

Full Length Research Paper

Spore density and diversity of Arbuscular mycorrhizal fungi in medicinal and seasoning plants

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Arbuscular mycorrhizal fungi (AMF) set mutualistic symbiosis with most plants. Understanding this association and meet the diversity of AMF in both the medicinal and the seasoning herbs is very important, since these plants have increasingly contributed to improving the quality of human life. The aim of this study was to assess the spore density, taxonomic diversity, and root colonization by AMF in experimental beds of rosemary (*Rosmarinus officinalis* L.), nasturtiums (*Tropaeolum majus*), mint (*Mentha crispa* L.), boldo (*Peumus boldus*), oregano (*Origanum vulgare*) and chamomile (*Matricaria chamomilla*), all planted in the Medicinal Plant Nursery of the Paranaense University - UNIPAR, Umuarama – PR. Soil samples (0 to 10 cm depth) and plant roots were collected in two periods, June and November 2011. Colonization of plant roots by AMF ranged 17 to 48%. The rosemary treatment was highly responsive to the sampling periods, with only 17% of root colonization in June compared with 48% in November. The AMF spore density was higher in June than in November for all species of plants studied. Among the AMF identified within this study, the dominant genus was *Glomus* sp., followed by *Acaulospora* sp. in all plants analyzed. Greater knowledge over diversity and density of AMF spores can strongly contribute to the sustainable management of nutrition for medicinal and seasoning plants, particularly on phosphorus supply.

Key words: Diversity of mycorrhizal fungi, symbiosis, mycorrhizae, medicinal and seasoning plants.

INTRODUCTION

Arbuscular mycorrhizal fungi (AMF) compose a key functional group of the soil biota that can substantially contribute to plant yields and ecosystem sustainability in crop production strategies. Presently, applications of beneficial microbial inoculants (biofertilizers) are increasingly attracting attention toward sustainable agriculture and life quality as a consequence of the need to solve health and environmental problems resulting

from the excessive use of agrochemicals through conventional farming practices (Gianinazzi et al., 2010).

The AMF are commonly found in nature and very important as biofertilizers. They belong to Phylum *Glomeromycota*, Class *Glomeromycetes* and form a monophyletic group of fungi classified into four orders, thirteen families, and nineteen genera, with somewhat 215 species already described (Siqueira et al., 2010).

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AMF form mutualistic symbiosis with the roots of most plants. Through this symbiosis, the fungus gets carbohydrates and other elements essential to their development from the host plant, forming new spores by sporulation processes. In contrast, the host plant obtains from the soil, with the help of the fungus, water and inorganic nutrients such as phosphorus (P), benefits by getting long and bulky roots, and acquires resistance to pathogens and abiotic stress, such as the presence of heavy metals and water shortage (Carrenho et al., 2007; Smith and Read, 2008).

Studies on mycorrhizal symbiosis with medicinal and seasoning plants are scarce. However, some of these few studies have shown that AMF can increase the production of secondary compounds containing medicinal active ingredients in plants under mycorrhizal symbiosis, in addition to promoting their growth (Faria et al., 2000; Russomano et al., 2008).

The aim of this study was to assess the content of soil organic matter (SOM), spore density and AMF root colonization in plants of rosemary (*Rosmarinus officinalis* L.), nasturtiums (*Tropaeolum majus*), mint (*Mentha crispa* L.), boldo (*Peumus boldus*), oregano (*Origanum vulgare*) and chamomile (*Matricaria chamomilla*) cultivated in experimental beds (plots) in the Medicinal Plant Nursery of the Paranaense University - UNIPAR, Umuarama – PR, in two periods, June and November, 2011.

MATERIALS AND METHODS

Experimental field: Soil and root sampling

Root and soil samples were collected at the Medicinal Plant Nursery of the Paranaense University - UNIPAR - Campus II, in the Umuarama city, northwestern Paraná State at coordinates S 23° 46' 11.34" and WO 53° 16' 41.78".

For each plant of rosemary, nasturtiums, mint, boldo, oregano and chamomile were assigned three experimental beds. Roots and rhizosphere soil were sampled in three points of each bed, giving a total of 9 replications per plant species in a completely randomized design. The beds received organic compost (coffee leaf straw transformed by fermentation process of composting) before being planted. Then, as with plants, the beds were irrigated daily by spraying when needed. Plants with the exception of boldo and rosemary, were at the phenological stage of pre-flowering.

The roots and soil sampling was performed at 0 to 10 cm, about 10 cm away from the stem of each plant, into two periods: June and November 2011. Sampling was done at the same point in each bed for the two periods. In each plot, three samples were collected for approximately 0.5 kg of soil, placed in plastic bags and stored in a refrigerator (4°C) until laboratory analyzes.

A soil sample was collected for chemical and granulometric analyses. One portion of that sample was utilized to determine the soil chemical characteristics at *Solo Fértil* Laboratory in the city of Umuarama, Paraná, Brazil. The characteristics determined were: pH in CaCl₂, Ca²⁺, Mg²⁺ and Al³⁺ extracted in KCl (1 Mol L⁻¹), and P and K⁺ extracted in Mehlich-1. All the analyses followed the CELA/PR standards to obtain a greater reliability of the results (Table 1). The other portion of the soil sample was intended for identification of the density and taxonomy of AMF spores. In both periods, the thinner roots of the plants were collected at three

points of each bed (n = 9), washed in water, placed in flasks with preserving solution containing ethyl alcohol, acetic acid and formaldehyde (1:1:1) and stored in a refrigerator (4°C) (Souza, 2000) until laboratory analyzes for determining the percentage of AMF root colonization.

Spore density of arbuscular mycorrhizal fungal

The spores were extracted from 50 g of soil subsamples using the wet sieving method (Gerdemann and Nicolson, 1963). Each sample was suspended in 1 L of water and agitated in a beaker, kept at rest for 1 min so that the rougher particles of the soil were decanted, and then the content was poured on two juxtaposed sieves with 0.710 mm and 0.053 mm opening; the procedure repeated for four times. The material remained at the 0.053 mm sieve was transferred to 50 mL Falcon tubes, centrifuged in distilled water (3000 rpm, 3 min), and supernatant discarded. Next, saccharose solution (50%) was added into the tubes and they were agitated and centrifuged (2000 rpm, 2 min). The spores in the supernatant were transferred to the 0.053 mm sieve, washed to eliminate saccharose excess, transferred to Petri dishes and then counted under stereoscopic lens (40X).

Characterization and diversity of AMF

In Glomeromycota, taxonomy can be performed through morphological analysis of the formation, structure and germination of AMF spores. Spores were fixed on semi-permanent slides in two separate groups: one group with PVLG (polyvinyl alcohol and glycerol) resin and the other with PVLG resin + Melzer, and counted under a microscope (Morton et al., 1993). The sporocarps were carefully broken and the spores were counted.

Species taxa of AMFs were identified using Schenck and Pérez (1988) and INVAM - International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (<http://invam.caf.wvu.edu>) in addition to other species descriptions. From the number of individuals of each genus, the indexes of dominance (Simpson) and diversity (Shannon-Wiener) were estimated according to Souza et al. (2010). They were calculated according to the equations:

$$C = -\sum (X_i/X_0) \times \log (X_i/X_0) \quad \text{Simpson,}$$

$$H' = -\sum (X_i/X_0)^2 \quad \text{Shannon-Wiener,}$$

where X_i is the spore density of each genus in 100 g of soil, X_0 is the total spore density of all AMF genera.

AMF root colonization

To determine AMF root colonization, six plants with roots were collected from each subplot (beds) and washed in running water. Plant roots were freshly cut at the length of ±1.5 cm so that they can be represented as the whole radicular system. The lab procedure was done according to Phillips and Hayman (1970), where sample roots are placed in 10% KOH and closed in plastic Falcon Tubes. After heating the tubes with roots in water bath at 90°C for 1 h, the KOH solution was removed and the roots were washed in running water. A solution of 1% HCL was added in the tubes with roots and agitated for acidification for 5 min; next, the solution was removed. Then, roots were stained by adding 0.05% trypan blue to the tubes, which were heated in water bath at 90°C for 30 min. At the end of the process, the roots were preserved in lactoglycerol. The root segments were examined in stereoscopic microscope (100x) for AMF structures and percentage root length colonization was

Table 1. Chemical properties of the experimental soil (0 – 10 cm) sampled in the experiment area during 1st and 2nd sampling period – June and November of 2011.

Plant	pH	P	C	Al ³⁺	H ⁺ +Al ³⁺	Ca ²⁺ +Mg ²⁺	Ca ²⁺	Mg ²⁺	K ⁺	SB	CEC	V
	CaCl ₂	mg dm ⁻³	g dm ⁻³	----- Cmol _c dm ⁻³ -----			-----					(%)
1st sampling – June												
Rosemary	4.63	52.40	7.60	0.0	4.96	3.50	2.00	1.50	0.15	3.65	8.61	42.42
Boldo	5.24	21.00	6.04	0.0	3.68	4.38	3.38	1.00	0.21	4.58	8.26	55.45
Chamomile	4.72	90.00	7.21	0.0	4.96	7.63	3.88	3.75	0.21	7.83	12.79	61.22
Nasturtiums	5.47	173.60	8.77	0.0	3.68	5.75	3.25	2.50	0.21	5.96	9.64	61.81
Mint	5.32	282.80	8.57	0.0	4.28	6.25	4.25	2.00	0.21	6.46	10.74	60.13
Oregano	5.20	57.40	8.57	0.0	4.28	5.50	3.25	2.25	0.21	5.71	9.99	57.14
2nd sampling – November												
Rosemary	5.17	60.10	6.62	0.0	3.97	4.50	2.75	1.75	0.15	4.65	8.62	53.96
Boldo	5.30	23.70	7.21	0.0	3.97	6.88	4.00	2.88	0.10	6.98	10.95	63.74
Nasturtiums	5.69	248.20	10.91	0.0	3.42	6.25	3.50	2.75	0.26	6.51	9.93	65.55
Mint	5.46	207.20	7.60	0.0	3.97	5.25	4.25	1.00	0.15	5.4	9.37	57.65
Oregano	5.08	69.90	7.60	0.0	4.28	4.88	2.63	2.25	0.15	5.03	9.31	54.02

P – Phosphorus; C – Carbon; Al³⁺ – Aluminium; H⁺+Al³⁺ – Potential Acidity; Ca²⁺ – Calcium; Mg²⁺ – Magnesium; K⁺ – Potassium; SB – Sum of Bases; CEC – Cation Exchange Capacity; V – Bases saturation

estimated according slide method (Giovannetti and Mosse, 1980) for each replication of each treatment.

Statistical analysis

Data was subjected to one-way ANOVA using general linear model with mixed-effects and balanced design, considering each plant species as one treatment, and compared with the Duncan's test ($p \leq 0.05$), by using SPSS version 16.0 for Windows (SPSS Inc., Chicago, IL, USA). To comply with ANOVA assumptions, the data was previously checked with the Levene's test. In the two periods June and November, *t*-test was done with independent bilateral averages.

RESULTS AND DISCUSSION

Soil chemical analysis from the first sampling period (June, 2011) presented the highest P level in soil with mint (282.8 mg dm⁻³), followed by soil with nasturtiums (173.6 mg dm⁻³) and boldo (21 mg dm⁻³) (Table 1).

Both the P fixation and the natural P levels of soils vary according to plant variety or cultivar. Changes also may occur in the AMF activity depending on the conditions of soil fertility (Siqueira et al., 2010). However, the amount of organic matter from composting added to the beds is not exactly known for evaluating fertility within this study.

In general, stable levels of P were observed in the analysis of soils collected from the second sampling (November, 2011), with an increase in P levels observed only in the soil cultivated with nasturtiums (Table 1).

The availability of nutrients is affected by the soil pH. In this study, results of soil pH (Table 1) are according to the literature, which indicates values of 6 to 7 as ideal to

grow most of plant species (Corrêa Junior and Scheffer, 2009); remembering that there are plants that can tolerate lower pH. On the other hand, the low pH, as of the soil with rosemary (4.63 – Table 1), may affect the mycorrhizal association with plants. This is due to the variation of the solubility of elements such as Al, Fe, Mn and Cu, which at toxic levels may reduce the germination of spores and germ tubes, reducing the sporulation of AMF (Lambais and Cardoso, 1989). Studies show that soil pH regulates the mycorrhizal condition and controls the distribution of AMF species (Moreira et al., 2003).

The C levels in the soil differed between sampling periods (Table 1). For example, the bed with nasturtiums had 8.77 g dm⁻³ C in June and started having 10.91 g dm⁻³ C in November. According to Kaschuk et al. (2010; 2011), the C level in soils can also be used as indicator of their fertility and quality as it supplies biological activity, maintains environmental quality and promotes the health of plants and animals.

The roots of all plants analyzed were colonized by AMF. The AMF root colonization in June was significantly lower than in November (Table 2), with averages of 25.72 and 35.36%, respectively. Boldo, mint, oregano and nasturtiums had no significant differences in AMF root colonization (Table 2). Rosemary had a significant increase in AMF root colonization in November compared to June. It indicates that AMF root colonization was maximum in winter season and lowest in early summer season. Kumar et al. (2010) observed similar results for *Spilantes acmella*, *Withania somnifera*, *Salvia officinalis*, *Mentha spicata* when AMF root colonization was significantly higher in November than in June.

Among all plants analyzed within this study, mint

Tabela 2. Means of AMF soil spore density ($\text{n}^\circ \text{g}^{-1}$ of dry soil) and AMF root colonization (%) (\pm standard deviation, $n = 9$) in June and November, 2011.

Plant	AMF root colonization		AMF spore density	
	June	November	June	November
Rosemary	17.40 \pm 4.83 Bb	48.04 \pm 8.26 Aa	21.15 \pm 4.41 BCa	2.84 \pm 0.70 Ab
Boldo	30.74 \pm 4.92 Aa	45.67 \pm 8.82 Aa	16.80 \pm 3.11 Ca	3.01 \pm 1.09 Ab
Chamomile	20.80 \pm 4.82 B	ND	37.30 \pm 8.60 A	ND
Nasturtiums	21.88 \pm 4.31 Ba	18.35 \pm 4.45 Ba	16.52 \pm 2.37 Ca	3.05 \pm 0.47 Ab
Mint	42.37 \pm 7.47 Aa	45.35 \pm 7.01 Aa	19.96 \pm 2.65 BCa	3.84 \pm 0.77 Ab
Oregano	19.28 \pm 2.65 Ba	16.55 \pm 3.29 Ba	32.11 \pm 4.75 ABa	3.18 \pm 0.46 Ab
<i>p</i> value	> 0.001	> 0.002	> 0.001	0.902

ND = Not determined. Means followed by the same capital letter in the column are not significantly different by the Duncan test ($p \leq 0.05$) and by the same minor letter in the line did not differ by the t-test ($p \leq 0.05$).

showed the highest percentage of colonization, with 42.37% (June) and 45.35% (November), followed by boldo with 30.74% (June) and 45.67% (November). This result indicates that both the mint and the boldo are the species that are more depending on the AMF associations.

As AMF started establishing on the thinnest roots, temperature and humidity were likely to influence AMF root colonization as well as the nutrients intake by plants in this study (Smith and Read, 2008). According to Carrenho et al. (2007), the process of root colonization can be influenced by changes in seasonal periods. It was also observed in other studies in which the best root colonization was in the rainy season (Kumar et al., 2010). However, Radhika and Rodrigues (2010) states that mycorrhizal root colonization is present in all seasons, suggesting a plant dependence on AMF throughout the year.

A root can be colonized by more than one species of mycorrhizal fungus (Dood et al., 2000) and a fungus species can grow at different rates when associated to different species of plants (Smith and Read, 2008). Still, there may be several colonizing rates across genotypes of the same plant species (Grahman and Eissenstat, 1994).

Gupta et al. (2002) observed an increased percentage of root colonization in plants inoculated with AMF compared with respective controls of non-inoculated plants. The authors also noted a possible difference in response to mycorrhizal colonization across varieties of the same plant, as observed in the three cultivars of mint inoculated with *G. fasciculatum* in their study.

The major density of AMF spores (number of spores g^{-1} of dry soil) was observed in the period of June, with emphasis on soils with oregano (32.11) and chamomile (37.29), which showed densities significantly higher than the soils of other plants studied (Table 2). However, AMF spores were present in November with minor density, but with no significant difference when compared to June (Table 2). It can be explained here by a wetter weather

(data not shown) affecting directly fungus sporulation. Kumar et al. (2010) observed similar results for soils with *S. acmella* and *Mellisa officinalis*, in which AMF spores density was significantly lower in November than in June. Radhika and Rodrigues (2010) reported a density of AMF spores varying in function of seasons with a higher number of spores in August than in January. On the other hand, Coppetta et al. (2006) developed a study suggesting that the density of spores in soils is most dependent on the extent of root colonization between the AMF and the plant.

Negative and significant correlation ($p = 0.035$) was observed between AMF root colonization and spore density. Similar results were found by Radhika and Rodrigues (2010) when they studied thirty-six medicinal plant species.

Studies demonstrate that the seasonality of mycorrhizal colonization is usually a function of environmental conditions as temperature, humidity, phenology and physiological condition of the plant (Mohammad et al., 1998; Brundrett, 2002) and in this way, significant differences in both the root colonization and the density of AMF spores were observed in this study (Table 2).

Phyla Glomeromycota is identified mainly from the analysis of the formation, structure and germination of spores. In this study, *Glomus* was the most predominant genus within the diversity of AMF in the two sampling periods (June and November - Tables 3 and 4, respectively). *Acaulospora*, *Gigaspora*, *Scutellospora* and *Pacispora* were others genera found in this study, but with lower frequency (Table 5). Similar results were found by Radhika and Rodrigues (2010) in samples of soils cultivated with thirty-six medicinal plant species. The great diversity of AMF found in this study indicates that the plants studied form a symbiotic-mandatory association with the AMF, regardless of the period analyzed.

The indexes of Shannon diversity and Simpson dominance (Table 5) revealed great spore diversity among species of plants regardless of the sampling

Table 3. Taxonomy and number of spores per species of mycorrhizal fungi (Phylum *Glomeromycota*) in the 1st sampling period (June, 2011) determined according to Siqueira et al. (2010).

Plant	Order	Family	Genus	Species	Nº
Rosemary	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Glomus aff. Lamellosum</i>	23
				<i>Glomus mosseae</i>	11
				<i>Glomus microaggregatum</i>	30
				<i>Glomus aff. Tortuosum</i>	3
				<i>Gigaspora margarita</i>	2
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Glomus claroideum</i>	3
				<i>Glomus etunicatum</i>	7
				<i>Acaulospora sp.</i>	1
				<i>Acaulospora koskei</i>	2
				<i>Glomus macrocarpum</i>	2
Boldo	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Glomus aff. Lamellosum</i>	27
				<i>Glomus claroideum</i>	1
				<i>Glomus microaggregatum</i>	5
				<i>Glomus mosseae</i>	1
				<i>Glomus aff. Deserticola</i>	9
				<i>Glomus constrictum</i>	1
				<i>Glomus etunicatum</i>	10
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Acaulospora delicata</i>	5
				<i>Acaulospora koskei</i>	27
				<i>Acaulospora morrowiae</i>	13
		Gigasporaceae	<i>Gigaspora</i>	<i>Acaulospora (Entrophospora) colombiana</i>	3
				<i>Acaulospora sp. (scro-reticulata)</i>	1
				<i>Gigaspora margarita</i>	1
Scutellosporaceae	<i>Scutellospora</i>	<i>Scutellospora aff. Verrucosa</i>	1		
		<i>Glomus aff. Lamellosum</i>	7		
Chamomile	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Glomus mosseae</i>	7
				<i>Glomus claroideum</i>	10
				<i>Gigaspora margarita</i>	1
	Diversispolares	Gigasporaceae	<i>Gigaspora</i>	<i>Scutellospora calospora</i>	1
				<i>Scutellospora</i>	1
		Scutellosporaceae	<i>Scutellospora</i>	<i>Glomus mosseae</i>	4
				<i>Glomus macrocarpum</i>	13
Nasturtiums	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Glomus aff. Lamellosum</i>	4
				<i>Glomus tortuosum</i>	31
				<i>Glomus aff. Deserticola</i>	5
				<i>Glomus claroideum</i>	2
				<i>Glomus geosporum</i>	1
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Glomus invermaium</i>	5
				<i>Glomus etunicatum</i>	9
				<i>Acaulospora sp. (scro-reticulata)</i>	2
				<i>Acaulospora koskei</i>	2
				<i>Entrophospora infrequens</i>	4
Mint	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Gigaspora margarita</i>	1
				<i>Glomus claroideum</i>	3
				<i>Glomus aff. lamellosum</i>	3
				<i>Glomus mosseae</i>	1
				<i>Glomus macrocarpum</i>	1
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Glomus constrictum</i>	1
				<i>Glomus etunicatum</i>	3
				<i>Acaulospora scrobiculata</i>	4
		Gigasporaceae	<i>Gigaspora</i>	<i>Acaulospora delicata</i>	1
				<i>Acaulospora koskei</i>	1
Oregano	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Acaulospora delicata</i>	1
				<i>Gigaspora margarita</i>	2
				<i>Gigaspora decipiens</i>	1
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Glomus macrocarpum</i>	4
				<i>Glomus aff. lamellosum</i>	1
<i>Glomus claroideum</i>	1				
Diversispolares	Gigasporaceae	<i>Gigaspora</i>	<i>Acaulospora koskei</i>	2	
			<i>Gigaspora decipiens</i>	1	

Table 4. Taxonomy and number of spores per species of mycorrhizal fungi (Phylum *Glomeromycota*) in the 2nd sampling period (November, 2011) determined according to Siqueira et al. (2010).

Plant	Order	Family	Genus	Species	Nº	
Rosemary	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Glomus tortuosum</i>	5	
				<i>Glomus aff. lamellosum</i>	5	
				<i>Glomus mosseae</i>	2	
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Glomus aff. luteum</i>	3	
				<i>Glomus claroideum</i>	1	
				<i>Acaulospora (Entrophospora) colombiana</i>	2	
				<i>Gigaspora</i>	4	
Boldo	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Gigaspora decipiens</i>	4	
				<i>Glomus margarita</i>	1	
				<i>Glomus aff. lamellosum</i>	5	
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Glomus aff. luteum</i>	1	
				<i>Glomus claroideum</i>	3	
				<i>Acaulospora koskei</i>	3	
				<i>Acaulospora (Entrophospora) colombiana</i>	4	
		Scutellosporaceae	<i>Scutellospora</i>	<i>Scutellospora rubra</i>	1	
				<i>Scutellospora heterogama</i>	1	
Nasturtiums	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Glomus tortuosum</i>	6	
				<i>Glomus aff. lamellosum</i>	7	
				<i>Glomus geosporum</i>	1	
				<i>Glomus aff. luteum</i>	1	
				<i>Glomus constrictum</i>	1	
				<i>Glomus mosseae</i>	2	
				<i>Glomus microaggregatum</i>	30	
Oregano	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Gigaspora decipiens</i>	1	
				<i>Glomus aff. lamellosum</i>	3	
	Diversispolares	Acaulosporaceae	<i>Acaulospora</i>	<i>Glomus mosseae</i>	4	
				<i>Gigaspora</i>	1	
Mint	Glomerales	Glomeraceae	<i>Glomus</i>	<i>Gigaspora gigantea</i>	1	
				<i>Glomus aff. lamellosum</i>	3	
				<i>Glomus geosporum</i>	2	
				<i>Glomus macrocarpum</i>	1	
				<i>Glomus etunicatum</i>	1	
				<i>Glomus margarita</i>	1	
				<i>Glomus aff. luteum</i>	1	
Diversispolares	Gigasporaceae	<i>Gigaspora</i>	<i>Gigaspora ramisporophora</i>	2		
			Acaulosporaceae	<i>Acaulospora</i>	<i>Acaulospora scrobiculata</i>	4
			Pacisporaceae	<i>Pacispora</i>	<i>Pacispora robiginia</i>	1

period. Shannon index for rosemary increased from 0.066 in June to 0.329 in November, indicating greater diversity of AMF in the period of June compared to November. However, the opposite was observed for nasturtiums (Table 5).

The plants of mint, oregano and bold had similar AMF indexes of Shannon diversity and Simpson dominance in the two sampling periods studied, thus these plants were efficient in their symbiosis in both seasons.

Among all AMF genera found, *Glomus sp.* was the most frequent in all species and in both periods analyzed in this study (Table 5). The dominance of *Glomus sp.* in

soil cultivated with rosemary and sampled in June was 0.931 whereas in soil under nasturtiums and sampled in November was 0.96 (Table 5). Spores of *Glomus sp.* were observed in soils cultivated with all species studied. Their frequency was higher than 52%, reaching 98% in soils cultivated with nasturtiums.

Conclusions

All plants showed levels of AMF root colonization, and spore density of AMF decreased in June when compared

Table 5. AMF genera frequency, indexes of Shannon diversity and Simpson dominance (June and November, 2011).

Plantas	<i>Glomus Acaulospora Gigaspora Scutellospora Pacispora</i>					Shannon	Simpson
	sp.	sp.	sp.	sp.	sp.		
----- Relative frequency (%) -----							
June							
Rosemary	96.47	3.53	0	0	0	0.066	0.931
Boldo	51.88	46.22	0.95	0.95	0	0.341	0.482
Chamomile	92.30	0	3.85	3.85	0	0.141	0.854
Nasturtiums	89.15	9.65	1.20	0	0	0.165	0.804
Mint	54.55	31.82	13.63	0	0	0.419	0.417
Oregano	66.67	22.22	11.11	0	0	0.368	0.506
November							
Rosemary	72.72	9.10	18.18	0	0	0.329	0.570
Boldo	52.63	36.84	0	10.53	0	0.409	0.423
Nasturtiums	97.96	0	2.04	0	0	0.043	0.960
Mint	56.25	25	12.50	0	6.25	0.403	0.394
Oregano	77.78	11.11	11.11	0	0	0.296	0.629

to November. The taxonomic diversity of AMF varied among species of medicinal and seasoning plants studied. Spores of *Glomus* sp. were observed in all species studied. Their frequency was higher than 52%, reaching 98% in soils cultivated with nasturtiums.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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