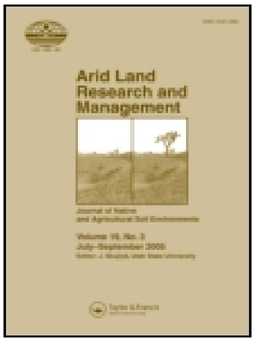

Arid Soil Research and Rehabilitation

ISSN: 0890-3069 (Print) (Online) Journal homepage: http://www.tandfonline.com/loi/uasr19

AM fungal abundance and activity in a chronosequence of abandoned fields in a semiarid mediterranean site

A. Roldan , C. Garcia & J. Albaladejo

To cite this article: A. Roldan , C. Garcia & J. Albaladejo (1997) AM fungal abundance and activity in a chronosequence of abandoned fields in a semiarid mediterranean site , Arid Soil Research and Rehabilitation, 11:3, 211-220, DOI: 10.1080/15324989709381474

To link to this article: http://dx.doi.org/10.1080/15324989709381474

Published online: 09 Jan 2009.

AM Fungal Abundance and Activity in a Chronosequence of Abandoned Fields in a Semiarid Mediterranean Site

A. ROLDAN
C. GARCIA
J. ALBALADEJO

Centro de Edafología y Biología Aplicada del Segura/CSIC
Murcia, Spain

When soils of semiarid areas are used for agriculture, their arbuscular mycorrhizal (AM) fungal propagules undergo a series of changes, and when agricultural land is abandoned it is very difficult for these populations and the plant cover to recover. We studied soils that have been abandoned for different lengths of time (3–45 years) in a homogeneous semiarid area in order to observe changes in their physical and chemical properties and in the AM fungi propagules, comparing them with soils that continue to be cultivated and a soil that has never been cultivated. The data we collected clearly showed that agricultural use reduces soil fertility and lowers AM fungi populations compared to the soil kept in its natural state. After abandonment, there is a 5-year period when the soils undergo a greater degree of degradation after which they slowly recover, with AM fungi propagules reaching values similar to those of the virgin soil after 45 years or so. Although the physical and chemical properties followed a similar pattern of recovery, it was not possible to establish significant correlations between these parameters and the recovery of the AM fungi. Except in isolated cases, the distribution of AM fungi did not seem to be influenced by position on the landscape, with their recovery appearing to be more influenced by the presence of host plants. In this sense, the rhizosphere of Anthyllis cytisoides *showed normal values of root infection and spore numbers even during the initial stages following abandonment.*

Keywords AM spores, *Anthyllis cytisoides*, root infection, soil characteristics

In the semiarid areas of southeastern Spain, changing patterns of economic development and gradual urbanization have led to the abandonment of large tracts of poorly productive land, a tendency that has been accentuated recently by the agricultural policies of the European Union. Recovery of these abandoned lands is particularly difficult in arid and semiarid regions. When a soil is put to agricultural use it undergoes a series of physical, chemical, and microbiological changes, one of the most important of which is the changes that affect root-inhabiting microorganisms. Arbuscular mycorrhizal (AM) fungi play an impor-

Received 24 September 1996; accepted 10 December 1996.

The research for this article was carried out as part of the Mediterranean Desertification and Land Use (MEDALUS) research project (EPOC-CT90-0014-SMA). We are indebted to Prof. J. B. Thornes and Dr. J. Obando (both at King's College London) for their advice during the field sites selection.

Address correspondence to Dr. A. Roldán, Centro de Edafología y Biología Aplicada del Segura/CSIC, Apartado 4195, 30080 Murcia, Spain.

Arid Soil Research and Rehabilitation, 11:211–220, 1997

0890-3069/97 $12.00 + .00

tant role in rehabilitating arid soils (Allen, 1989). Their symbiosis is the most common in arid plant species (Newman & Reddell, 1987) and has been repeatedly demonstrated to improve the uptake of nutrients and water by plants (Harley & Smith, 1983; Lansac et al., 1995).

Although the AM fungi–host plant symbiosis is not specific, there may be a certain degree of compatibility (Smith & Gianinazzi-Pearson, 1988; Díaz et al., 1992). For this reason spontaneous processes of plant colonization are limited in abandoned lands since, although AM fungi are presumably present, their quantity and/or quality may not be sufficient for the recovery of spontaneous vegetation (Barea, 1990). Despite the importance of mycorrhizal symbiosis in plant cover development in abandoned soils, the process has been little studied (Abbott & Robson, 1991), especially in semiarid zones. In this study we evaluate changes in the AM fungi propagules in fields abandoned for different lengths of time in a semiarid area of southeastern Spain.

Materials and Methods

Field Study Site

The survey was carried out in the Rambla del Chortal experimental area on the La Torrecilla range in Murcia Province (southeastern Spain) at a mean altitude of 600 m. The climate is semiarid Mediterranean with an average annual rainfall of 300–350 mm occurring mostly in autumn and spring. The mean annual temperature is 16°C. The predominant soils are developed from fillites with a sandy loam texture (Sánchez et al., 1980).

In an area measuring approximately 2 km^2, 7 sampling sites were chosen to include fields abandoned for different times, a zone in which barley was cultivated, and a zone covered with natural vegetation (*Quercus rotundifolia* Lam. forest). Some characteristics of the zones are shown in Table 1. The dates when the fields were abandoned were provided by the landowners; the date for site 5 is approximate and may differ from the actual date by 5 years.

Table 1
Characteristics of the sampling sites in Rambla del Chortal

Site	Length of abandonment (years)	Catena length (m)	Average slope (degrees)	Exposure	Predominant soils[a]
S1	3	45	16	SE	Lithic Xeric Torriorthent
S2	7	30	11	SE	Lithic Xeric Torriorthent
S3	15	47	12	SE	Lithic Xeric Torriorthent
S4	30	193	22	SE	Entic Haploxeroll
S5	45	61	19	SE	Entic Haploxeroll
S6	Natural Control	50	22	SE	Entic Haploxeroll
S7	Cultivated Control	35	16	SE	Xeric Torriorthent

[a]According to Soil Survey Staff (1975).

Sampling Procedures

The areas were sampled in April 1994. In sites S1 to S5 three transects perpendicular to the slope were made in the upper, middle, and lower parts. Five soil samples (0–15 cm depth) were taken in each transect in the rhizosphere of randomly chosen patches of vegetation (15 samples per site). From sites S6 and S7 five rhizosphere soil samples were taken without being divided by transects. Five samples of the rhizosphere of *Anthyllis cytisoides* L. were taken from sites S1 to S6.

Measurements

The soil samples were air-dried and sieved with a 2-mm screen in order to separate roots and stones. Four subsamples were taken of separated roots, and the percentage of root length colonized by AM fungi was calculated by the gridline intersect method (Giovannetti & Mosse, 1980) after staining with trypan blue (Phillips & Hayman, 1970).

Spore numbers in soil were determined after processing three 50-g sieved soil subsamples by the wet sieving and decantation procedure outlined by Gerdemann and Nicolson (1963). Only those spores appearing viable were counted. The water content of the soils was measured gravimetrically, and spore numbers were calculated per 100 g dry soil.

For each locality, portions of soil samples were mixed and analyzed to characterize the physical and chemical properties of the soils. Electrical conductivity and pH were measured in a 1/5 (solid/liquid) aqueous extract.

Total organic carbon (TOC) was determined by the method of Yeomans and Bremmer (1989). The carbohydrate content was determined spectrophotometrically as the blue-green complex formed when sugars are heated with anthrone in sulfuric acid (Brink et al., 1960).

Total P and K contents were determined by calorimetry (Murphy & Riley, 1962) and flame photometry (Schollemberger & Simon, 1954), respectively, after nitric–perchloric digestion of the samples.

Results and Discussion

Physical and Chemical Characteristics of the Sites

The pH of the soils studied ranged from 7.93 to 8.84 (Table 2), which probably reflects the $CaCO_3$ content of the soils (Thomas, 1967). The highest pH values occurred in sites S2 and S3, perhaps indicating the lesser degree of acidity from root exudates. The soils abandoned for the longest period of time (S4 and S5) showed the lowest pH values, perhaps because of the incorporation of plant debris, which subsequently decomposed.

Although electrical conductivity values were low compared to that of adjacent semiarid soils developed on loams (García et al., 1994), they did not appear to change with the duration of abandonment.

The principal source of organic matter in the soils is plant debris, both from the aerial parts incorporated into the soil after the growth periods and from the roots and rhizomas that remain in the soil. Other sources are water-soluble organic compounds and polysaccharides excreted by roots, together with contributions from microbial and animal activities. The data point to a great difference between the TOC of the natural (S6) and cultivated control (S7); agricultural activity clearly reduced the soil organic matter content. The TOC of the abandoned areas (S1–S5) increased gradually with time, with the TOC of

Table 2
Analytical data of soils from the sites studied

Sites (years of abandonment)	Organic C ($g\ kg^{-1}$)	Carbohydrates ($mg\ kg^{-1}$)	Total K ($mg\ kg^{-1}$)	Total P ($mg\ kg^{-1}$)	pH	Electrical conductivity ($S\ m^{-1}$)
S1 (3)	13.1c	967d	217b	241c	8.47b	0.14b
S2 (7)	5.5e	370e	246ab	327a	8.81a	0.08c
S3 (15)	9.2d	505e	223b	205d	8.84a	0.08c
S4 (30)	18.8b	1421c	219b	230cd	7.93d	0.10c
S5 (45)	23.2a	1788b	165c	279b	8.26a	0.17a
S6 (natural control)	23.3a	2044a	282a	336a	8.44b	0.13b
S7 (cultivated control)	7.7d	403e	252ab	149e	8.55b	0.09c

Note. Values in columns sharing a letter do not differ significantly at $p < .05$ by Duncan's test.

S5 being equal to that of land kept under natural vegetation. The recovery of organic matter content in the abandoned lands was very slow and took 45 years before levels were reached similar to those occurring before human intervention.

One of the most important carbon fractions that indicates a soil's fertility is that of total carbohydrates. Many authors have shown the correlation between the percentage of stable aggregates (necessary for maintaining soil structure) and the total carbohydrate content (Cheshire et al., 1983; Díaz et al., 1994; Roldán et al., 1994). The behavior of total carbohydrates parallels that of TOC, as is evident from the high correlation coefficient between both parameters ($r = 0.949$; $p < .0001$, Table 3).

The concentrations of K and P recorded are usual for soils in this zone (García et al., 1994). Note that the cultivated soil showed a high K content (probably due to fertilization)

Table 3
Spearman rank correlations among soil analytical variables

Component	Organic C	Carbohydrates	Total K	Total P	pH	Electrical conductivity ($S\ m^{-1}$)
Organic C	1	0.949^{c}	−0.252	-0.390^{a}	-0.624^{c}	0.662^{c}
Carbohydrates		1	−0.115	0.453^{b}	-0.587^{c}	0.684^{c}
Total K			1	0.001	0.203	-0.341^{a}
Total P				1	−0.005	0.361^{a}
pH					1	-0.312^{a}
Electrical conductivity						1

[a]Significant at $p < .05$.
[b]Significant at $p < .01$.
[c]Significant at $p < .001$.

and low concentration of P. These nutrients did not show a clear pattern in the abandoned soils, although the highest values were always apparent in the natural wooded site.

Our data show that agricultural activity impoverishes the soil. This impoverishment (particularly the organic matter content) continues immediately after abandonment, perhaps due to erosion phenomena (Albaladejo et al., 1996). Recovery of soil fertility and quality is slow, and only after 45 years can the soil be regarded as similar to that of the natural control.

AM Fungal Abundance and Activity

The number of AM fungi spores varied significantly with the duration of abandonment (Table 4). The lowest concentration was observed in site S1 (abandoned for 3 years), with the levels rising gradually until reaching a maximum in sites S5 and S6 (45 years after abandonment and natural control, respectively). Note that the number of spores remained stable between 15 and 30 years after abandonment (S3 and S4); the data for these same sites showed no significant difference with respect to those for the cultivated control (S7). As regards their position on the slope, this factor did not seem to modify the number of AM fungi spores with the exception of site S2, in the lower part of which significantly higher values were noted. In S5, on the other hand, significantly lower values were observed in the lower part.

The percentage of root infection behaved quite differently (Table 4), with the lowest values occurring in the cultivated site and S1 with no significant difference between them. These values increased positively and significantly (but still showed no difference between each other) in older sites (S2, S3, and S4). Site S5 showed root infection percentages higher than the other sites and very near those of the natural control (S6). As regards their position in the slope, the differences were even less than in the case of spore number, and in no case did this factor introduce significant differences.

Agricultural practices can seriously affect the natural microbial populations in a soil, and such an alteration can be very evident in the case of AM fungi due to their obligately

Table 4
AM fungi spore numbers and percentage of infected root length in abandoned fields of different ages

Sites (years of abandonment)	AM spores/100 g dry soil at position in the slope			Percent infected root length at position in the slope		
	Top	Middle	Base	Top	Middle	Base
S1 (3)	9a	8a	10a	14A	10A	148A
S2 (7)	60b	69b	114c	39BC	38BC	36B
S3 (15)	118c	115c	130c	44C	44C	438C
S4 (30)	111c	119c	118c	45C	46C	42C
S5 (45)	204de	223e	180d	62D	66D	68DE
S6 natural control		207e			74E	
S7 cultivated control		110c			17A	

Note. Values are means of five replicates. Spore data sharing a lowercase letter do not differ significantly at $p < .05$ by Duncan's test. Infection data sharing a capital letter do not differ significantly at $p < .05$ by Duncan's test.

symbiotic nature, which limits their presence to host plants. In our study the cultivated control showed normal spore numbers, although the percentage of root infection was low. The cultivation of barley over many years (the exact time is unknown) may have favored the proliferation of an adapted AM fungi population. For this reason, spore numbers remained high after harvesting (when sampling took place), although root infection of the few plants that colonize the land between growing seasons was the lowest of all the sites studied.

The study zone is subjected to a Mediterranean rainfall regime with intense autumn precipitations that result in severe erosion of the soil (Albaladejo, 1990). Under such conditions a soil that ceases to be cultivated rapidly loses its surface horizons rich in organic matter (Milton et al., 1994), while the percentage of plant cover and the AM fungi diminish (Aziz & Habte, 1990; Roldán & Albaladejo, 1993), thus possibly explaining why soils S1 and S2 show the lowest number of AM propagules.

The renewal of endomycorrhizal colonization is conditioned by several factors (Allen, 1987, 1988). It is widely accepted that the spores of AM fungi are dispersed by wind and animals in semiarid areas (Warner et al., 1987; Allen et al., 1989) although their establishment in a particular site will depend on a soil's characteristics (Abbott & Robson, 1991; Díaz et al., 1992) and on the established vegetation (Gemma & Koske, 1990). Our study pointed to a gradual improvement in the physical and chemical properties of the soil of abandoned plots, but no significant correlation ($p < .05$) existed between these parameters and AM fungi propagules. Neither did the position of a site on the slope appear to be a determining factor in endomycorrhizal fungi colonization, as would be expected if dispersion by wind were the determining factor in recolonization.

Changes in AM fungi populations after severe disturbance can be linked to colonization by mycorrhizal plant species (Gemma & Koske, 1990. Roldán & Albaladejo, 1993). Our study shows the data corresponding to spore numbers and root infection in the sites studied obtained by random sampling, that is, with no reference to the number of mycorrhizal plants present at the sampling point. For comparison purposes, we also sampled in the rhizospheres of *Anthyllis cytisoides,* which is a pioneering legume in the colonization of degraded soils and totally dependent on endomycorrhizal symbiosis to survive (Díaz et al., 1992; Herrera et al., 1993). This plant was present in all the sampling sites except in the cultivated site (S7).

After the abandonment of a cultivated soil (S1), the rhizosphere of *A. cytisoides* showed a much higher AM fungi spore number than samples of mixed rhizospheres from the same site (Figure 1). This suggested that the decrease in the number of spores after abandonment was influenced by the absence of mycorrhizal plants among the first colonizers, which in our study zone were mainly representatives of the mostly nonmycorrhizal Brassicaceae family. The number of spores in the *A. cytisoides* rhizosphere remained stable even after 7 years of abandonment (S2) but increased gradually in sites S3–S6. Note that in S5 and in the natural control, the levels of AM fungi propagules were significantly higher in the mixed rhizospheres than in the *A. cytisoides* rhizosphere.

AM infection in pure stands of *A. cytisoides* always exceeded 50% (Figure 2) and in all cases was significantly higher than in mixed stands except in the natural control, where the differences were not statistically significant. This seems to suggest that soil has a sufficiently high potential for mycorrhizal infection even in the early stages of abandonment, although it must not be forgotten that the sampling of *A. cytisoides* rhizospheres was not random and that this plant, due to its pioneering characteristics, probably colonizes the few patches of soil where mycorrhizal inocula were present. This should not be extrapolated to the rest of the study area.

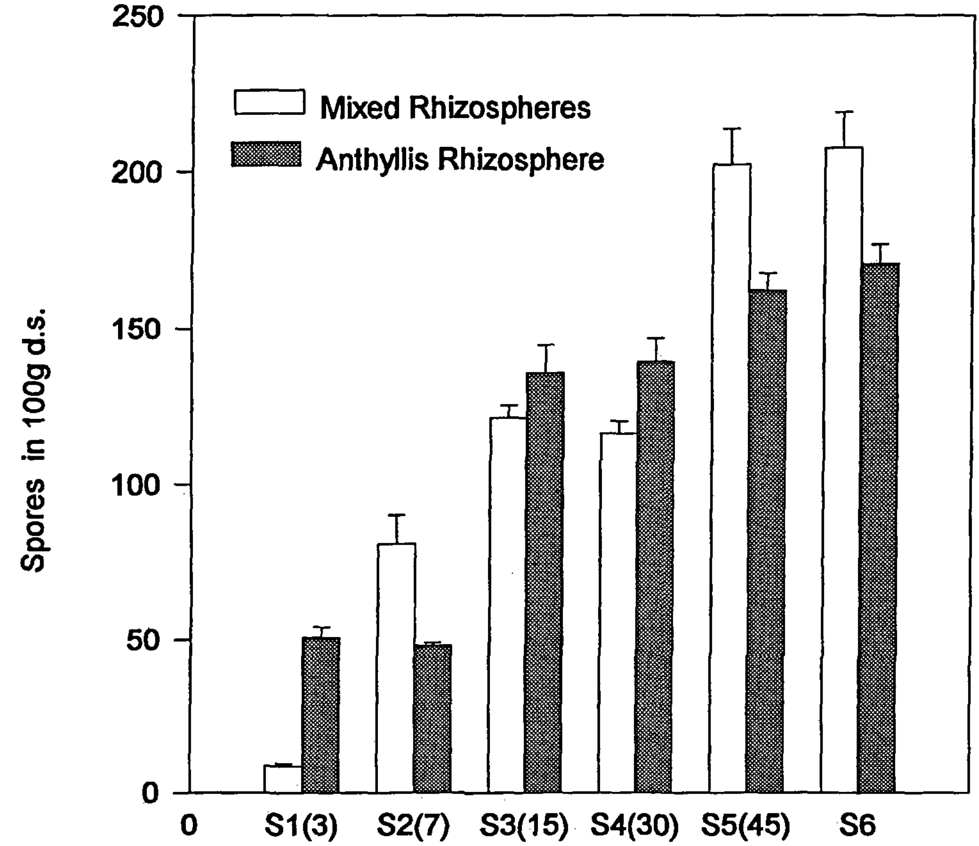

Figure 1. Comparison between AM fungi spore numbers in mixed rhizospheres and *Anthyllis cytisoides* rhizosphere in abandoned lands. Bars represent standard error ($n = 15$ for mixed rhizospheres, $n = 5$ for *A. cytisoides* rhizosphere).

Conclusions

Abandonment of a cultivated area is bound to strongly affect the edaphic ecosystem. In semiarid zones, the loss of soil fertility and of the mycorrhizal component is very pronounced during the initial stages of abandonment. Reestablishment of suitable conditions is a very slow process, which includes edaphogenetic processes and recolonization by vascular plants and their symbiotic mycorrhizal fungi. In the area studied, AM fungi propagules recovered gradually and only reached levels comparable to the uncultivated control after 45 years. This recovery of AM fungi propagules seemed to be connected with the progressive introduction of mycorrhizal host plants, which, in turn, may depend on an improvement of the soil's physical and chemical characteristics. It seemed clear that any attempt to regenerate the plant cover of abandoned soils should take all these factors into consideration and that the proper management of mycorrhizal symbiosis through the introduction of inoculated plants should be investigated as a possible mechanism for accelerating the regeneration process of soils.

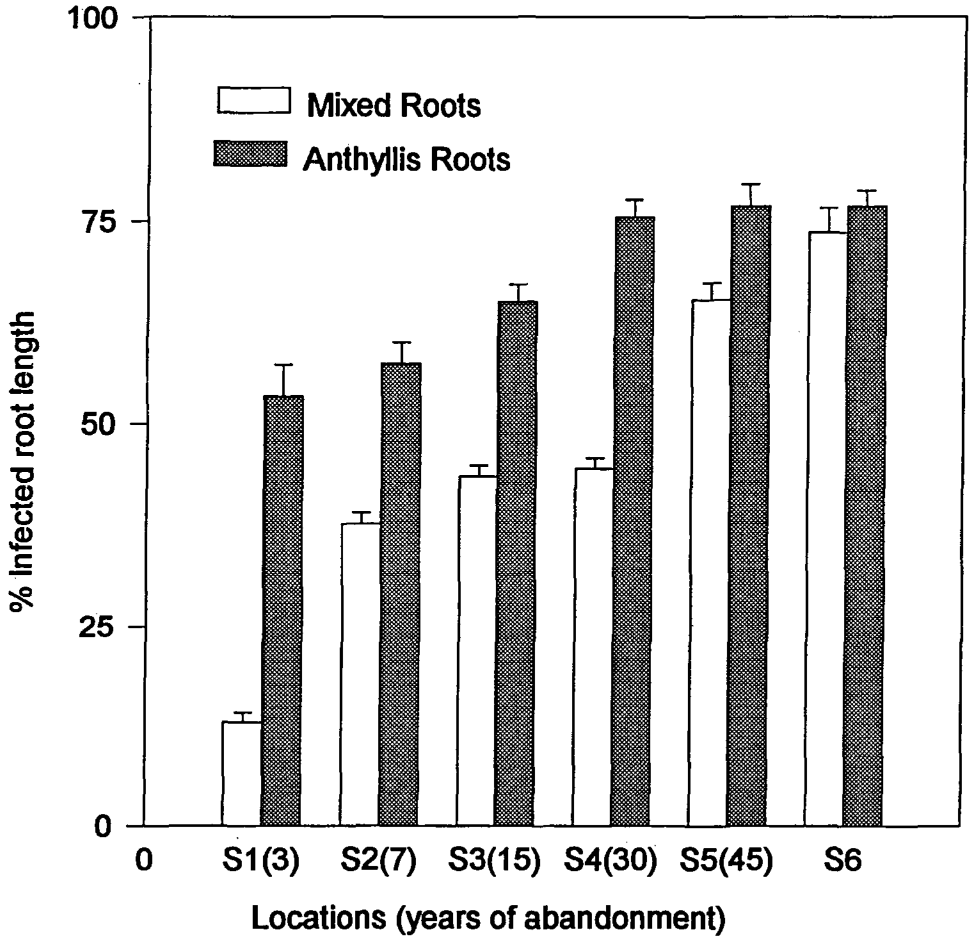

Figure 2. Comparison between percentage of colonizated root length in mixed rhizospheres and *Anthyllis cytisoides* rhizosphere in abandoned lands. Bars represent standard error (n = 15 for mixed rhizospheres, n = 5 for *A. cytisoides* rhizosphere).

References

Abbott, L. K., and A. D. Robson. 1991. Factors influencing the occurrence of vesicular-arbuscular mycorrhizas. *Agriculture, Ecosystems and Environment* 35:121–150.

Albaladejo, J. 1990. Impact of the degradation processes on soil quality in arid Mediterranean environment, pp. 133–215, in J. L. Rubio and R. J. Rickson, eds., *Strategies to Combat Desertification in Mediterranean Europe*. Commission of the European Communities, Luxembourg.

Albaladejo, J., V. Castillo, and A. Roldán, 1996. Rehabilitation of degraded soils by water erosion in semiarid environments, pp. 265–278, in J. L., Rubio and A. Calvo, eds., *Soil Degradation and Desertification in Mediterranean Environments*. Geoforma Ediciones. Logroño.

Allen, M. F. 1987. Re-establishment of mycorrhizas on Mount St. Helens: Migration vectors. *Transactions of the British Mycological Society* 88:413–417.

Allen, M. F. 1988. Re-establishment of VA mycorrhizas following severe disturbance: Comparative patch dynamics of a shrub desert and a subalpine volcano. *Proceedings Royal Society of Edinbourgh* 94:63–71.

Allen, M. F. 1989. Mycorrhizae and rehabilitation of disturbed arid soils: Processes and practices. *Arid Soil Research and Rehabilitation* 3:229–241.

Allen, M. F., L. E. Hipps, and G. L., Wooldridge, 1989. Wind dispersal and subsequent establishment of VA mycorrhizal fungi across a successional arid landscape. *Landscape Ecology* 2:165–171.

Aziz, T., and M. Habte. 1990. Enhancement of endomycorrhizal activity through nitrogen fertilization in cowpea grown in an oxisol subjected to simulated erosion. *Arid Soil Research and Rehabilitation* 4:131–139.

Barea, J. M. 1990. Micorrizas vesículo-arbusculares, pp. 271–278, in J. Casadesus and F. Ruiz-Berraquero, eds., *Microbiologia 1990*. Secretariado de Publicaciones de la Universidad de Sevilla, Sevilla.

Brink, R. H., P. Dubar, and D. L. Linch. 1960. Measurement of carbohydrates in soil hydrolizates with anthrone. *Soil Science* 89:157–166.

Cheshire, M. V., G. P. Sparling, and C. M. Mundie. 1983. Effect of periodate treatment of soil on carbohydrate constituents and soil aggregation. *Journal of Soil Science* 34:105–112.

Díaz, G., A. Roldán, and J. Albaladejo. 1992. Influencia del tipo de suelo sobre las pautas de colonización y eficiencia en la simbiosis micorrícica de seis especies de *Glomus, Cryptogamie Mycologie* 13:47–56.

Díaz, E., A. Roldán, A. Lax, and J. Albaladejo. 1994. Formation of stable aggregates in degraded soil by amendment with urban refuse and peat. *Geoderma* 63:277–288.

García, C., T. Hernandez, and F. Costa. 1994. Microbial activity in soils under Mediterranean environmental conditions. *Soil Biology and Biochemistry* 26:1185–1191.

Gemma, J. N., and R. E. Koske. 1990. Mycorrhizae in recent volcanic substrates in Hawaii. *American Journal of Botany* 77:1193–1200.

Gerdemann, J. W., and J. H. Nicolson, 1963. Spores of mycorrhizal *Endogone* species extracted from soil by wet sieving and decanting. *Transactions of the British Mycological Society* 46:235–240.

Giovannetti, M., and B. Mosse. 1980. An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. *New Phytologist* 84:489–500.

Harley, J. L., and S. E. Smith. 1983. *Mycorrhizal symbiosis*. Academic Press, London.

Herrera, M. A., C. P. Salamanca, and J. M. Barea. 1993. Inoculation with selected arbuscular mycorrhizal fungi and rhizobia to recover desertified Mediterranean ecosystems. *Applied and Environmental Microbiology* 59:129–133.

Lansac, A. R., A. Martín, and A. Roldán. 1995. Mycorrhizal colonization and drought interactions of Mediterranean shrubs under greenhouse conditions. *Arid Soil Research and Rehabilitation* 9:167–175.

Milton, S. J., W. R. J. Dean, M. A. du Pleiss, and W. R. Siegfried. 1994. A conceptual model of arid rangeland degradation. *Bioscience* 44:70–76.

Murphy, J., and J. P. Riley. 1962. A modified single solution method for the determination of phosphate in natural waters. *Analitica Chimica Acta* 27:31–36.

Newman, E. I., and P. Reddell. 1987. The distribution of mycorrhizas among families of vascular plants. *New Phytologist* 106:745–751.

Phillips, J. M., and D. S. Hayman. 1970. Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhiza fungi for rapid assessment of infection. *Transactions of the British Mycological Society* 55:159–161.

Roldán, A., and J. Albaladejo. 1993. Vesicular-arbuscular mycorrhiza (VAM) fungal populations in a xeric Torriorthent receiving urban refuse. *Soil Biology and Biochemistry* 25:451–456.

Roldán, A., F. García-Orenes. and A. Lax. 1994. An incubation experiment to determine factors involving aggregation changes in an arid soil receiving urban refuse. *Soil Biology and Biochemistry* 26:1699–1707.

Sánchez, J. A., F. Artes, and J. Albaladejo. 1980. Suelos silíceos de la zona Sur-Occidental de la Región de Murcia: Caracteres y aptitudes de utilización, pp. 77–84, in *Proceedings 3 Congreso Nacional de Química*. European Federation of Chemical Engineering Publication Series 12, Sevilla.

Schollemberger, C. J., and R. H. Simon. 1954. Determination of exchange capacity and exchangeable bases in soils. *Soil Science* 59:13–24.

Smith, S. E., and V. Gianinazzi-Pearson. 1988. Physiological interactions between symbionts in vesicular-arbuscular mycorrhizal plants. *Annual Reviews of Physiology, Plant Molecular Biology* 39:221–244.

Soil Survey Staff. 1975. *Soil taxonomy: A basic system of soil classification for making and interpreting soil surveys*. Agriculture Handbook 436. Soil Conservation Service, USDA, Washington, DC.

Thomas, G. W. 1967. Problems encountered in soil testing methods, pp. 37–54, in *Soil testing and plant analysis, Part I*. Soil Science Society of America. Madison, Wisconsin.

Warner, N. J., M. F. Allen, and J. A. MacMahon. 1987. Dispersal agents of vesicular-arbuscular mycorrhizal fungi in a disturbed arid ecosystem. *Mycologia* 79:721–730.

Yeomans, J. C., and J. M. Bremmer. 1989. A rapid and precise method for routine determination of organic carbon in soil. *Communications in Soil Science and Plant Analysis* 19:1467–1476.