, the free, two-way flow of nutrients and information is beneficial to both partners. The more extensive the mycelial tissue, the more effective the cooperation and the greater the impact on the overall health of the plant. Because the metabolism of the two components of the grafted tree differ to varying degrees, this flow can be disrupted and unfavorable conditions can develop. The smallest difference in metabolism occurs when individuals of the same species are combined (e.g., apricot scion grafted onto apricot rootstock). The more distantly related the rootstock, the greater the difference and the weaker the mycorrhizal colonization. According to our present studies, rooting of plum rootstocks under the edaphic conditions of the experimental plantation is not suitable for apricot. The roots of 'Apricot seedling' were the most mycorrhizal colonized (both poorly and highly), 'Wavit' was the least. Based

on our results, 'Apricot seedling', 'Myrobalan 29C' and 'Rootpac R' are the most suitable for the apricot scion cultivars, in that order. The effect of the scion variety on the formation of mycorrhizae in the root system of the rootstock is a very complex task. There are not many test results on this subject and the effect is not very clear. The vigorously growing 'Gönci magyar kajszi' shows a better susceptibility to high colonization, while the moderately growing 'Ceglédi szilárd' shows a higher proportion of poor colonization. Every tested rootstock showed more affinity in combination with 'Gönci Magyar kajszi', except for 'Rootpac R'.

Consideration of the specific scion cultivar and its compatibility with mycorrhizal fungi can help maintain the orchard. Some rootstock cultivars may benefit more from mycorrhizal associations than others, depending on their nutrient requirements and local soil and environmental conditions, but this area is not well understood. Soil tests and assessment of mycorrhizal colonization levels are needed to clarify the effects of mycorrhizae on apricot rootstock growth and fruit production.

Authors' Contributions

The contributions of authors to the manuscript should be specified in this section; according to the type of Conceptualization, Á.M. and E.M.P.; methodology, L. Sz. and J. L. B.; formal analysis, J. L. B.; investigation, L. Sz.; writing—original draft preparation, Á.M. and E.M.P.; writing—review and editing, Á.M.; supervision, L. Sz.; All authors read and approved the final manuscript.

Ethical approval (for researches involving animals or humans)

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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