Chapter 18

Alterations in Root Morphology of Rootstock Peach Trees Caused by Mycorrhizal Fungi

José Luis da Silva Nunes, Paulo Vitor Dutra de Souza, Gilmar Arduino Bettio Marodin, José Carlos Fachinello and Jorge Ernesto de Araújo Mariath

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/51586

1. Introduction

The morphology of plant roots have gained prominence in various branches of knowledge, especially in the Biological and Agricultural Sciences, according to the same being one of the main features of the plant body related to the supply and support of plant (Marschner, 1995). Agricultural practices of soil management require special attention in the relations of the roots of different plants with different managements employees, because the health of plants is dynamically linked to these delicate relatio (Silva et al., 2005). This is because the management practices linked monocultures allow the reproduction of micro-organisms that cause crop damage, and the common use of pesticides to alleviate this problem (Bressan & Vasconcelos, 2002).

In this sense, studies are being conducted with the objective of evaluating the possibility to reduce the use of these chemicals in the control of harmful micro-organisms, ranging from research on structural strength of the plant, past the front of the dynamic plant managements, to the use of microorganisms considered beneficial plants (Bressan & Vasconcelos, 2002). On this last point, the arbuscular mycorrhizal fungi (AMF) colonize the root system of most plants, and one of the most reported benefits has been a greater phosphorus absorption by the mycorrhized plants (Nunes et al, 2006), forming a mutualistic symbiosis type biotrophic (Dodd, 2000). This symbiosis is widely distributed in the plant kingdom, occurring in 83% of dicotyledonous plants, in 79% of monocots and in all Gymnosperms, without altering the external appearance of the root (Wilcox, 2002). Moreover, the occurrence of symbiosis is widespread in most habitats, both natural ecosystems and in ecosystems altered by human activities (Sylvia et al., 2001).

In this respect, mutualism is manifested in the bidirectional exchange of nutrients, where the plant comes from carbohydrates to the fungus, while it provides you with water and nutrients, especially for the case of phosphorus (Smith et al., 2003). Although the result of symbiosis be beneficial for the phytobionts, the effectiveness varies in function of the combination the vegetal species and fungus involved in the association (Smith et al., 2003).

By mechanisms promoted by the AMF, the external hypha and mycelia increase the root capacity to exploit the soil results in greater nutrient absorption (Siqueira et al., 2002). However, this absorption has also been related to alterations in the morphological properties of the root of the host plant (Moreira & Siqueira, 2002).

The root system morphology is determined genetically, and can vary among species and individuals in function of environmental factors, such as water availability, nutrients and temperature (Tokeshi, 2000) and the plasticity of the root system can also be influenced by AMF (Berta et al., 1995). The root morphology influences the fast development of the root system and is critical for the successful establishment of most horticultural and fruit plants (Bressan & Vasconcellos, 2002).

This fact is fundamental to the understanding of the effects of the AMF on root development, especially in the case of rootstock plants (Berta et al., 1995). However, the relationships involved in the formation of this symbiosis, since the signaling between the phytobionts, the early stages of the colonization process, as well as possible alterations in the morphological structure of the roots (Berta et al., 1995), in order to be considered a complete understanding relations between the symbionts

There is little information about such relationships, as well as morphological changes produced by mycorrhizal infection in plant tissues (Souza et al., 2000). Some authors report that the AMF does not cause major morphological changes in roots (Cooper, 1984), but studies showed that the AMF induces changes in the architecture (Berta et al., 1995; Norman et al., 1996), especially in the increase of the root ramification, in the morphology (Berta et al., 1995; Bressan & Vasconcelos, 2002; Kothari et al., 1990; Norman et al., 1996,) and the anatomy (Berta et al., 1995) the roots of different plant species.

Most infections of the root system of plants by soil microorganisms imply relations between the actors involved, these relations are based on compatibility between symbionts or the ability of the microorganism to overcome the defense mechanisms of plants (Paszkowski, 2006). The study of morphological relationships between the symbionts highlight the determinants of compatibility that allow the symbiosis occurs involving taxonomically distinct groups of plants and AMF infective (Panstruga, 2003).

The objective of this study was to relate the morphology and root system development of plants of the rootstock cultivars of the peach trees Aldrighi and Okinawa with root colonization by AMF species and the influence of this relationship on nitrogen, phosphorus and potassium absorption and the vegetative development of the plants.

2. Material and methods

2.1. Execution area

The study was carried out under shading (Okinawa cultivar) and a greenhouse (Aldrighi cultivar) at the UFRGS Agronomic Experimental Station, county of Eldorado do Sul, RS, located at latitude 30° 05' South and longitude 51° 39' West from 2004 to 2005.

2.2. Plant and fungal material

Seeds from the two rootstock cultivars were stratified in sterilized sand and placed in a refrigerator at 4°C for 45 days to break the seed dormancy.

Afterwards the seeds were sown on a bed of sterilized sand in a greenhouse. When they were about 5 cm long, the seedlings were replicated to 5 liters black plastic bags containing substrate consisting of clay soil, sand with medium particle size and decomposed black acacia bark residue (1:1:1, V:V.V). The substrate was previously disinfected with formaldehyde solution at 10%.

The AMF species tested were *Acaulospora sp.* (Trappe), *Glomus clarum* (Nicol. and Schenck) and *Glomus etunicatum* (Becker and Gerd) for the Okinawa cultivar and for the Aldrighi cultivar the same treatments were tested along with *Scutellospora heterogama* (Nicol. and Gerd.). The AMF species were inoculated by adding to each plastic bag 30 g of roots and rhyzospheric soil of oregano (*Origanum vulgare* Link) containing AMF structures. The inoculum was placed in a layer situated in the mid-part of the recipient.

A randomized block design was used, with 20 plants per plot and four replications, in a total of 320 plants for the Okinawa cultivar and 400 plants for the Aldrighi cultivar.

2.3. Determination of roots colonization and plant responses

When the plants had diameter for grafting (360 days for the Okinawa cultivar and 180 days for the Aldrighi cultivar) the height was assessed of the 20 plants in each plot, from the root-stem junction to the tip of the main stem, using a measuring tape, and the main stem diameter, at the root-stem junction and plant height using a pachymeter.

In addition, 5 plants were used from each replication of the treatments, for determination of leaf area, through the use of leaf area meter mark Li-Cor (LI - 3000). After, the shoot was dried and ground and where the fractions were removed for evaluation of plant tissue nitrogen, phosphorus and potassium content by digestion, distillation and spectrophotometry flames, following the methodology by Tedesco et al. (1995).

Five second order roots with similar length and diameter were collected from the root system to assess the root colonization rate (by the ratio number of infected segments/total

analyzed). To determine the colonization rate the radicels were stained following methodology reported by Phillips and Hayman (1970).

2.4. Determination of reserve substances

Samples of the aerial part (leaves, stems and stem) and dried roots were ground in the mill, coupled with a sieve of 20 meshes per inch. Each sample was collected approximately one gram for determination of reserve substances.

A similar procedure was carried out with samples of roots. After each sample individually packaged in bags made of special screen for the filtration of food products and brought back to 65°C oven to constant weight, recording the weight of each bag, after, were digested in order to extract all components of plant tissue (carbohydrates, fats, fatty acids, etc.) that were not fibers (cellulose, hemicellulose and lignin), as conventionally known as reserve substances the method described by Priestley (1965).

The samples were placed in one liter Erlenmeyer flask containing an aqueous solution with 5% trichloroacetic acid (99%) and 35% methanol (99.8%) remained on heating gas burner, under a hood with hood, by eight hours. From the third hour to eight hours, distilled water was added to the solution, as it would evaporate in order to always maintain the same volume of liquid sufficient to maintain the samples immersed in the solution.

After the samples were rinsed with distilled water again and put in stove to dry at 65°C until constant weight. The difference in mass of the samples before and after digestion constisted substance content of the buffer that contained samples.

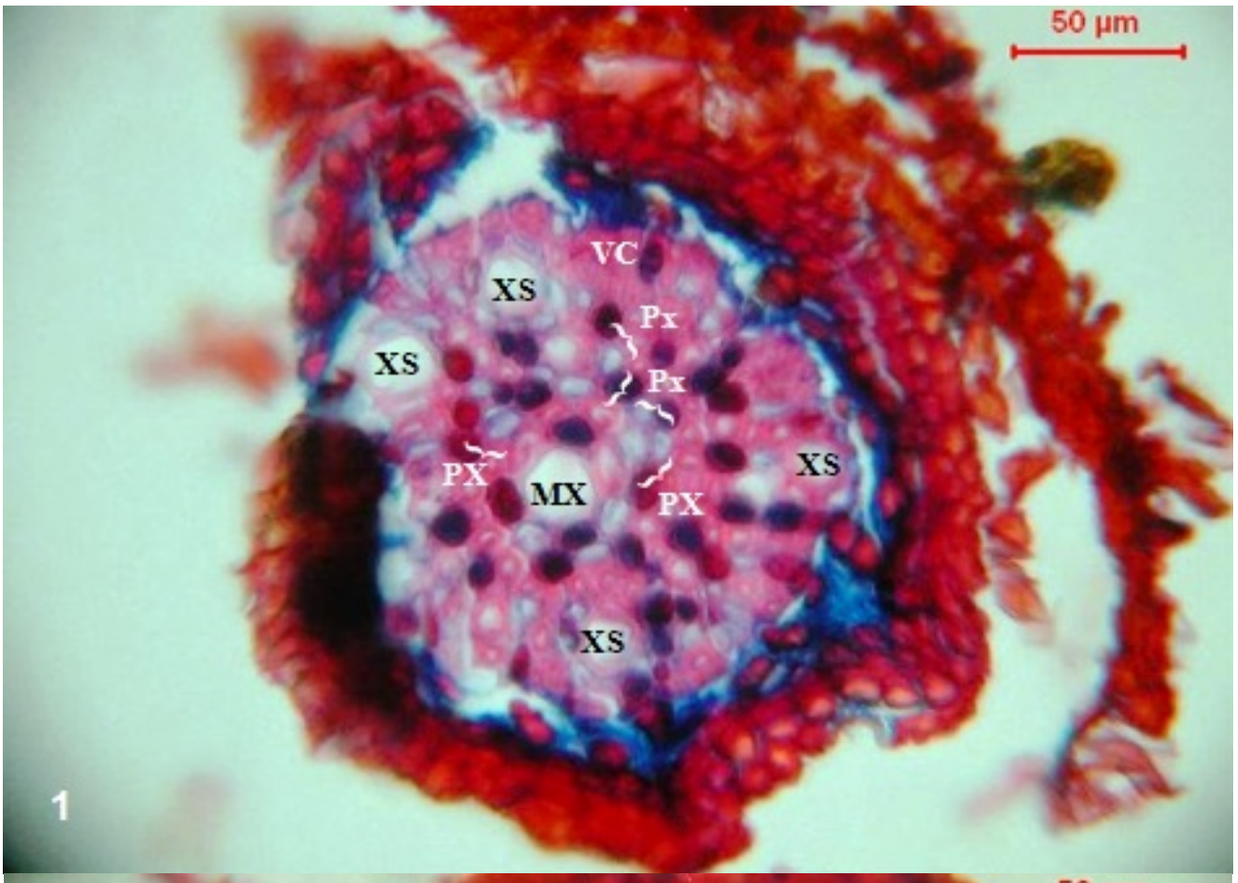
2.5. Histological studies

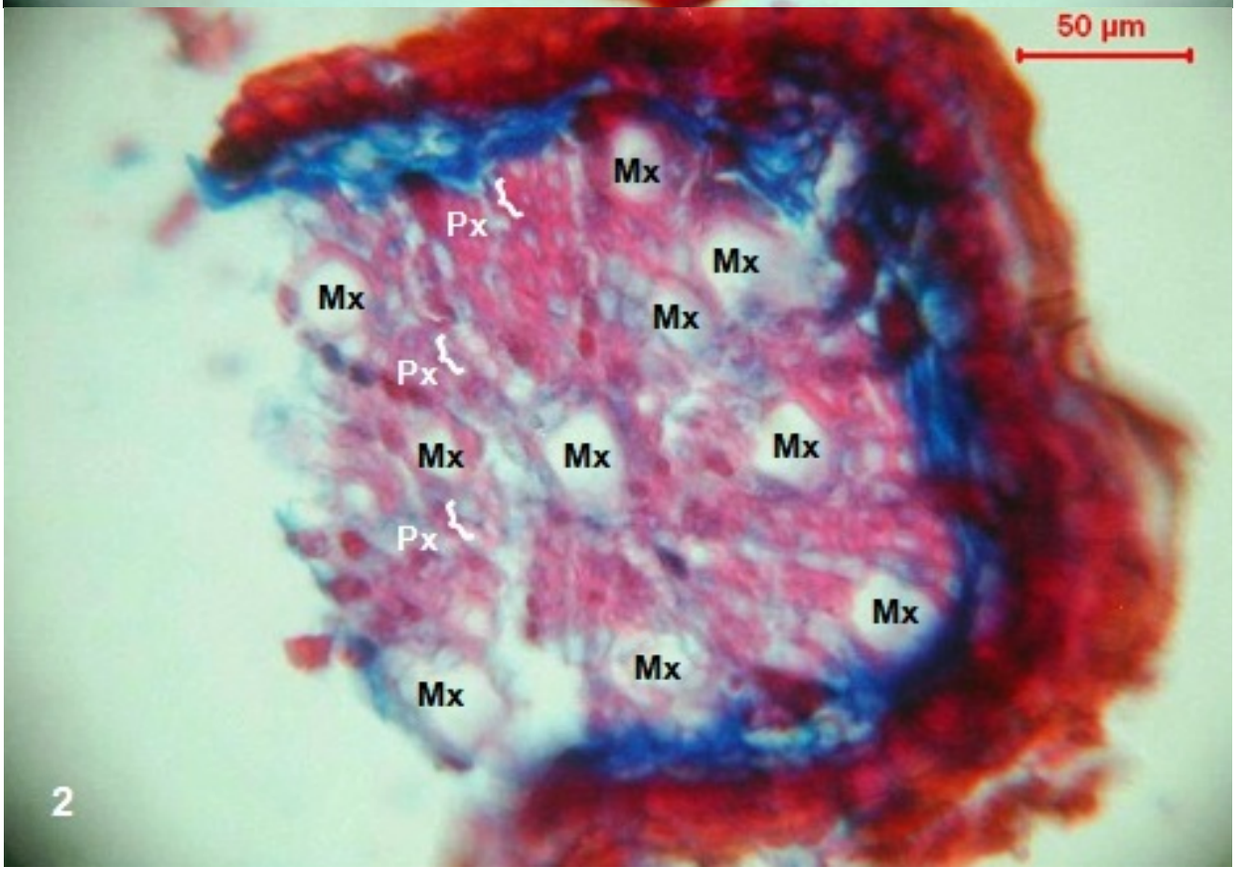
Secondary roots with similar diameter were used for the morphological studies, as shown in Tables 1 and 2. The histological studies followed the methods described by Johansen (1940), where 1 cm long samples were dehydrated and blocked in paraffin, and 10-15μm thick slices were made using a manual microtome.

The slices were placed on slides, removed the paraffin with xylol, rehydrated for later staining with aqueous Safranin (1%) and Toluidine Blue O (0.05%), and than dehydrated again and the preparations mounted in Canada balsam with a coverslip.

These sections were observed under a Leica DM microscope with 400X magnification. The images were captured with a Nikon CoolPix 990 digital camera (Photos of José Luis da Silva Nunes) and analyzed using the "WCIF Image J" software.

The morphometric parameters measured in the roots were area, diameter, number and perimeter of the tracheal element cells, regardless of the stage of ontogenetic development (primary or secondary) and, from the primary xylem, only the metaxylem was measured, because the protoxylem collapsed at the end of its differentiation (Figures 1 and 2).


50 µm
VC
XS
Px
XS
Px
PX
MX
PX
XS
XS
1


50 µm
Mx
Px
Mx
Mx
Mx
Px
Mx
Mx
Mx
Px
Mx
Mx
Mx
2

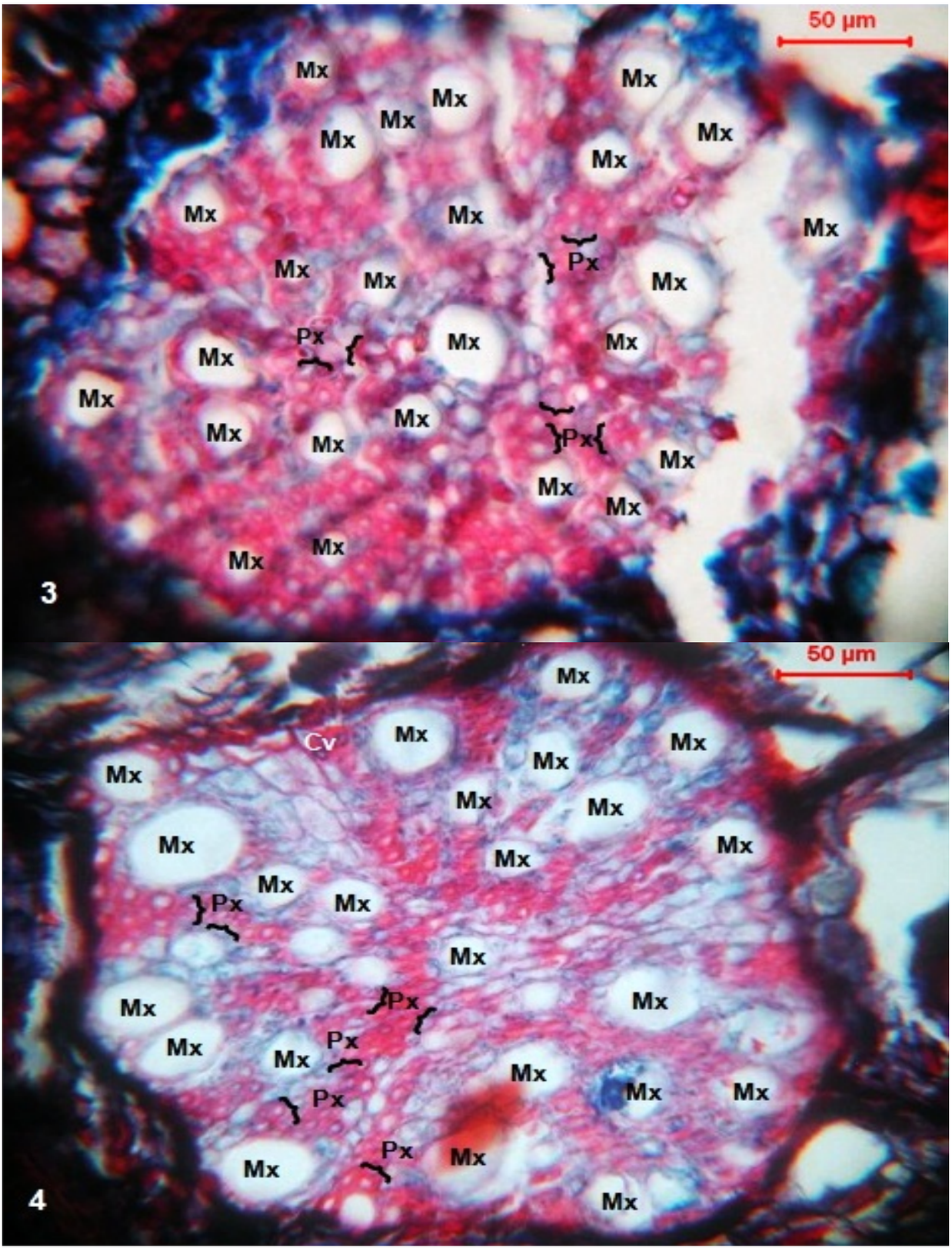
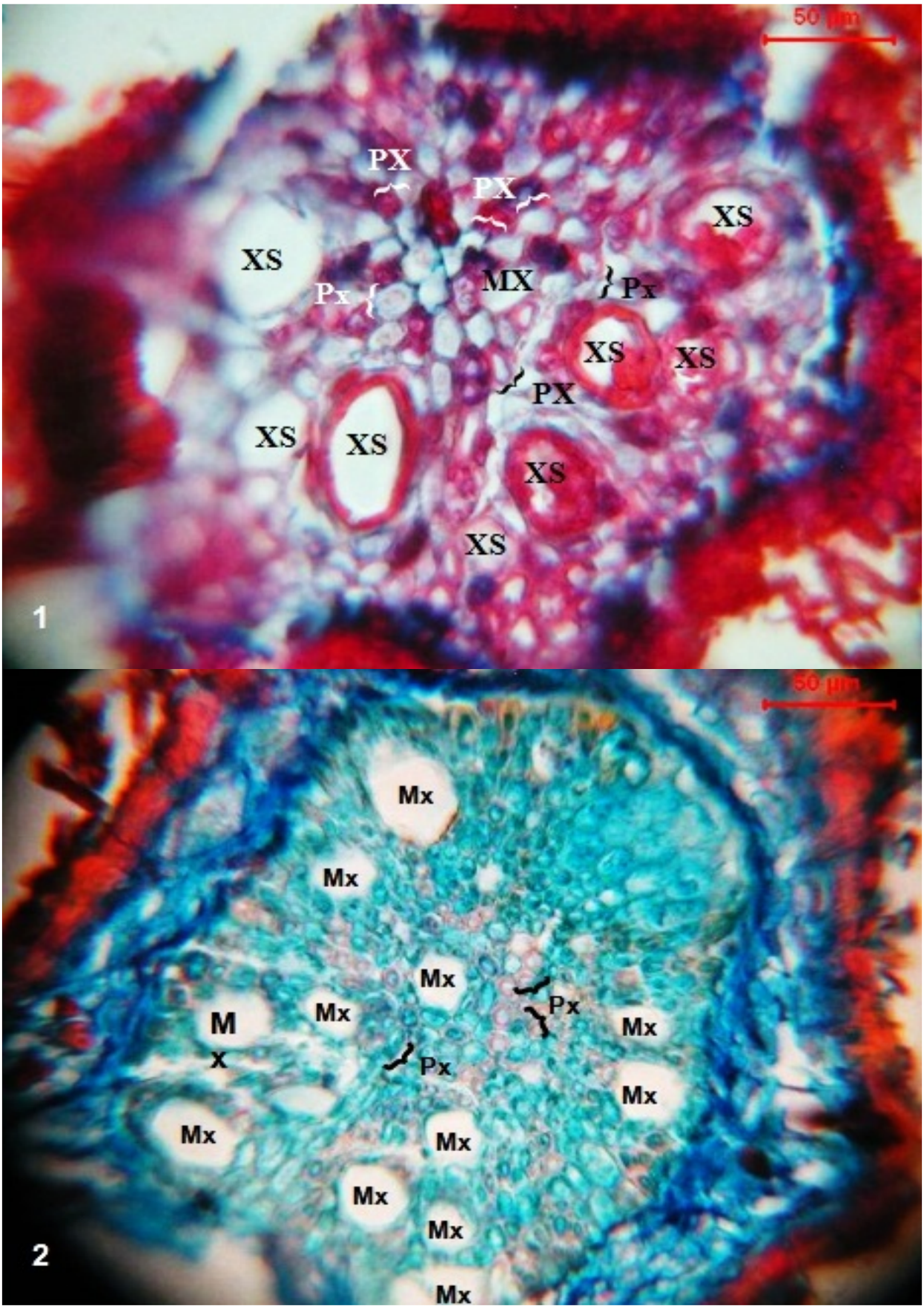
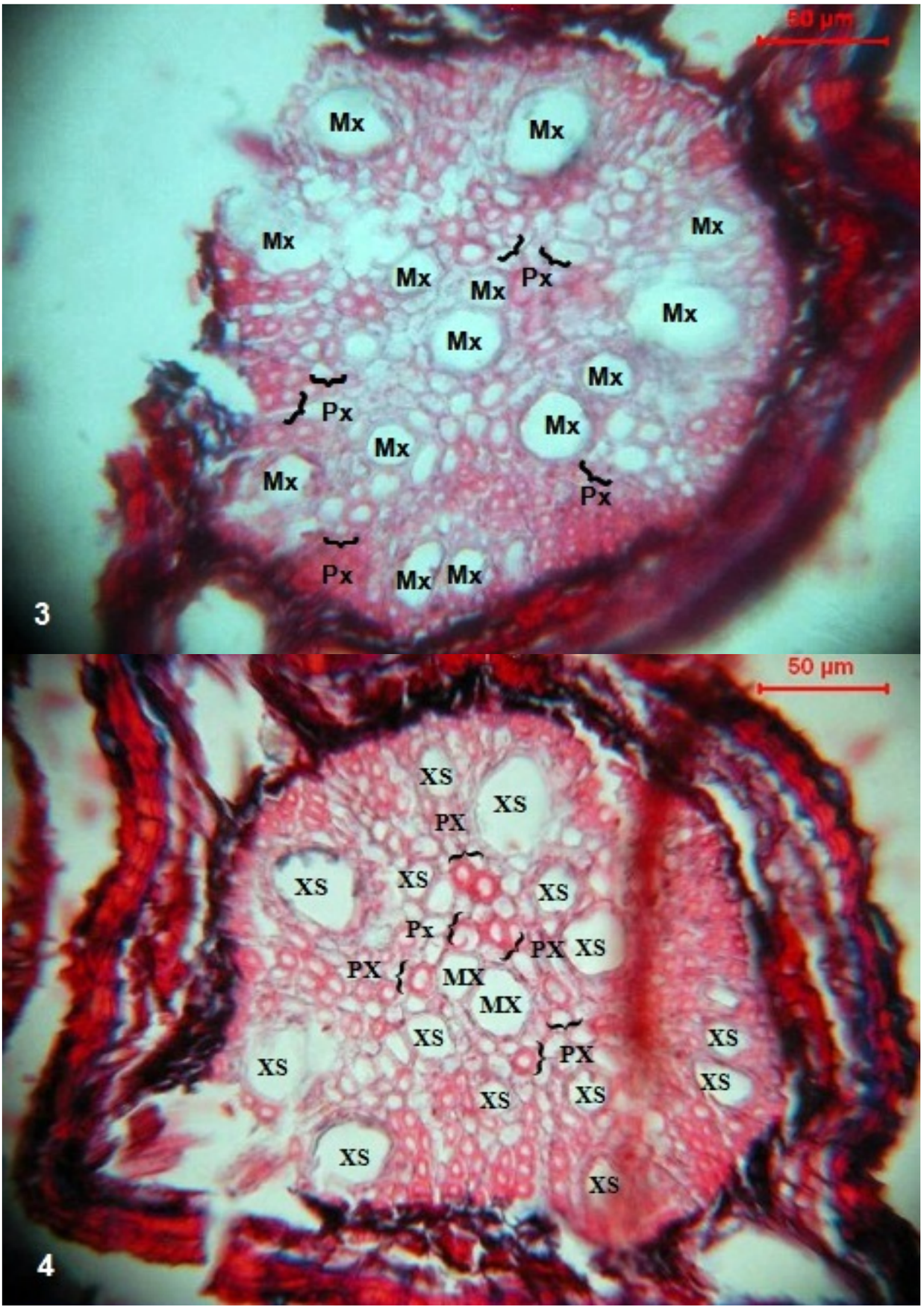


Figure 1. Cross sections of secondary roots of peach tree rootstock Okinawa without inoculation (1) and inoculated with AMF (2 – *G. clarum*, 3 – *G. etunicatum*, 4 – *Acaulospora sp*.). MX – metaxylem; PX – protoxylem; XS – secondary xylem. Scale 50 µm.


50 µm
PX
PX
XS
XS
MX
Px
Px
XS
XS
PX
XS
XS
XS
XS
1
50 µm
Mx
Mx
Mx
Mx
M
x
Px
Px
Mx
Mx
Mx
Mx
Mx
Mx
Mx
2


50 µm
Mx
Px
3
50 µm
XS
PX
MX
4

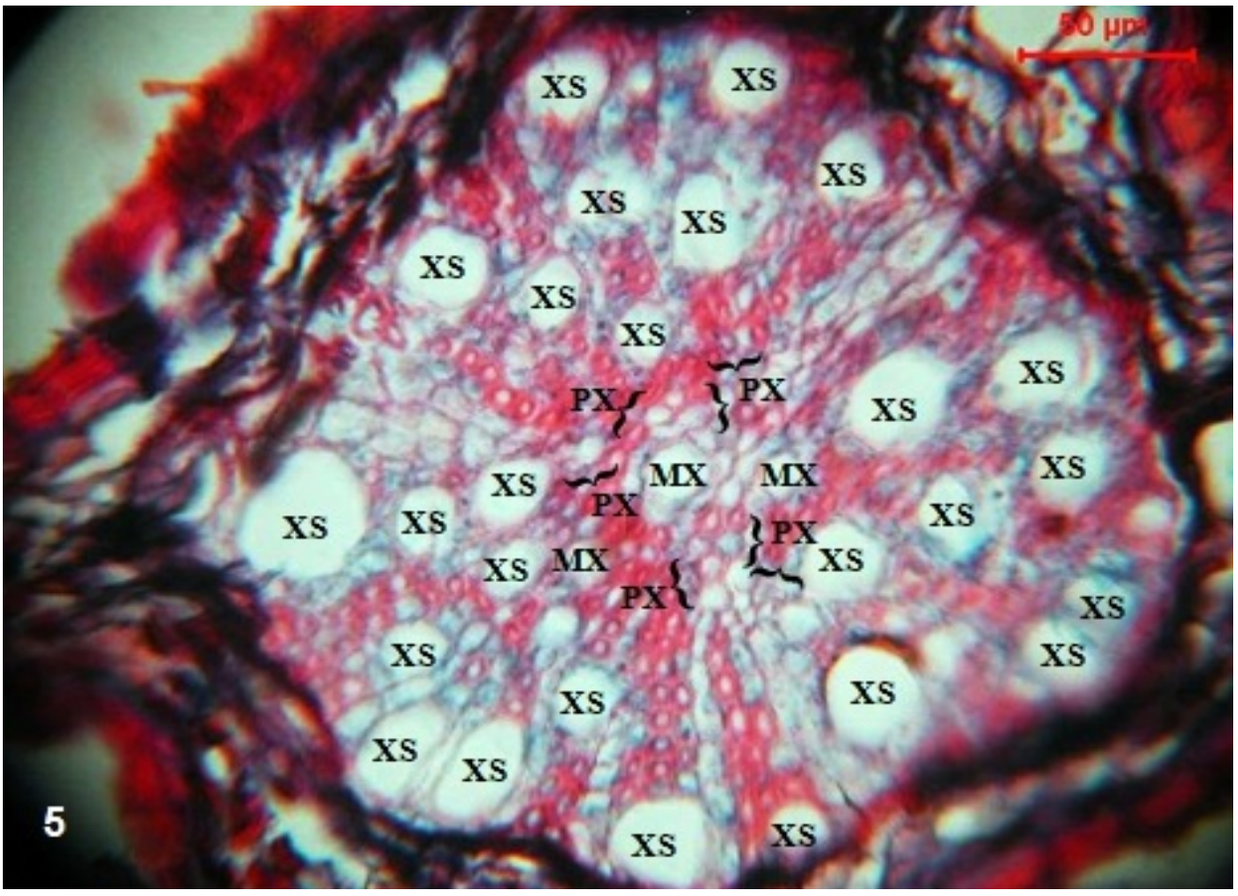


Figure 2. Cross sections of secondary roots of peach tree rootstock Aldrighi without inoculation (1) and inoculated with AMF (2 – *G. clarum*, 3 – *G. etunicatum*, 4 – *Acaulospora sp*., 5 - *Scutellospora heterogama*). MX – metaxylem; PX – protoxylem; XS – secondary xylem. Scale 50 µm.

2.6. Statistics

The data were submitted to an analysis of variance by the SAS program and the measurements were compared by the Duncan test (Duncan, 1955) at the level of 5% significance.

3. Results

The results regarding the effect of the AMF on the conductor tissue of the roots of the Okinawa cultivar showed that the treatments with the *Acaulospora sp.* and *G. etunicatum* species performed similarly for the number and diameter of the metaxylem and secondary xylem cells, superior to the other treatments, while for cell area and perimeter, *Acaulospora sp.* was superior to the other treatments. The treatment with *G. etunicatum* presented cell area, perimeter and diameter similar to *G. clarum*, superior to the controls. The treatment with *G. clarum* presented the number of cells of these xylem classes similar to that of the controls. There was an inverse performance for cortical thickness, where the control plants presented the highest results, followed by the plants inoculated with *G. clarum* while those inoculated with *Acaulospora sp.* and *G. etunicatum* presented similar but lower results, compared to the control plants (Table 1).

Treatment	Diameter of root (μm)	Cortex thickness (μm)	Metaxylem e secondary xylem			
			Number of Cell	Cell diameter (μm)	Cell perimeter (μm)	Cell area (μm²)
Acaulospora sp.	957,14^{ns}	118,12c	38,53a	5,50a	16,86a	58,55a
G. clarum	958,32^{ns}	129,85b	26,00b	5,11b	15,61b	48,46b
G. etunicatum	961,35^{ns}	119,31c	34,60a	5,26ab	15,70b	50,45b
Testemunha	952,61^{ns}	139,39a	22,00b	4,04c	11,77c	41,14c
V. C. (%)	5,17	7,41	5,01	10,09	6,02	13,22

Table 1. Root morphology of secondary roots of the Okinawa cultivar rootstock inoculated with three AMF species (*Acaulospora sp.*, *G. clarum* and *G. etunicatum*), collected 360 days after sowing. Eldorado do Sul, RS, 2005. Means followed by the same letter in the column do not differ by the Duncan test at 5% significance. ^{ns}Non-significative.

For the Aldrighi cultivar, the treatment with the *S. heterogama* species presented better results than the other treatments for the parameters number of cells, cell area and perimeter of the metaxylem and the secondary xylem, but performed similarly to *G. etunicatum* for cell diameter, and was superior to the other treatments for this parameter (Table 2).

Treatment	Diameter of root (μm)	Cortex thickness (μm)	Metaxylem e secondary xylem			
			Number of Cell	Cell diameter (μm)	Cell perimeter (μm)	Cell area (μm²)
Acaulospora sp.	945,01^{ns}	118,08c	33,40b	4,87b	12,95c	52,95c
G. clarum	939,11^{ns}	128,85b	24,93c	4,93b	13,10c	53,10c
G. etunicatum	955,23^{ns}	107,31d	30,20b	6,17a	16,86b	56,86b
S. heterogama	960,12^{ns}	105,22d	37,00a	7,06a	22,70a	62,70a
Testemunha	940,04^{ns}	138,39a	16,33d	3,87c	11,57d	41,57d
V. C. (%)	4,76	6,11	5,54	7,84	6,84	11,32

Table 2. Root morphology of secondary roots of the Aldrighi cultivar rootstock inoculated with four AMF species (*Acaulospora sp.*, *G. clarum*, *G. etunicatum* and *S. heterogama*) collected 180 days after sowing. Eldorado do Sul, RS, 2005. Means followed by the same letter in the column do not differ by the Duncan test at 5% significance. ^{ns}Non-significative.

G. etunicatum performed similarly to *Acaulospora sp.* regarding the number of cells that was better than *G. clarum* and the control. Furthermore, *G. etunicatum* performed better than *Acaulospora sp.*, *G. clarum* and the controls for the other parameters. The plants inoculated with *Acaulospora sp.* performed better than *G. clarum* for the number of cells and similarly to the other parameters. The control plants presented lower results for all the parameters in the assessment of the xylem classes. However, regarding cortical thickness, the control plants performed better than the other treatments, followed by *G. clarum* that was better than *Acaulospora sp.* that was better than the treatments with *S. heterogama* and *G. etunicatum* that presented similar results.

The inoculation with AMF species accelerated the growth of the plants of the Okinawa cultivar rootstock, inducing greater height, diameter, leaf area and greater nitrogen, phosphorus and potassium content, compared with the control. All presented root colonization rates were over 90%. *Acaulospora sp.* was the species that was most efficient among the AMF tested giving greatest height, diameter and nutritional state compared to the other species. The plants inoculated with *G. clarum* and *G. etunicatum* presented to mediate growth and nutritional state, and were similar (Table 3).

Treatment	H (cm)	D (mm)	L.A. (cm²/ plant)	Nutrients (%)			Colonization (%)
				N	P	K	
Acaulospora sp.	136,46a	8,42a	197,01a	2,35a	0,16a	2,07a	97,00a
G. clarum	126,65b	7,79b	163,00b	2,23b	0,15b	1,74b	91,76b
G. etunicatum	129,04b	7,87b	167,00b	2,22b	0,15b	1,82b	92,62b
Control	119,23c	7,24c	142,03c	2,05c	0,14c	1,60c	00,00c
V. C. (%)	3,88	2,17	2,54	2,56	2,61	4,75	2,42

Table 3. Height (H), root-stem junction diameter (D), leaf area (L.A.), percentage of nitrogen (N), phosphorus (P) and potassium (K) in the plant tissue and root colonization (%) of the Okinawa cultivar rootstock inoculated with three AMF species (*Acaulospora sp.*, *G. clarum* and *G. etunicatum*), collected 360 days after sowing. Eldorado do Sul, RS, 2005. Means followed by the same letter in the column do not differ by the Duncan test at 5% significance.

For the plants of the Aldrighi cultivar, only the *S. heterogama* and *G. etunicatum* species were shown to be efficient for the height parameter (Table 4).

Treatment	H (cm)	D (mm)	L.A. (cm²/ plant)	Nutrients (%)			Colonization (%)
				N	P	K	
Acaulospora sp.	130,94c	6,28c	125,00c	2,99c	0,16b	2,40b	30,00c
G. clarum	129,80c	6,24c	119,01c	2,96c	0,17b	2,44b	28,50c
G. etunicatum	138,36b	6,82b	137,00b	3,33b	0,20a	2,72a	91,50b
S. heterogama	143,97a	7,29a	173,10a	3,74a	0,22a	2,76a	97,75a
Control	129,70c	5,88d	105,00d	2,65d	0,16b	2,29c	00,00d
V. C. (%)	1,55	2,77	2,44	6,64	10,60	3,97	2,91

Table 4. Height (H), root-stem junction diameter (D), leaf area (L.A.), percentage of nitrogen (N), phosphorus (P) and potassium (K) in the plant tissue and root colonization (%) of the Aldrighi cultivar rootstock inoculated with four AMF species (*Acaulospora sp.*, *G. clarum*, *G. etunicatum* and *S. heterogama*), collected 180 days after sowing. Eldorado do Sul, RS, 2005. Means followed by the same letter in the column do not differ by the Duncan test at 5% significance.

These species were the only ones to present root colonization rates of over 90%. All of the AMF species were efficacious for the root-stem junction diameter and leaf area parameters and only varied in the response intensity. In all the assessments of plant growth and nutritional states, invariably *S. heterogama* induced greater growth compared to the other AMF species, while *G. etunicatum* induced intermediate performance and *Acaulospora sp.* and *G. clarum* presented similar performance.

Inoculation with AMF increased content of reserve substances to plants of cv. Okinawa, especially when inoculated with *Acaulospora sp*. In the shoots of rootstock plants inoculated with *G. etunicatum* and *G. clarum* showed intermediate levels. In roots, the plants inoculated with *G. etunicatum* also showed intermediate values, while those inoculated with *G. clarum* not differ from the controls (Table 5).

Treatment	Reserve substances (% in the plant)	
	Shoot	Roots
Acaulospora sp.	39,81a	28,38a
G. clarum	35,05b	21,02c
G. etunicatum	35,53b	24,28b
Control	27,29c	19,41c
V. C. (%)	5,24	2,58

Table 5. Reserve substances of shoots (leaves and stems) and roots of plants of the Okinawa cultivar rootstock inoculated with three AMF species (*Acaulospora sp*., *G. clarum* and *G. etunicatum*), collected 360 days after sowing. Eldorado do Sul, RS, 2005. Means followed by the same letter in the column do not differ by the Duncan test at 5% significance.

In reviewing the data on the percentage of reserve substances from plants of cv. Aldrighi, present in the tissue of the shoot, it appears that the plants were inoculated with the AMF species had percentages higher than uninoculated plants (Table 6).

Treatment	Reserve substances (% in the plant)	
	Shoot	Roots
Acaulospora sp.	34,69b	22,03c
G. clarum	35,50a	22,99bc
G. etunicatum	36,61a	24,92b
S. heterogama	38,25a	28,27a
Control	28,57c	19,80c
V. C. (%)	2,76	3,26

Table 6. Reserve substances of shoots (leaves and stems) and roots of plants of the Okinawa cultivar rootstock inoculated with four AMF species (*Acaulospora sp*., *G. clarum*, *G. etunicatum* and *S. heterogama*), collected 180 days after sowing. Eldorado do Sul, RS, 2005. Means followed by the same letter in the column do not differ by the Duncan test at 5% significance.

For the shoot, plants inoculated with *G. clarum*, *G. etunicatum* and *S. heterogama* showed percentages of reserve substances statistically similar but superior to *Acaulospora sp*., which in turn was higher than the control. In roots, the plants were inoculated with *S. heterogama* presented the greatest results, while those inoculated with *G. etunicatum* showed intermediate levels, statistically similar to *G. clarum* that, in turn, was similar to those inoculated with *Acaulospora sp*. and the control.

4. Discussion

4.1. Anatomy and morphology changes in roots

It was observed that inoculation with AMF reduced the cortex thickness of inoculated plants in both cultivars, associated to increase in most of the morphological parameters of the root xylem assessed for the Okinawa cultivar and for all of those of the Aldrighi cultivar (Tables 1 and 2).

The main effect of the AMF occurred on the metaxylem, that is, one of the categories of the primary xylem, whose conductor cells differentiate later and are larger in diameter (Costa et al., 2003) and also on the secondary xylem cells. On the other hand, the AMF not did not seem to exercise effect on the protoxylem, that are conductor cells of the primary xylem that differentiate first, that is, they acquire secondary lignin walls early (Apezzato-da-Glória & Hayashi, 2003) that reduce the possibility of the AMF acting on the growth of this category of cells of the primary xylem.

The decrease in the cortex area seems to be directly linked to the increase in the number of cells in the metaxylem and the secondary xylem of the plants inoculated with AMF. The control plants presented a smaller number of metaxylem and secondary xylem cells that were smaller in diameter compared to the cells of the inoculated plants, especially in the case of the treatments with the species *Acaulospora sp.* (cv. Okinawa) and *S. heterogama* (cv. Aldrighi).

The mycelia of endomycorrhizal fungi were extracted from roots of *Ophrys lutea* (Orchidaceae) and placed in culture medium kept and in the dark for three weeks (Barroso et al., 1986). Then the derivatives released in the culture medium were extracted, whose greatest concentrations consisted of indol-3-acetic acid and indol-3-ethanol acid, showing the ability of these fungi to synthesize hormones. The authors concluded that the identification of these compounds in the mycelia extract suggested the transference of these compounds from the fungi to the host plants in the phase when symbiosis was established.

Roots colonized by AMF presented an increase in auxin and cytokinin production that are involved in the increase or continuity of the growth of the conductor tissue cells, especially in the size and number of the cells of the metaxylem and the secondary xylem (Hirsch et al., 1997). According to the same authors, the establishment of symbiosis would lead to the production of biochemical signals that would activate genes involved in the production of these plant hormones, and thus the same signals would be responsible for the formation of

the root nodes on legumes colonized by *Rhizobium sp*. These authors showed that in roots of the MN 1008 alfalfa mutant cultivar, that carries out the transcription of these signals in the absence of the symbiants, the responses of the root tissue were identical to those of roots of plants colonized both by AMF and *Rhizobium meliloti*.

Thus it can be inferred that the presence of AMF would favor the constant differentiation of the xylem tracheal elements, that coincides with the results obtained in this study for both the root stock cultivars.

There appear to be possible variable effects on root morphology, according to the AMF species and the plant species involved in the symbiosis that also influences the size and growth of the xylem cells, that was also observed in this study, because some species presented variable performance in increasing the size and number of cells, in function of the cultivar used, and in function of the AMF species used for the same cultivar (Atkinson et al., 1994). The species *G. fasciculatum* and *G. etunicatum* induced modification in the roots of the Elsanta and Cambridge Favorite strawberry cultivars, but did not cause any alteration in the morphology of the roots of the Rhapsody cultivar, shedding the variable effect of these AMF species on different cultivars of the same plant species (Norman et al., 1996).

Moreover, roots of plants were colonized by AMF may or may not show increases in longevity, depending on plant species and the fungi involved in symbiosis (Atkinson et al, 2003; Eissenstat et al., 2000; Hodge et al., 2000). However, the morphological attributes of the roots that may be affected by the AMF, such as roots and branches of the diameter of the conducting tissue, has a direct influence on increasing the longevity of roots (Wells et al., 2002). In addition to increasing longevity, root colonization by AMF provides a quick renewal of the root system, increasing the rate of substitution of roots that have collapsed.

4.2. Acquisition of nutrients and benefits

The increase in the absorption and transport volume of nutrients such as nitrogen, that is a constituent of proteins (Tedesco et al., 1995), phosphorus that is essential for a cell division and photosynthesis metabolism and potassium that acts on the electric equilibrium of the cells and on the stomata opening and closing (Tedesco et al., 1995), is vital for plant growth. This contributed to greater responses of the inoculated plants in terms of plant development that was observed in this study for both the cultivars, especially in the plants where there were the highest percentages of root colonization (Tables 3 and 4).

The AMF obtain carbohydrates from their host plants and provide nutrients, especially phosphate. In the case of phosphate, depending on the combination plant-fungus, the acquisition can be performed wholly or partly by the fungus (Smith et al., 2003). The metabolic pathway of nutrient acquisition starts with the uptake by hyphae-soil interface (Benedetto et al., 2005). In hyphae, the nutrient is transported to structures of the fungus in the roots (Ohtomo et al., 2005), where it is transferred to the plant via arbúsculo (Nagy et al.,

2005). The route of transfer of carbohydrates from the plant to the AMF follows the opposite direction (Nagy et al., 2005; Ohtomo et al., 2005).

The benefits given by the AMF on xylem development is associated to many action mechanisms of these fungi, that act directly or indirectly on the plants (Souza et al., 2000). One of the positive effects of the AMF is in function of the presence of the external mycelia, which play an important role in slow diffusion nutrient absorption, such as phosphorus and potassium (Minhoni & Auler, 2003; Souza et al., 2000; Tobar et al., 1994), increasing the nutritional content of the plants (An et al., 1993; Barea, 1991). Associated to this, the modifications caused by the AMF in the xylem structure, such as increase in the number and diameter of the metaxylem and secondary xylem cells, permitted a greater flow of nutrient absorption, such as nitrogen, phosphorus and potassium, translocated to the upper part of the plant, culminating in accelerated growth (Souza, 2000; Souza et al., 2000).

The fact that the AMF species induce major development parameters such as height, diameter and leaf area per plant provides greater photosynthesis and, consequently, a higher level of production of assimilates (Nunes et al., 2006). This report confirms the data obtained in this study with respect to the reserve substances of shoots of both cultivars, for all species used provided an increase in leaf area compared to control (Tables 3 and 4). There is also agreement with other authors, who found higher levels of reserve substances in the tissues of plants inoculated with AMF (Theodore et al., 2003; Sena et al., 2004, Souza et al., 2005).

Another fact to be noted is that only the AMF species that provided the greatest results for height, diameter and leaf area for both cultivars (*Acaulospora sp*. Okinawa cultivar, *S. heterogama* for Aldrighi cultivar and *G. etunicatum* for both cultivars) yielded significant differences in plant reserves, both to the tissues of shoots and roots to the tissues, compared to other treatments. Plants with greater height and leaf area, are more light-gathering capacity and production of assimilates, which allows a higher flow of carbohydrates into the root system where one part would be used by the AMF in its nutrition and accumulation in structures buffer (vesicles, where *Acaulospora sp*. and *G. etunicatum*), and the rest would be accumulated in the storage tissue of the plant in the form of reserve substances (Souza et al., 1999; Scatena & Scremin-Dias, 2003). Moreover, the larger diameter provided by the AMF, would increase the upward flow of water and nutrients, and sap formulated in the downward (Mazzoni-Viveiros & Trufem, 2004).

5. Conclusion

Plants inoculated with AMF have changes in the morphological structure of the roots, such as reduction of the cortex and increased the number and size of cells of the metaxylem, which provides greater volume of water and nutrients translocated to the top of the plant. This benefits plants, accelerating its vegetative growth, improving the content of macronutrients and allowing the production and accumulation of assimilates.

Author details

José Luis da Silva Nunes
BADESUL Desenvolvimento, Brasil

Paulo Vitor Dutra de Souza, Gilmar Arduino Bettio Marodin and Jorge Ernesto de Araújo Mariath
Universidade Federal do Rio Grande do Sul, Brasil

José Carlos Fachinello
Universidade Federal de Pelotas, Brasil

Acknowledgement

To Ministério da Agricultura, Pecuária e Abastecimento (MAPA) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for research support and grants of the authors.

6. References

An Z.Q., Shein T., Wang H.G. (1993). Mycorrhizal fungi in relation to growth and mineral nutrition of apple seedlings. *Scientia Horticulturae*, Vol.54, Nº 4 (July 1993), pp. 275 – 285, ISSN 0304-4238

Appezzato-da-Glória B., Hayashi A.H. (2003). Raiz. In: *Anatomia Vegetal*, Appezzato-da-Glória B., Carmello-Guerreiro S.M. (eds), pp. 267 – 287, UFV, ISBN 85-7269-240-1, Viçosa, Brasil

Atkinson D., Berta G., Hooker J.E. (1994). Impact of mycorrhizal colonization on root archicture, roots longevity and the formation of growth regulators. In: *Impact of Arbuscular Mycorrhizas on Sustainable Agriculture and Natural Ecosystems*, Gianinazzi S., Schüepp H. (eds), pp. 89 – 99, Birkhäuser Verlag, ISBN 3-7643-5000-8, Basel, Switzerland

Atkinson, D., Blanck, K.E., Forbes, P.J., Hooker, J.E., Baddeley, J.A., Watson, C.A. (2003). The influence of arbuscular mycorrhizal colonization and environment on root development in soil., Vol.54, Nº 4 (December 2003), pp. 751 – 757, ISSN 1365-2389

Barea J.M. (1991). Vesicular-arbuscular mycorrhizae as modifiers of soil fertility. In: *Advances in Soil Science*, STEWART, B.S. (ed), pp. 01 – 40, Springer-Verlag, ISBN 0-3879-7354-0, New York, USA

Barroso J., Neves H.C., Pais M.S. (1986). Production of indole-3-ethanol and indole-3-acetic acid by the mycorrhizal fungus of *Ophrys lutea* (Orchidaceae). *New Phytologist*, Vol.103, Nº 4 (December 1986), pp. 745 – 749, ISSN 1469-8137

Benedetto, A., Magurno, F., Bonfante, P., Lanfranco, L. (2005). Expression profiles of a phosphate transporter gene (GmosPT) from the endomycorrhizal fungus *Glomus mosseae*. *Mycorrhiza*, Vol.15, Nº 8 (December 2005), pp. 620 – 627, ISSN: 0940-6360

Berta G., Trotta A., Fusconi A., Hooker J.E., Munro M., Atkinson P., Giovannetti M., Morini S., Fortuna P., Tisseranti B., Gianinazzi-Pearson V., Gianinazzi S. (1995). Arbuscular mycorrhizal induced changes to plant growth and root system morphology in *Prunus cerasifera*. *Tree Physiology*, Vol.15, Nº 5 (May 1995), pp. 281 – 293, ISSN 0829-318X

Bressan W., Vasconcellos C.A . (2002). Alterações morfológicas no sistema radicular do milho induzidas por fungos micorrízicos e fósforo. *Pesquisa Agropecuária Brasileira*, Vol.37, Nº 4 (Abril 2002), pp. 509 – 517, ISSN 0100-204X

Cooper K.M. (1984). Physiology of VA Mycorrhizae associations. In: *VA Mycorrhiza*. Powel CL, Bagyaraj J (eds), pp. 155 – 186, CRC, ISBN 08-493-569-46, Boca Raton, USA

Costa C.G., Callado C.H., Coradin V.T.R., Carmello-Guerreiro S.M. (2003). Xilema. In: *Anatomia Vegetal*, Appezzato-da-Glória B., Carmello-Guerreiro S.M. (eds), pp. 129 – 154, UFV, ISBN 85-7269-240-1, Viçosa, Brasil

Dodd, J. C. (2000) The role of arbuscular mycorrizal fungi in agro – and natural ecosystems. *Outlook on Agriculture*, Vol.29, Nº 1 (March 2000), p. 55 – 62, ISSN 0030-7270

Duncan, D.B. (1955). Multiple range and multiple F tests. *Biometrics*, Vol.11, Nº 1 (March 1955), pp. 1- 42, ISSN 1947-2006

Eissenstat, D.M., Wells, C.E., Yanai, R.D., Whitbeck, V.L. (2000) Building roots in a changing environment: implications for root longevity. *New Phytologist*, Cambridge, Vol.147, Nº 1 (July 2000), pp. 33 – 42, ISSN 1469-8137

Hirsch A.M., Fang Y., Asad S., Kapulnik Y. (1997). The role of phytohormones in plant-microbe symbioses. *Plant and soil*, Vol.194, Nº 2 (January 1997), pp. 171 – 184, ISSN 0032-079X

Hodge, A., Robinson, D., Fitter, A.H. (2000). An arbuscular mycorrhizal inoculum enhances root proliferation in, but not nitrogen capture from, nutrient-rich patches in soil. *New Phytologist*, Cambridge, Vol.147, Nº 3 (September 2000), pp. 575 - 584, ISSN 1469-8137

Johansen D.A. (1940). *Plant microtechnique*. McGraw-Hill, ISBN 007592, New York, USA

Kothari B.K., Maschner, H., George, E. (1990) Effect of VA mycorrhizal fungi and rhizosphere microorganisms on root and shoot morphology, growth and water-relations in maize. *New Phytologist*, Vol.116, Nº 2 (October 1990), p. 303 – 311, ISSN 1469-8137

Marschner, H. (1995) *Mineral nutrition of higher plants*. Academic Press, ISBN 978-0124735415, San Diego, USA

Mazzoni-Viveiros, S.C., Trufem, S.F.B. (2004) Efeitos da poluição aérea e edáfica no sistema radicular de *Tibouchina pulchra* Cogn. (Melastomataceae) em área de mata Atlântica: associações micorrízicas e morfologia. *Revista Brasileira de Botânica*, Vol.27, Nº 2 (Abril/ Junho 2004), pp. 337 – 348, ISSN 0100-8404

Minhoni M.T.A., Auler P.A.M. (2003) Efeito do fósforo, fumigação do substrato e fungo micorrízico arbuscular sobre o crescimento de plantas de mamoeiro. *Revista Brasileira de Ciência do Solo*, Vol.27, Nº 5 (Outubro 2003), pp. 841 – 847, ISSN 0100-0683

Moreira F.M.S., Siqueira J.O. (2002) *Microbiologia e bioquímica do solo*. Editora UFLA, ISBN 85-8769-233-X, Lavras, Brasil

Nagy, R., Karandashov, V., Chague, V., Kalinkevich, K.; Tamasloukht, M., Xu, G., Jakobsen, I., Levy, A.A., Amrhein, N., Bucher, M. (2005). The characterization of novel mycorrhiza-specific phosphate transporters from *Lycopersicon esculentum* and *Solanum tuberosum* uncovers functional redundancy in symbiotic phosphate transport in solanaceous species. *Plant Journal*, Vol.42, Nº 2 (April 2005), pp. 236 – 250, ISSN 1365-313X

Norman J.R., Atkinson D., Hooker J.E. (1996). Arbuscular mycorrhizal-fungal-induced alteration to root architecture in strawberry and induced resistance to the pathogen *Phytophthora fragariae*. *Plant and Soil*, Vol.185, Nº 2 (September 1996), pp. 191 – 198, ISSN 0032-079X

Nunes M.S., Soares, A.C.F., Soares Filho, W.S., Lêdo, C.A.S. (2006). Colonização micorrízica natural de porta-enxertos de citros em campo. *Pesquisa Agropecuária Brasileira*, Vol.41, Nº 3 (Março 2006), pp. 525 – 528, ISSN 0100-204X

Ohtomo, R., Saito, M. (2005). Polyphosphate dynamics in mycorrhizal roots during colonization of an arbuscular mycorrhizal fungus. *New Phytologist*, Vol. 167, Nº 2 (August 2005), pp. 571 – 578, ISSN 1469-8137

Panstruga, R. (2003). Establishing compatibility between plants and obligate biotrophic pathogens. *Current Opinions in Plant Biology*, Vol.6, Nº 4 (August 2003), p. 320-326, ISSN: 1369-5266

Paszkowski, U. (2006). Mutualism and parasitism: the yin and yang of plant symbioses. *Current Opinions in Plant Biology*, Vol.9, Nº 4 (August 2006), p.364 – 370, ISSN: 1369-5266

Phillips J.M., Hayman D.S. (1970). Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Transactions of the British Mycological Society*, Vol.55, Nº 1 (January 1970), pp. 157-160, ISSN 0007- 1536

Priestley, G.A. (1965) New method for the estimation of the resources of apple tress. *Journal of the Science of Food and Agriculture*, Vol.16, Nº 12 (December 1965), pp. 717 – 721, ISSN 1097-0010

Scatena, V.L., Scremin-Dias, E. (2003). Parênquima, Colênquima e Esclerênquima. In: *Anatomia Vegetal*, Appezzato-da-Glória B., Carmello-Guerreiro S.M. (eds), pp. 109 – 127, UFV, ISBN 85-7269-240-1, Viçosa, Brasil

Sena, J.O.A., Labate, C.A., Cardoso, E.J.B.N. (2004) Caracterização fisiológica da redução de crescimento de mudas de citros micorrizadas em altas doses de fósforo. *Revista Brasileira da Ciência do Solo*, Vol.28, Nº 5 (Setembro/ Outubro 2004), pp. 827 – 832, ISSN 0100-0683

Siqueira J.O., Lambais M.R., Stürmer S.L. (2002) Fungos micorrízicos arbusculares. *Biotecnologia, Ciência & Desenvolvimento*, Vol.25 (Março/Abril 2002), pp. 12 – 21, ISSN 1414-4522

Silva, L.M.S.; Alquini, Y.; Cavallet, V.J. (2005) Inter-relações entre a anatomia vegetal e a produção vegetal. *Acta Botanica Brasilica*, Vol.19, Nº 1 (Janeiro/ Março 2005), p. 183 – 194, ISSN 0102-3306

Souza, F.A., Trufem, S.F.B., Almeida, D.L., Silva, E.M.R., Guerra, J.G.M. (1999) Efeito de pré-cultivos sobre o potencial de inoculo de Fungos Micorrízicos Arbusculares e produção de mandioca. *Pesquisa Agropecuária Brasileira*, Vol.34, Nº 10 (Outubro 1999), pp. 1913 – 1923, ISSN 0100-204X

Souza P.V.D., Agustí M., Abad M., Almela V. (2000). Desenvolvimento vegetativo e morfologia radicular de Citrange Carrizo afetado por ácido indolbutírico e micorrizas arbusculares. *Ciência Rural*, Vol.30, Nº 2 (Março/Abril 2000), pp. 249 – 255, ISSN 0103-8478

Souza P.V.D. (2000). Interação entre micorrizas arbusculares e ácido giberélico no desenvolvimento vegetativo de plantas de Citrange Carrizo. *Ciência Rural*, Vol.30, Nº 5 (Setembro /Outubro 2000), pp. 783 – 787, ISSN 0103-8478

Souza, P.V.D., Carniel, E., Schimitz, J.A.K., Silveira, S.V. (2005) Influência de substratos e fungos micorrízicos arbusculares no desenvolvimento do porta-enxerto Flying Dragon (*Poncirus trifoliata*, var. monstruosa Swing.). *Revista Brasileira de Fruticultura*, Vol.27, Nº 2 (Agosto 2005), pp. 285 – 287, ISSN 0100-2945

Smith, S.E., smith, F.A., Jakobsen, I. (2003) Mycorrhizal fungi can dominate phosphate supply to plants irrespective of growth responses. *Plant Physiology*, Vol.133, Nº 2 (October 2003), pp. 16–20, ISSN 0032-0889

Sylvia, D. M., Chellemi, D. O. (2001) Interations among root-inhabiting fungi and their implications for biological control of root pathogens. *Advances in Agronomy*, Vol.73, Nº 1 (April 2001), pp. 1 – 33, ISBN 978-0-12-000773-8

Tedesco M.J., Gianello C., Bissani C.A., Bohnen H., Volkweiss S.J. (1995). *Análises de solo, plantas e outros materiais (Boletim Técnico, 5*), UFRGS/Departamento de solos, ISBN 000-148837, Porto Alegre, Brasil

Theodoro, V.C.A., Alvarenga, M.I.N., Guimarães, J., Mourão Junior, M. (2003) Carbono da biomassa microbiana e micorriza em solo sob mata nativa e agroecossistemas cafeeiros. *Acta Scientiarum: Agronomy*, Vol.25, Nº 1 (Maio 2003), pp. 147 – 153, ISSN 1679-9275

Tobar R., Azcón R., Barea J.M. (1994) Inproved nitrogen uptake and transport from ^{15}N-labelled nitrate by external hyphae of arbuscular mycorrhiza under water stressed condictions. *New Phytologist*, Vol.126, Nº 1 (January 1994), pp. 119 – 122, ISSN 1469-8137

Tokeshi, H. (2000) Doenças e pragas agrícolas geradas e multiplicadas pelos agrotóxicos. *Fitopatologia Brasileira*, Vol.25 (Janeiro 2000), pp. 264-270, ISSN 0100-4158

Wells, C.E., Glenn, D.M., Eissenstat, D.M. Changes in the risk of fine-root mortality with age: a case study in peach, *Prunus persica* (Rosaceae). *American Journal of Botany*, Vol.89, Nº 1 (January 2002), pp. 79 – 87, ISSN 0002-9122

Wilcox, H. E. (2002). Mycorrhizae. In: *Plants roots*, Waisel, Y., Eshel, A., Kafkafi, U. (eds), Marcel Dekker, ISBN 0-8247-0631-5, New York, USA

IntechOpen