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S. Fontenla · R. Godoy · P. Rosso · M. Havrylenko

Root associations in *Austrocedrus* forests and seasonal dynamics of arbuscular mycorrhizas

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Abstract Despite the recognized importance of arbuscular mycorrhizal (AM) fungi in forest ecosystems, their is little information about their occurrence in natural forest ecosystems of Patagonia. This study investigated root associations in two *Austrocedrus* forests and the seasonal dynamics of AM of *Austrocedrus chilensis*. Most of the vascular flora in both forests had arbuscular mycorrhizae (80.5 and 70.5% of species, respectively). The dynamics of mycorrhiza formation and spore number did not differ between these forests but varied with the season. Root colonization was lowest in September at the end of the quarter with the most rain, and spore numbers were highest in September and in December at the beginning of the dry season.

Key words *Austrocedrus chilensis* · Cypress · Patagonia · Spores · Seasonality

Introduction

Austrocedrus chilensis (D. Don) Florin et Boutelje is an important evergreen coniferous tree, endemic to temperate South American forests in Argentina and Chile (Donoso Zegers 1993). These forests form a discontinuous belt extending from the Andes to the border of the Patagonian steppe. According to Cabrera (1956), they belong to the Austral Region, Subantarctic Province, which is characterized by caducifolious and evergreen forest dominated by *Nothofagus* spp. and *A. chilensis.* At its eastern border, however, the vegetation is different and is dominated by xeric grasses and shrubs, with floristic elements of the Neotropical Region, Patagonia Province.

Despite the central role of soil microorganisms and root symbionts in the ecology of forest ecosystems, information on the distribution and abundance of these organisms is scarce (Perry et al. 1987; Allen 1991). This is especially true of natural forest ecosystems in Patagonia. The aim of this study was to survey root associations in *Austrocedrus* forests and to quantify the seasonal dynamics of the arbuscular mycorrhizae (AM) of *A. chilensis.*

Materials and methods

This study was conducted in the Nahuel Huapi National Park (40°8′ to 41°35′ S and 71°20′ to 71°57′ W), Argentina. A 1500-m^2 study plot was established in each of two forests (Limay and Llao-Llao) on Andosols in which allophane dominates middle horizons (Fitzpatrick 1984). The Limay plot (altitude 825 m), with an annual average precipitation of ca. 940 mm, was situated on the eastern slope of the Cerro Las Chivas. Its climate is dry, subhumid microthermal with little or no excess water, according to Thornthwaite's classification (Muñoz and Garay 1985). The Llao-Llao plot was located in the Cerro Llao-Llao foothills (altitude 800–850 m) with an annual average precipitation of ca. 2000 mm. Its climate is humid microthermal with moderate water deficiency and low thermal concentration in summer (Muñoz and Garay 1985). December through February is the warm, dry season and most precipitation (60–70%) occurs during the cold winter months of June through August.

The composition of the plant community in each plot was characterized according to the taxonomy and nomenclature of Correa (1971) and Dimitri (1972). Sørensen's index of similarity was calculated for the two forest communities (Sørensen 1948). The densities of *A. chilensis* were calculated in each plot.

The mycorrhizal status of the total vascular flora was determined from samples of three individuals of each species collected in December 1990. Root collection depended upon plant size. Small plants were collected whole, but tree roots were sampled only when they were clearly attached to the tree. All root samples were cleared and stained according to the method of Bevege (1968). Roots were considered to have AM when arbuscules were observed.

S. Fontenla (✉) · P. Rosso · M. Havrylenko
Centro Regional Universitario Bariloche,
Universidad Nacional de Comahue,
Quintral 1250 (CP 8400) S.C. de Bariloche,
Rio Negro, Argentina
Fax: +54-944-22111; e-mail: soniaf@uncmai.edu.ar

R. Godoy
Instituto de Bótanica, Universidad Austral de Chile, Casilla 567,
Valdivia, Chile

To examine the mycorrhizal dynamics of *A. chilensis*, the fine roots of nine different trees at each time point were sampled at 3-month intervals for 1 year. All sampled individuals were live, mature trees of 10–40 cm DBH (diameter at breast height). The gridline intersect method of Giovannetti and Mosse (1980) was used to quantify the frequency of AM colonization. One hundred root segment intersections with a gridline were examined per sample and the frequency of colonization was determined as the percentage of intersections with arbuscules. Root colonization was compared by two-way analysis of variance (ANOVA) after arcsine transformation.

Soil (ca. 400 g) from around the roots of the root-sampled *A. chilensis* trees was collected every 3 months for 1 year and the total number of spores of AM fungi determined by wet-sieving (45–500 μm) and sucrose density centrifugation (48% w/v; Walker et al. 1982). Three 50-g sub-samples per soil sample were used for spore extraction (Daniels and Skipper 1982). These counts were analyzed by two-way ANOVA.

Results

Austrocedrus chilensis dominated both plots, with densities of 0.117 trees per m^2 in Llao-Llao and 0.077 in Limay. The flora comprised 71 species, with 44 and 40 species present in the Limay and Llao-Llao plots, respectively (Table 1). Sørensen's index of similarity between the plots was 33%.

Four different types of mycorrhizal associations were found. In Llao-Llao, 87% of the species had mycorrhizae, made up of 81% AM and 6% (one species each) ectomycorrhizae, ericoid mycorrhizae, or orchid mycorrhizae. Only AM associations (with 71% of species) were found in the Limay plot.

As in previous studies (Fontenla et al. 1991), *A. chilensis* had only AM. Fig. 1A shows the frequency of AM colonization by season. The colonization frequency did not differ significantly between the two plots ($F = 3.579$, $P = 0.0768$), but there was a significant effect of season ($F = 4.476$, $P = 0.0183$); the interaction was not significant ($F = 1.954$, $P = 0.1617$). Considering the mean number of AM fungus spores for each plot by season (Fig. 1B), the plots did not differ significantly ($F = 0.809$, $P = 0.3913$), but there was a significant effect of season ($F = 11.654$, $P = 0.0003$); the interaction was not significant ($F = 0.507$, $P = 0.6829$).

Discussion

The forests studied are dominated by *A. chilensis* which forms AM. *A. chilensis* is similarly mycorrhizal elsewhere in Nahuel Huapi National Park (unpublished data) and in temperate forests in southern Chile (Godoy et al. 1994). The plant communities in the two forests differed considerably, as shown by the low index of similarity of floristic composition (33%), which reflects the substantial difference in total precipitation.

The majority of the vascular flora of both plots had mycorrhizal associations, with AM clearly dominating. Only Llao-LLao had other types of associations, which involved few species. This accords with previous find-

Table 1 Plant species, life-forms, and root associations in two *Austrocedrus chilensis* forests. A blank space indicates the absence of a species from that forest plot. *AM* arbuscular mycorrhiza, *E* ectomycorrhizae, *ER* ericoid mycorrhizae, *F* Frankia nodules, *NM* no mycorrhizal infection, *O* orchid mycorrhizae, *R* Rhizobium nodules

Plant species	Root associations	
	Llao-Llao plot	Limay plot
Ferns		
Adiantaceae		
Adiantum chilense Kaulf.	AM	
Aspidiceae		
Rumohra adiantiformis (Forst.) Ching.	AM	
Herbs		
Amaryllidaceae		
Alstroemeria aurea Graham	AM	
Asclepiadaceae		
Cynanchum nummularifolium Hooker et Arn.		AM
Asteraceae		
Crepsis setosa Hall. f.		AM
Leuceria achillaeifolia Hooker et Arnott		NM
Solidago chilenesis Meyer		AM
Boraginaceae		
Cynoglossum creticum Miller		AM
Caryophyllaceae		
Cerastium arvense Linné		NM
Silene andicola Gillies ex Hook. et Arnot	NM	
Stellaria media (L.) Villars	AM	
Cyperaceae		
Carex aphylla Knuth	NM	
Carex boelckeiana Barros	NM	NM
Uncinia richleriana Steudel	NM	
Fabaceae		
Vicia nigricans Hooker et Arnot	AM + R	AM + R
Geraniaceae		
Geranium sessiliflorum Cavanilles	AM	AM
Hydrophyllaceae		
Phacelia magellanica (Lam.) Cav.		NM
Iridaceae		
Sisyrinchium sp.		NM
Loasaceae		
Caiophora sylvestris (Poepp.) Urban et Gilg		NM
Loasa berghii Hiernonymus		NM
Orchidaceae		
Chloraea virescens (Willd.) Lindley	O	
Oxalidaceae		
Oxalis valdiviensis Barnéoud		AM
Poaceae		
Aira caryophyllea Linné		NM
Festuca argentina (Speg.) Parodi		AM
Festuca pallescens (St. Yves) Parodi		AM
Hordeum comosum Presl.		AM
Koeleria vurilochensis Caldéron		NM
Pao lanuginosa Poiret ap. Lamarck		AM
Trisetum flavescens (Linné) Palisot de Beauvois		AM

Table 1 (continued)

Plant species	Root associations	
	Llao-Llao plot	Limay plot
Polygonaceae		
Rumex acetosella Linné		NM
Portulacaceae		
Montia perfoliata (Donn) Howell		NM
Rhamnaceae		
Discaria articulata (Phil.) Miers		M+F
Rosaceae		
Acaena ovalifolia Ruiz et Pavón	AM	
Acaena pinnatifida Ruiz et Pavón		AM
Fragaria chiloensis (L.) Duchesne	AM	AM
Rubiaceae		
Galium aparine L.		NM
Relbuneum hypocarpium (L.) Hemsl.	AM	
Relbuneum richardianum (Gill. ex Hook. et Arn.) Hicken		AM
Scrophulariaceae		
Calceolaria crenatiflora Cav.	AM	
Calceolaria polyrrhiza Cav.		AM
Verbascum thapsus L.		AM
Umbelliferae		
Eryngium paniculatum Cav. et Dombey ex F. Delaroche	AM	
Osmorhiza chilensis Hooker et Arnott	AM	AM
Valerianaceae		
Valeriana laxiflora DC.	AM	
Violaceae		
Viola maculata Cavanilles	AM	AM
Shrubs		
Anacardiaceae		
Schinus patagonicus (Phil.) Johnston	AM	AM
Asclepiadacea		
Cynanchum sp.	AM	
Asteraceae		
Baccharis racemosa (Ruiz et Pav.) De Candolle	AM	
Mutisia decurrens Cavanilles	AM	AM
Mutisia spinosa Ruiz et Pavón	AM	AM
Senecio bracteolatus Hooker et Arnott		AM
Berberidacea		
Berberis buxifolia Lamarck	AM	AM
Berberis darwinii Hooker	NM	
Celastraceae		
Maytenus chubutensis (Speg.) L. O'Donnell et Sleumer	AM	AM
Maytenus magellanica (Lam.) Hooker fil.	AM	
Elaeocarpaceae		
Aristotelia chilensis (Molina) Stuntz	AM	
Ericaceae		
Pernettya mucronata (L.f.) Gaud.	ER	
Escalloniacea		
Escallonia rubra (Ruiz et Pavón)	AM	
Flacourtiaceae		
Azara microphylla J. D. Hooker	AM	
Grossulariaceae		
Ribes magellanicum Poiret	AM	AM
Poaceae		
Chusquea culeou Desvaux	AM	
Rhamnaceae		
Colletia hystrix Clos	AM+F	
Discaria articulata (Phil.) Miers		AM+F
Discaria chacaye (G. Don) Tortosa		AM+F
Rosaceae		
Rosa rubiginosa L.		AM
Thymeleacea		
Ovidia andina (Poepp.) Meissner	AM	
Trees		
Araliacea		
Pseudopanax laetevirens (Gay) Franchet	AM	
Celastraceae		
Maytenus boaria Molina		AM
Cupresaceae		
Austrocedrus chilensis (Don) Florin et Boutelje	AM	AM
Fagacea		
Nothofagus dombeyi (Mirb.) Blume	E	
Myrtaceae		
Luma apiculata (DC.) Burret	AM	
Proteaceae		
Lomatia hirsuta (Lam.) Diels		NM
Rhamnaceae		
Discaria chacaye (G. Don) Tortosa		AM+F

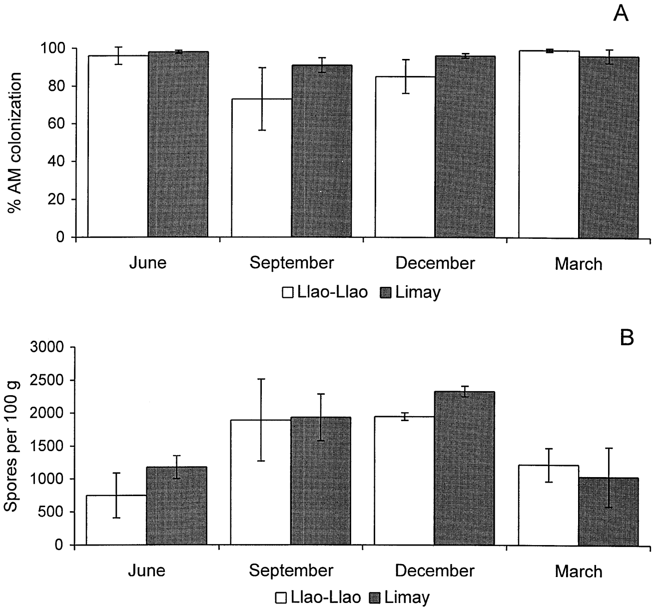

Fig. 1A, B Occurrence of arbuscular mycorrhizae on *Austrocedrus chilensis* and of arbuscular mycorrhizal fungus spores in two Patagonian forests. **A** Percent root colonization; **B** Mean number of spores per 100 g dry soil by month of collection (*error bars* are the error of estimate) ings in temperate woods in the Austral area of Chile (Carrillo et al. 1992), in temperate forests at other latitudes (Mohankumar and Mahadevan 1987; Berliner and Torrey 1989; Mayr and Godoy 1989) and in general (Trappe 1987; Allen 1991; Brundrett 1991).

Most plants in the world within the same families and genera are usually associated with the same mycorrhiza type as we observed in this study (Table 1; Brundrett 1991). However, some plant species benefit from mycorrhizal association only under certain conditions (Koide and Li 1990) and thus occasional species in the families Asteraceae, Berberidaceae, Hydrophyllaceae, Poaceae and Rubiaceae may not have mycorrhizae (Harley and Harley 1987a,b, 1990). We found no colonization in two species of Poaceae (*Aira caryophyllea* and *Koeleria vurilochensi*) and in one species of Rubiaceae (*Galium aparine*). In our study, *Berberis darwinii* (Berberidaceae) lacked mycorrhizae, but *B. buxifolia* had AM. Both of these species form AM in southern Chile and AM are present in other species of this family (Carrillo et al. 1992; Godoy et al. 1994).

A third group of species belong to families considered non-mycorrhizal, e.g., Caryophyllaceae, Cyperaceae, Polygonaceae, Portulacaceae and Proteaceae (Tester et al. 1987; Harley and Harley 1990). However, we found *Stellaria media* (Caryophyllaceae) to have AM. Arbuscular mycorrhizas have also been observed in *Stellaria cuspidata* (Carrillo et al. 1992).

We found tripartite symbiotic associations, both AM and actinorrhizae, in three species of Rhamnaceae, as well as a tripartite symbiosis with AM and Rhizobium in *Vicia nigricans* (Fabaceae).

In our study of two forests with similar seasonality, slightly dissimilar soils (Havrylenko et al. 1989), different precipitation regimes, and different vegetation, we observed similar seasonal patterns of mycorrhizal dynamics. When the number of spores in soil was high, the frequency of colonization was low, and vice versa. Louis and Lim (1987) also observed that high root colonization followed high numbers of spores in surrounding soil. Several factors could contribute to such a pattern, including seasonal sporulation (Sutton and Barron 1972), seasonal variation in the development of the host plant (Giovannetti 1985), and seasonal nutrient availability (Louis and Lim 1987).

This is the first report of which we are aware concerning root associations of forest trees in Patagonia, Argentina. It contributes data necessary for future studies of the relationship of microorganisms to the other biological and environmental components of this complex ecosystem.

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