Chapter

Effects of Arbuscular Mycorrhizal Fungi on Plant Adaptation to Arid Ecosystem of Bou-Hedma National Park in Tunisia

Mahmoudi Neji, Mahdhi Mosbah and Mars Mohamed

Abstract

Plants interact with beneficial microbes living in their rhizosphere, promoting their growth and development. In arid ecosystems, specific plant-associated microbes grant plants access to nutrients that would otherwise be inaccessible. Arbuscular mycorrhizal fungi (AMF) are probably one of the better known below-ground functional networks with plants. AMF plays a crucial role in plant performance and consequently in ecosystem functioning. AMF activities also determine the bio-availability of nutrients and therefore soil fertility. The main objective of the present study was to evaluate the plant-AMF interactions on soil functions under arid ecosystem in Tunisia. AMF colonization was evaluated by visual observation of AMF in fine roots of *Astragalus corrugatus* and *Lotus creticus* on Bou-Hedma National Park in Tunisia. Mycorrhizal colonization varied between plants, and the spore number was significantly different across rhizosphere soils. Statistical analysis showed a clearly positive correlation between the number of spores and plant-mycorrhizal intensity. For microbiological proprieties, our results showed that mycorrhizal plants improved significantly the different microbiological parameters. The results of the present study specified the association plant-AMF and highlight AMF importance as a tailored mechanism of plant adaptation to arid ecosystems.

Keywords: beneficial microbes, arid ecosystems, arbuscular mycorrhizal fungi (AMF), mycorrhizal colonization, microbiological proprieties

1. Introduction

Most arid and semiarid regions of Tunisia are frequently exposed to high temperatures and prolonged droughts, which reduce natural vegetation biodiversity and increase soil erosion and sand dune advance [1–3]. Under arid conditions, plant adaptation and survival depend obligatory on the plants' strategies to overcome drought and deficiencies on nutrients [4]. Low soil fertility in these types of ecosystems makes plants highly dependent on symbiotic association as plant's strategies. The developed symbioses, especially "mycorrhizal symbioses," have been noted for their importance in the stability of the ecosystem subject to degradation and desertification. The symbiotic association with mycorrhizal fungi plays a

significant role in the vegetative cover durability, especially in degraded soil of arid ecosystems [5].

Arbuscular mycorrhizal fungi (AMF) is a widespread group of soil fungi in natural ecosystems, forming a symbiotic association with roots of most of the terrestrial plants [6]. AMF play a significant role in the dynamic of ecosystem and growth and adaptation of plants to their environments [7]. AMF improve the ability of the plant to absorb water and nutrients from the soil, thus increasing their growth and development under unfavorable conditions [8]. AMF protect also their host plants against divers' biotic and abiotic stresses because of their beneficial roles in improving growth and drought tolerance [9].

AMF formed a symbiotic association between plant roots and the surrounding soils. Root systems play a crucial function in the acquisition of nutrients by exploiting a good volume of soil absorption [4]. When colonizing the roots of plant species, AMF developed two important structures: intra- and extra-radical mycelium. The intra-radical hyphae penetrate the root cells and develop some structures (developed vesicles and arbuscules) to improve the exchanges between the fungi and the plant host [10]. The extra-radical hyphae developed beyond the root surface and colonized the surrounding substratum. AMF significantly increased in the area of root absorption of plants colonized by the increase in the volume of soil explored [11]. So, AMF formation was exactly an adaptive strategy that provides the plant with an increased ability for nutrients in soils with low nutrient availability, particularly in arid and semiarid ecosystems [12].

AMF created an advantaged space for microbial development and activities. They play a crucial role in terrestrial ecosystems' functioning, especially in arid ecosystem, where root exudates are the major carbon source supporting soil microbial activities [13]. Apart from the nutritional benefits of the host plant, AMF increased the tolerance to drought [14], salt stress [15], and protection from root pathogens. As an important ecological function, AMF enhances soil structure and stability through the formation of hydro-stable aggregates [16, 17].

Generally, the symbiotic association AMF-plant communities are strongly dependent on the local environmental factors [18]. Soil physical, chemical, and biological properties are key factors to determine the AMF community and activity [19]. It has been reported that edaphic and climatic characteristics affect the diversity and composition of the AMF communities [20]. Some studies established a strong relationship between AMF colonization and environmental conditions such as temperature, rainfall, and physicochemical properties of soils [21, 22].

Therefore, in order to understand the effects of mycorrhization on plant development and their impact in the ecology of plant communities, it is important to assess the activities of the AMF community. Due to the relevant and important role these fungi play in the stability of the ecosystem, in this study we investigated the plant-AMF interactions on soil functions under arid ecosystem in Bou-Hedma National Park in Tunisia.

2. Materials and methods

2.1 Study site

The study site is the Bou-Hedma National Park (34°39′N and 94°8′E). This area is located in the Governorate of Sidi Bouzid, in central and southern Tunisia (**Figure 1**). The park was established in 1980 and covers an area of 16,488 hectares of which 6000 are under full protection. The mean annual rainfall varies between 150 and 250 mm. The mean annual temperature is about 17.2°C, while the minimal

Figure 1.
Location of the studied site: Bou-Hedma national park.

and maximal monthly temperature means are, respectively, 3.8°C (January) and 36.2°C (August).

2.2 Roots and soil samples

Fine roots of *Astragalus corrugatus* and *Lotus creticus* were collected at 25 cm depth. Four replicate samples were collected at each plant. Roots were washed with distilled water to remove attached soil for root colonization assessment.

Separate soil samples were collected from underneath each plant species. Control soil was recovered in an area which does not contain any vegetation. The soil samples were sieved to 2 mm to remove plant debris, gravel, and earthworms. They were then stored at 4°C for further analysis.

2.3 Assessment of root colonization by AMF

Root mycorrhizal colonization was checked for 90 root pieces per plant. Root segments of 1–2 cm were cleaned in 10% (w/v) potassium hydroxide (KOH) at 90° C for 1 h and acidified with 1% (w/v) hydrochloric acid (HCl) for 5 min. Then, the root segments were stained for 90 min at 60°C in 0.05% trypan blue (Phillips and Hayman, 1970). AMF colonization percentage was evaluated according to the grid interception of Giovannetti and Mosse [23]. Mycorrhizal frequency and intensity were calculated as described by Trouvelot et al. [24].

The parameters evaluated are:

- The mycorrhizal frequency (F %), which reflects the importance of the host plant root system infection by mycorrhizal fungi:

$$F\% = 100\ X\ (N - N0)/N \tag{1}$$

where N, number of the observed fragments; N0, number of non-mycorrhizal fragments

- The mycorrhizal intensity (M %), which is defined as the proportion of the root invaded by endomycorrhizal:

$$M\% = (95n5 + 70n4 + 30n3 + 5n2 + n1)/N. \tag{2}$$

where n = number of fragments assigned with the index 0, 1, 2, 3, 4, or 5.

2.4 Extraction and enumeration of AMF spores from soil samples

AMF spores were extracted from 100 g of each soil sample using the wet sieving method in sucrose gradient as described by Gerdemann and Nicolson [25]. 100 g of each studied soil sample were submerged in 1 L of tap water. After 1 min of stirring and 30 s of settling, the supernatant was sieved through three nested sieves with mashes of 1000, 100, and 32 μm. The filtrate was collected and sieved again (each soil suspension was sieved twice). Deposition in the sieves of 32 μm was recovered in centrifuge tubes of 25 ml. A viscosity gradient was created by adding 25 ml of aqueous sucrose solution at 60%. After 2 min of centrifugation at 2000 at 3000 rpm/min, the supernatant was poured onto the sieve of 32 μm; the fraction retained was rinsed with distilled water to remove sucrose. After extraction, spores were counted under a stereomicroscope (40X magnification), and the average number was calculated per 100 g of dry soil.

2.5 Soil analysis

For chemical analyses, soil pH and electrical conductivity (Ec) were determined by the saturated paw method in water by pH meter and conductivity meter, respectively. Total nitrogen (TN) was determined by the Kjeldahl method, and soil organic carbon (Corg) was determined by the Walkley and Black method. All soil analyses were performed in triplicate.

2.6 Soil microbial biomass

Microbial biomass carbon (Cmic) present in the different soil samples was determined by the method of "fumigation–extraction" as described by "Amato and Ladd [26]". This method is based on the use of ninhydrin-N reactive compounds extracted from soils with potassium chloride (KCl) after an incubation period (around a 10-day fumigation period). All soil analyses were performed in triplicate.

2.7 Biochemical properties

For enzyme activities, phosphatase and β-glucosidase activities were determined as described by Caravaca et al. [27]. The p-nitrophenol (PNP) formed in phosphatase activity and the p-nitrophenol glucopyranoside in β-glucosidase were determined in a spectrophotometer at 398 nm. The dehydrogenase activity was determined according to Garcia et al. [28]. The iodonitrotetrazolium formazan (INTF) formed was measured using a spectrophotometer at 490 nm. All soil analyses were performed in triplicate.

2.8 Statistical analyses

All the statistical analyses were performed by using the SAS statistical package. The effects of AMF on soil proprieties were subjected to ANOVA for repeated measures. The least significant difference values at 5% level of significance ($P \leq 0.05$) were calculated to assess differences between parameters.

3. Results

3.1 Physical and chemical proprieties of the studied site

The different physical and chemical proprieties of studied soil are shown in **Table 1**. Data showed that the pH was basic (pH > 7) and the electrical conductivity

Granulometry	Available phosphorus (ppm)	Available nitrogen (ppm)	pH	Electrical conductivity (s m^{-1})	Organic carbon %	Water content %
Sandy loam	6.0 ± 0.2	110 ± 10	7.8 ± 0.1	2.0 ± 0.1	1.2 ± 0.1	3.2 ± 0.2

Table 1.
Physical and chemical properties of the studied site.

was 2 sm $^{-1}$. The present studied the soil studied was of sandy loam texture. A low values of water content and available P were registered. The mean soil organic carbon and total available nitrogen (TN) were around 1.2% and 110 ppm, respectively.

3.2 AMF colonization of roots

Microscopic examinations of root fragments (**Figure 2**) demonstrated the development of different endogenous structures characterizing mycorrhizal fungi (intracellular hyphae, vesicles of varying shapes and arbuscules) in the root cells of the two studied plants.

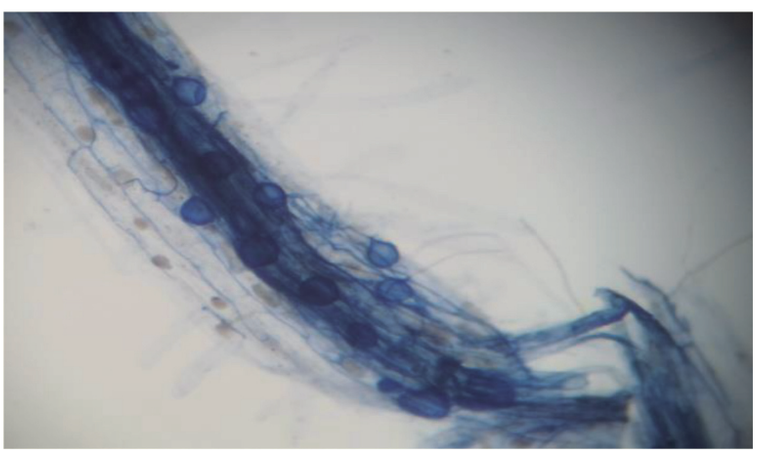

Figure 2.
Endogenous structures of AMF colonization (intraradical hyphae, and spores) within a root of A. corrugatus.

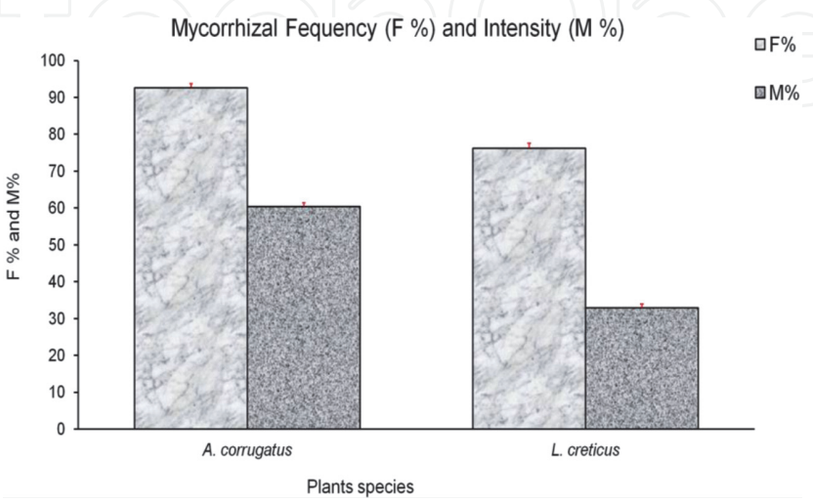

Figure 3.
Data from AMF colonization (mycorrhizal frequency and mycorrhizal intensity) of plant roots: A. corrugatus *And* L. creticus*. Error lines correspond to the standard deviation (n = 3). The significant differences calculated at P < 0.05.*

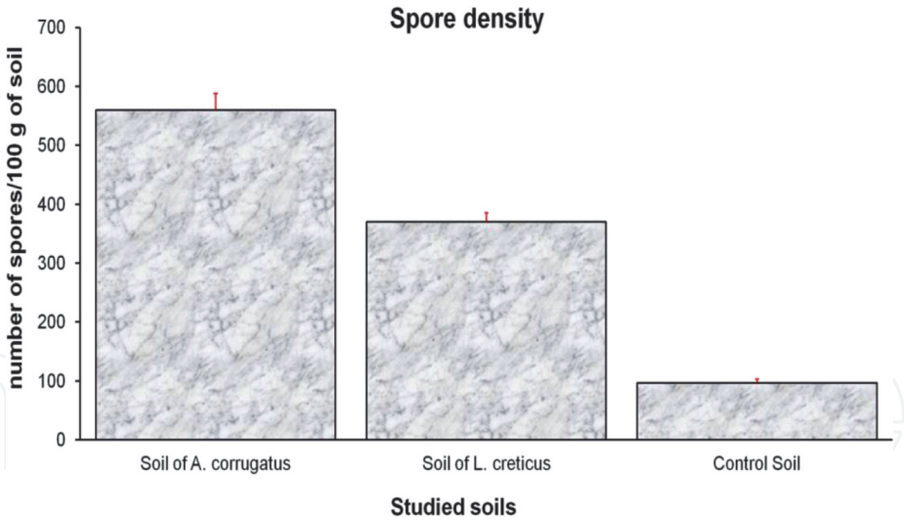

Figure 4.
Data from spores' density (number of spores) of studied soils. Error lines correspond to the standard deviation (n = 3). The significant differences calculated at P < 0.05.

Mycorrhizal frequency in the roots varied between the two studied Fabaceae plants, reaching 76.2% (*L. creticus*) and 92.6% (*A. corrugatus*) (**Figure 3**).

The mycorrhizal power that corresponds to the percentage of the mycorrhizal intensity of root cortex showed a significant difference (**Figure 3**). The roots of *A. corrugatus* have the highest mycorrhizal intensity (60.4%), while the roots of *L. creticus* have the lowest values (32.9%).

3.3 Abundance of spores' densities

AMF spores were detected in all studied soil samples (**Figure 4**). The majority number of AMF spores isolated from the rhizosphere varied significantly ($P < 0.05$) among the three studied soils; the average numbers ranged from 560 spores to 96 spores per 100 g dry soil. The highest value was recorded in the rhizosphere of *A. corrugatus* and the lowest in the control soil devoid of any plant cover (**Figure 4**).

Static analyses registered significant differences in the values of spore density (**Figure 4**) across the different studied samples, with an increasing trend in

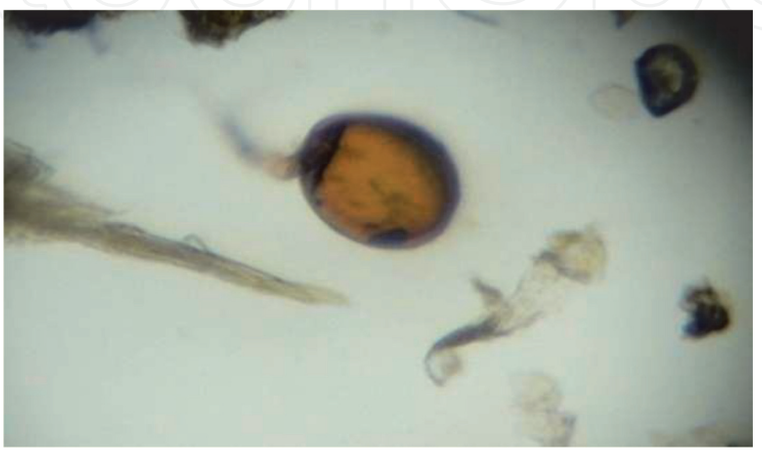

Figure 5.
Morphological appearance of some spores in a soil suspension of rhizospheric of A. corrugatus *(magnification: 100X).*

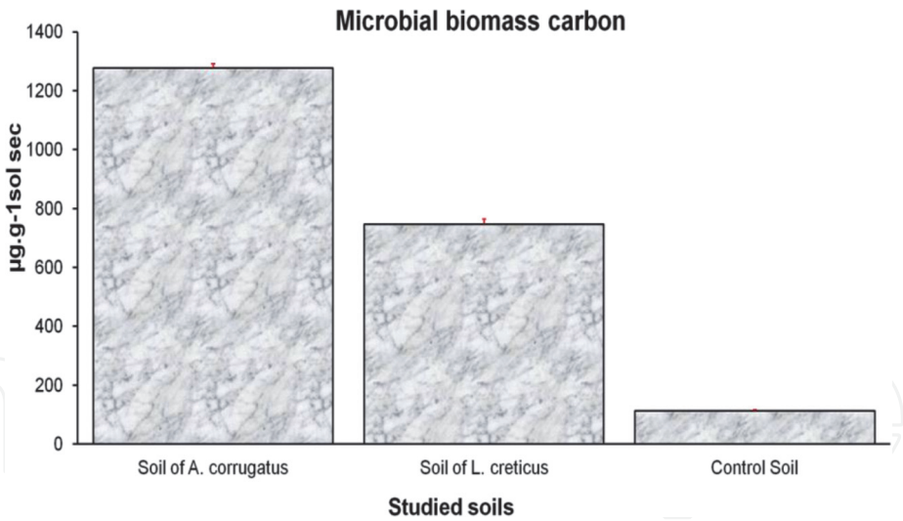

Figure 6.
Data from microbiological parameters (microbial biomass) of studied soils. Error lines correspond to the standard deviation (n = 3). The significant differences calculated at P < 0.05.

sporulation and with the increasing percentage of mycorrhizal colonization shown in the root samples of plant species.

A high variability in spore size, color, and shape was observed. The majorities of the spores were yellow-colored and did not exceed 70 µm in size (**Figure 5**).

3.4 Microbiological properties: microbial biomass

Microbial biomass varied significantly between the three studied soils (**Figure 6**). Rhizospheric soil of *A. corrugatus* presents the highest values (1277.22 µg C/g soil) compared with the control soil (112.31 µg C/g soil). The results showed a very significant effect of the mycorrhizal plant. However, across the different soil samples, **Figure 6** showed significant differences in the values of microbial biomass,

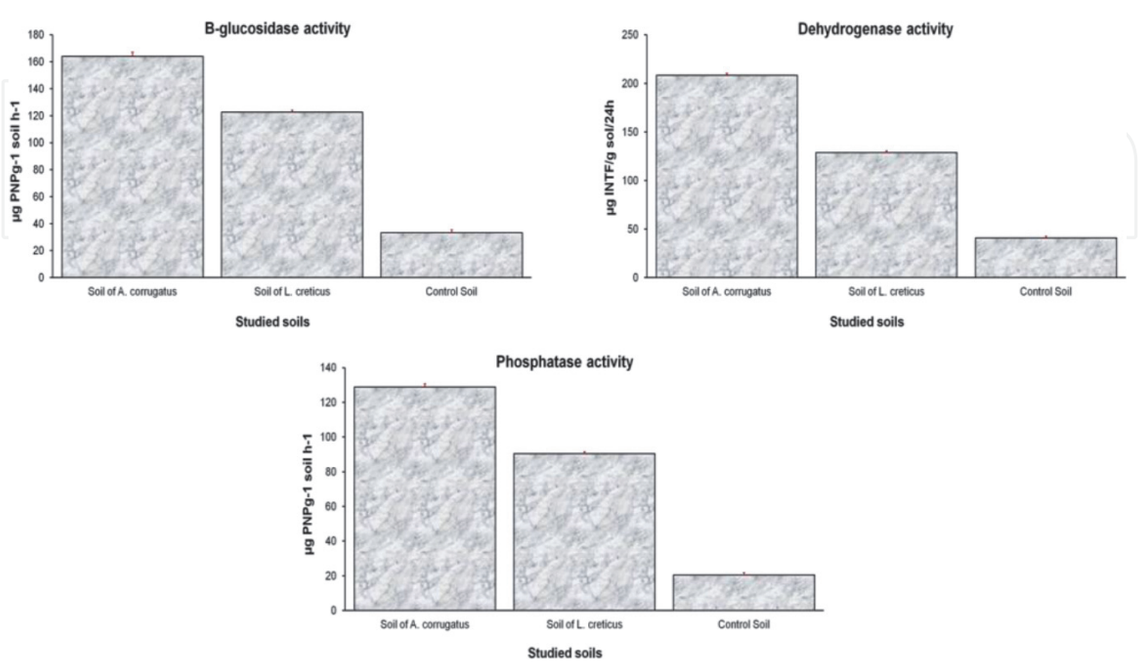

Figure 7.
Data from biochemical parameters (dehydrogenase, β-glucosidase and phosphatase activity) of studied soils. Error lines correspond to the standard deviation (n = 3). The significant differences calculated at P < 0.05.

with an increasing trend of microbial communities and with the increasing percentage of mycorrhizal colonization.

3.5 Biochemical properties: enzymatic activities

Across the different studied soils, static analyses indicated significant differences in the values of dehydrogenase activity (**Figure 7**), with an increasing trend in enzyme activity and with the increasing percentage of mycorrhizal colonization. Phosphatase and β-glucosidase activities showed the same trends as dehydrogenase activity, values were higher in the high mycorrhizal intensity, and they decreased along with soil devoid of any vegetation types (**Figure 7**).

4. Discussion

Arbuscular mycorrhizal fungi as symbiotic microorganisms were a major microbial component in the soil that is registered in all different climates and ecosystems [11]. AMF plays crucial roles in the restoration and retention of soil fertility, improving significantly the development of the host plant in degraded soils in the arid and semiarid region [5].

In general, the predominant mycorrhizal associations in natural ecosystems (such as our study area: Bou-Hedma National Park (**Figure 1**)) are arbuscular and vesicular mycorrhizal fungi [29]. The presence of different AMF structures such as vesicles and arbuscules (**Figure 2**) in the root cortex of the two studied plants indicates that these species had a higher mycotrophic status and the mycorrhizal activity can improve their successful growth in arid regions. Our result is consistent with the finding that more than 80% of terrestrial plants are mycotrophic plants [30]. The percentage of mycorrhizal colonization (**Figures 2** and **3**) varies between studied plants. However, many plant species have very different levels of dependence to AM fungi to satisfy their nutritional needs [31, 32]. All these can explain the results obtained in our study since *Astragalus corrugatus* showed the highest dependence on mycorrhizal fungi.

Our study revealed considerable variability in spore number. The spore densities detected in the different studied soil ranged from 96 to 560 spores per 100 g dry soil. The abundance of AMF spores in the different prospected soils suggests the high AMF diversity that can characterize our arid studied ecosystem.

Mycorrhizal plant species promote the development of different fungal propagules (spores) in their rhizospheric soil [33]. For this reason, as shown in **Figure 4**, the spore number registered in the rhizospheric soil of the two studied plants was higher than in the open areas (control soil).

As shown in the microscopic observation of spore suspensions (**Figure 5**), a high variability in spore size, color, and shape was observed, and the majority of the spores did not exceed 70 μm in size (a dominance of small spores). Thus, in general, the predominance of this type may be a strategy of selective adaptation to the major abiotic stress process and especially the drought stress [34].

We observed as indicated in **Figure 4** that spore densities in the plant rhizospheres are correlated significantly with the intensity colonization registered on their corresponding mycorrhizal root systems [35]. However, the highest mycorrhizal plant (*A. corrugatus*) registered the highest spore's number in their rhizospheric soil.

The plant-AMF symbiotic association is dependent on multiple factors: soil nutrient availability [nitrogen (N) and phosphorus (P)], environmental variables (temperatures), and soil properties (water, pH, salinity, and organic matter) [36].

In general, AMF play important roles for nutrient acquisition to the host plant [11]. Thus, the levels of mycorrhizal colonization are reduced by high value of P and N [20]. However, it is not the case in our area of study which is characterized by low soil nutrient availability (**Table 1**). Temperature, pH, and water availability also seem to affect the percentage of root AMF colonization. Thus, it has been reported that an important value of water content inhibits spore germination by increasing AMF mycelium growth [34]. As indicated in **Table 1**, our result showed that all environmental factors (low values) influence significantly AMF development on these types of ecosystems which explain the high level of AMF colonization and spore densities registered in the park.

It is well established that the rhizosphere is a very dynamic biological environment clearly distinct from the bulk soil (control soil), where microbial diversity and activity are characteristic. Various microbiological communities in the rhizospheric soil are essential for the occurrence of fundamental processes (biochemical cycling) that improve the formation and fertility of the soil and consequently the maintenance and sustainability of arid ecosystems [37]. The measurement of the status and activities of specific microbial communities contributing to the soil fundamental processes has the potential to provide particularly rapid and sensitive means of characterizing changes in soil quality and fertility [3].

Microbiological soil parameters are well known to be used as an important indicator of soil quality and dynamics. Biological parameters used are those engaged to characterize the size, structure, and dynamic of the microbial communities in the soil [38]. As a first biological indicator, soil microbial biomass carbon (Cmic) has been increasingly considered an environmental marker [39]. Our results (**Figure 6**) agree with those reports, which also showed the positive effect of the rhizospheric soil of mycorrhizal plant on Cmic compared to control soil devoid of vegetation. The important value of microbial density, estimated by the microbial carbon, was significantly registered in the soil recovered from two studied plants. The control soil (opened area) devoid of any vegetation has the lowest Cmic. This finding is in consistence with the reports, indicating at first that a soil-containing plant maintains a higher microbial biomass than soil devoid of vegetation cover [40] and also that plant species promote the development, the abundance, and the diversity of the microbial communities in their rhizosphere, which improve soil fertility and levels of organic matter in the soil [41, 42]. In general, plants can influence the microbial community by the secretion of different root exudates [43]. Our analyses showed that the high mycorrhizal plant registered the high densities on microbial communities.

However, similarly to roots, AMF are leaky and lose nutrients into the mycosphere, which will selectively promote the development of different microorganisms. In this context, it is to be expected that AMF play a crucial role in the biological characteristics of the rhizosphere [27, 44]. In this particular respect, the importance of the AMF in promoting the development of the microbial community is confirmed by the low levels of microbial carbon observed in the soil without plant cover (control).

It is notable that rhizosphere activity improves significantly plant fitness and soil quality because the microbial activity (as biochemical cycling) can help the host plant to better adapt to stress conditions concerning water and mineral deficit [45]. Soil enzymes take part in organic matter decomposition and the major nutrient cycling. They are important components for biochemical functioning of soils [3].

Soil enzyme activities have been used as good indicators of microbial activity. With regard to enzymes present in the different studied soils, dehydrogenase, phosphatase, and β-glucosidase are considered as a microbiological activity index of soil quality in semiarid and arid soils. The potential activities of the three studied enzymes

(**Figure** 7) were higher in the rhizosphere of the high mycorrhizal plants than in that of the bulk soil (soil devoid of vegetation cover). However, AMF increase the diversity of the carbon sources available to the microorganisms in the rhizosphere [46], which in part is due to their nutritional mode and excretion of catabolic enzymes to the surrounding medium. The importance of the soil microbial activity in association with enzyme activity was highlighted by the similarity in the activity patterns of the three studied enzymes, an indicator of microbial activity [47].

Furthermore, the major biological activity of soil was a highly sensitive indicator to biotic and abiotic environmental factors. It is a useful microbiological parameter of the soil improvement or degradation [48].

At this point it is clear that the impact of AMF on plant community composition and functioning was a key factor for ecosystem functioning and stability [49].

5. Conclusion

In conclusion, this study is among the firsts report on AMF communities associated with some herbaceous plant species in arid Tunisia and especially in protected ecosystem "Bou-Hedma National Park." Little is currently known about the AMF composition and activity and their impact on the stability of arid regions on Tunisia. *A. corrugatus* have the highest mycorrhizal colonization registered in their roots, and consequently they registered the highest spore density in their rhizospheric soil. Our study also reported that the soil microbial activity was improved with the high levels of AMF. Therefore, the influence of AMF on soil biochemical properties (enzymatic activities) and their dynamics is significant. The findings of the present study should be considered on appropriate soil management and the preservation of the protected ecosystem, especially in the arid region.

Acknowledgements

This work was supported by the Tunisian Ministry of High Education and Research Development. We are grateful to Bou-Hedma National Park for making the experimental site available.

Conflict of interest

We declare we do not have any conflict of interest.

DOI: http://dx.doi.org/10.5772/intechopen.92087

Author details

Mahmoudi Neji[1,2*], Mahdhi Mosbah[1,3] and Mars Mohamed[1]

1 Faculté des Sciences de Gabès, Unité de Recherche: Biodiversité et Valorisation des Bio-ressources en Zones Arides (BVBZA), Erriadh Zrig, Tunisie

2 Faculté des Sciences de Tunis, Université Tunis Al Manar, Tunisie

3 Center for Environmental Research and Studies, Jazan University, Kingdom of Saudi Arabia

*Address all correspondence to: neji.2012@yahoo.com

References

[1] Mahmoudi N, Cruz C, Mahdhi M, Mars M, Caeiro MF. Arbuscular mycorrhizal fungi in soil, roots and rhizosphere of Medicago truncatula: Diversity and heterogeneity under semi-arid conditions. PeerJ. 2019;**7**:e6401. DOI: 10.7717/peerj.6401

[2] Mosbah M, Philippe DL, Mohamed M. Molecular identification of arbuscular mycorrhizal fungal spores associated to the rhizosphere of Retama raetam in Tunisia. Soil Science & Plant Nutrition. 2018;**64**:335-341. DOI: 10.1080/ 00380768.2018.1431012

[3] Fterich A, Mahdhi M, Mars M. Impact of grazing on soil microbial communities along a chronosequence of Acacia tortilis subsp, raddiana in arid soils in Tunisia. European Journal of Soil Biology. 2012;**50**:56-63. DOI: 10.1016/j.ejsobi.2011.12.002

[4] Yao Q, Wang LR, Zhu HH, Chen JZ. Effect of arbuscular mycorrhizal fungal inoculation on root system architecture of trifoliate orange (*Poncirus trifoliata* L. Raf.) seedlings. Scientia Horticculturae. 2009;**121**:458-461. DOI: 10.1016/j.scienta.2009.03.013

[5] Duponnois R, Plenchette C, Prin Y, Ducosso M, Kisa M, Ba AM, et al. Use of mycorrhizal inoculation to improve reafforestation process with Australian Acacia in Saheliane cozones. Ecological Engineering. 2007;**29**:105-112. DOI: 10.1016/j.ecoleng.2006.09.008

[6] Gianinazzi S, Gollotte A, Binet MN, van Tuinen D, Redecker D, Wipf D. Agroecology: The key role of arbuscular mycorrhizas in ecosystem services. Mycorrhiza. 2010;**20**:519-530. DOI: 10.1007/s00572-010-0333-3

[7] Brundrett M. Mycorrhizal associations and other means of nutrition of vascular plants: Understanding the global diversity of host plants by resolving conflicting information and developing reliable means of diagnosis. Plant and Soil. 2009;**320**:37-77. DOI: 10.1007/s11104-008-9877-9

[8] Requena N, Perez-Solis E, Azcon-Aguilar C, Jeffries P, Barea JM. Management of indigenous plant microbe symbioses aids restoration of desertified ecosystems. Applied and Environmental Microbiology. 2001;**67**:495-498. DOI: 10.1128/aem.67.2.495-498.2001

[9] Yang HS, Zhang Q, Dai YJ, Liu Q, Tang JJ, Bian XM, et al. Effects of arbuscular mycorrhizal fungi on plant growth depend on root system: A meta-analysis. Plant and Soil. 2015;**389**:361-374. DOI: 10.1007/s11104-014-2370-8

[10] Van Aarle IM, Cavagnaro TR, Smith SE, Smith FA, Dickson S. Metabolic activity of *Glomus intraradices* in Arumand Paris-type arbuscular mycorrhizal colonization. The New Phytologist. 2005;**166**:611-618. DOI: 10.1111/j.1469-8137.2005.01340.x

[11] Smith SE, Read DJ. Mycorrhizal Symbiosis. San Diego: Academic Press; 2008. p. 787

[12] Allen MF. Mycorrhizal fungi: Highways for water and nutrients in arid soils. Vadose Zone Journal. 2007;**6**:291-297. DOI: 10.2136/vzj2006.0068

[13] Bago B, Vierheilig H, Piché Y, Ázcon-Aguilar C. Nitrate depletion and pH changes induced by the extraradical mycelium of the arbuscular mycorrhizal fungus Glomus intraradices grown in monoxenic culture. The New Phytologist. 1996;**133**:273-280. DOI: 10.1111/j.1469-8137.1996.tb01894.x

[14] Auge RM. Water relations, drought and vesiculararbuscular mycorrhizal symbiosis. Mycorrhiza. 2000;**11**:3-42. DOI: 10.1007/s005720100097

[15] Garg N, Chandel S. Effect of mycorrhizal inoculation on growth, nitrogen fixation, and nutrient uptake in *Cicer arietinum* (L.) under salt stress. Turkish Journal of Agriculture and Forestry. 2011;**35**:205-214. DOI: 10.3906/tar-0908-12

[16] Daynes CN, Field DJ, Saleeba JA, Cole MA, McGee PA. Development and stabilization of soil structure via interactions between organic matter, arbuscular mycorrhizal fungi and plant roots. Soil Biology and Biochemistry. 2013;**57**:683-694. DOI: 10.1016/j.soilbio.2012.09.020

[17] Peng S, Guo T, Liu G. The effects of arbuscular mycorrhizal hyphal networks on soil aggregations of purple soil in Southwest China. Soil Biology and Biochemistry. 2013;**57**:411-417. DOI: 10.1016/j.soilbio.2012.10.026

[18] Velázquez M, Stürmer SL, Bruzone C, Fontenla S, Barrera M, Cabello M. Occurrence of arbuscular mycorrhizal fungi in high altitude sites of the Patagonian Altoandina region in Nahuel Huapi National Park (Argentina). Acta Botânica Brasílica. 2016;**30**:521-531. DOI: 10.1590/0102-33062016abb0223

[19] Oehl F, Laczko E, Bogenrieder A, Karl Stahr K, Bösch R, Vander Heijden M, et al. Soil type and land use intensity determine the composition of arbuscular mycorrhizal fungal communities. Soil Biology and Biochemistry. 2010;**42**:724-738. DOI: 10.1016/j.soilbio.2010.01.006

[20] Johnson NC. Resource stoichiometry elucidates the structure and function of arbuscular mycorrhizas across scales. The New Phytologist. 2010;**185**:631-647. DOI: 10.1111/j.1469-8137.2009.03110.x

[21] Xu X, Chen C, Zhang Z, Sun Z. The influence of environmental factors on communities of arbuscular mycorrhizal fungi associated with *Chenopodium ambrosioides* revealed by MiSeq sequencing investigation. Scientific Reports. 2017;**7**:45134. DOI: 10.1038/srep45134

[22] Frater PN, Borer ET, Fay PA, Jin V. Nutrients and environment influence arbuscular mycorrhizal colonization both independently and interactively in *Schizachyrium scoparium*. Plant and Soil. 2018;**425**:493-506. DOI: 10.1007/s11104-018-3597-6

[23] Giovannetti M, Mosse B. An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. The New Phytologist. 1980;**84**:489-500. DOI: 10.1111/j.1469-8137.1980.tb04556.x

[24] Trouvelot A, Kough JL, Gianinazzi-Pearson V. Evaluation of VA infection levels in root systems. Research for estimation methods having a functional significance. In: Gianinazzi-Pearson V, Gianinazzi S, editors. Physiological and Genetical Aspect of Mycorrhiza. Paris: INRA Press; 1986. p. 1986

[25] Gerdemann W, Nicolson TH. Spores of mycorrhizal endogone species extracted by wet sieving and decanting. Transactions of the British Mycological Society. 1963;**46**:235-244. DOI: 10.1016/s0007-1536(63)80079-0

[26] Amato M, Ladd JN. Assay for microbial biomass based on ninhydrinreactive nitrogen in extracts of fumigated soils. Soil Biology and Biochemistry. 1988;**20**:107e114. DOI: 10.1016/0038-0717(88)90134-4

[27] Caravaca F, Alguacil MM, Torres P, Roldán A. Plant type mediates rhizospheric microbial activities and soil aggregation in a semiarid Mediterranean salt marsh. Geoderma. 2005;**12**:375e382. DOI: 10.1016/j.geoderma.2004.05.010

[28] Garcia C, Hernandez T, Costa F. Potential use of dehydrogenase activity

as an index of microbial activity in degraded soils. Communications in Soil Science and Plant Analysis. 1997;**28**: 123e134. DOI: 10.1080/0010362 9709369777

[29] Neree AO. Diversity and Dynamics of Mycorrhizal Associations in Tropical Rain Forests with Different Disturbance Regimes in South Cameroon. Vol. 2000. Netherlands: Wageningen University; 2000. p. 167

[30] Wang B, Qiu YL. Phylogenetic distribution and evolution of mycorrhizas in land plants. Mycorrhiza. 2006;**16**:299-363. DOI: 10.1007/ s00572-005-0033-6

[31] Bagayoko M, Buerker A, Lung G, Bationo A, Romheld V. Cereal/legume rotation effects on cereal growth in Soudano-Sahelian West Africa: Soil mineral nitrogen, mycorrhizae and nematodes. Plant and Soil. 2000; **218**:103-116. DOI: 10.1023/a: 1014957605852

[32] Barea JM, El-Atrach F, Azcon R. The role of VA mycorrhizas in improving plant N acquisition from soil as assessed with 15N. In: Flitton C, editor. The Use of Stable Isotopes in Plant Nutrition, Soil Fertility and Environmental Studies. Vienna: Joint IAEA, FAO, Division; 1991. pp. 677-808

[33] Lovelock CE, Andersen K, Morton JB. 2003: Arbuscular mycorrhizal communities in tropical forests are affected by host tree species and environment. Oecologia. 2003;**135**: 268-297. DOI: 10.1007/s00442-002-1166-3

[34] Boddington C, Dodd J. The effect of agricultural practices on the development of indigenous arbuscular mycorrhizal fungi. I. Field studies in an Indonesian ultisol. Plant and Soil. 2000; **218**:137-144. DOI: 10.1023/a:1014 966801446

[35] Houngnandan P, Yemadje RG, Kane A, Boeckx P, Van Cleemput O. Les glomales indigènes de la forêt claire à Isoberliniadoka (Craib et Stapf) à Wari-Maro au centre du Bénin. Tropicultura. 2009;**27**(2):83-87

[36] Liu Y, Shi G, Mao L, Cheng G, Jiang S. Direct and indirect influences of 8 yr of nitrogen and phosphorus fertilization on Glomeromycota in an alpine meadow ecosystem. The New Phytologist. 2012;**194**:523-535. DOI: 10.1111/j.1469-8137.2012.04050

[37] Barea JM, Pozo MJ, Azcón R, Azcón-Aguilar C. Microbial co-operation in the rhizosphere. Journal of Experimental Botany. 2005;**56**:1761-1778

[38] Schloter M, Dilly O, Munch JC. Indicators for evaluating soil quality. Agriculture, Ecosystems and Environment. 2003;**98**:255-262. DOI: 10.1016/s0167-8809(03)00085-9

[39] Franco I, Contin M, Bragato G, De Nobili M. Microbiological resilience of soils contaminated with crude oil. Geoderma. 2004;**121**:17-30. DOI: 10.1016/j.geoderma.2003.10.002

[40] Bastida F, Moreno JL, Hernandez T, Garcia C. Microbiological degradation index of soils in a semiarid climate. Soil Biology and Biochemistry. 2006;**38**: 3463-3473. DOI: 10.1016/j.soilbio.2006. 06.001

[41] Cao CY, Jiang DM, Teng XH, Jiang Y, Liang WJ, Cui B. Soil chemical and microbiological properties along a chronosequence of Caraganamicrophylla lam. Plantations in the Horqin sandy land of Northeast China. Applied Soil Ecology. 2008;**40**:78-85. DOI: 10.1016/j. apsoil.2008.03.008

[42] Sileshi G, Akinnifesi FK, Ajayi OC, Place F. Meta-analysis of maize yield response to planted fallow and green manure legumes in sub-Saharan

African. Plant and Soil. 2008;**307**:1-19. DOI: 10.1007/s11104-008-9547

[43] Dennis PG, Miller AJ, Hirsch PR. Are root exudates more important than other sources of rhizodeposits in structuring rhizosphere bacterial communities? Microbial Ecology. 2010; **72**:313-327. DOI: 10.1111/ j.1574-6941.2010.00860.x

[44] Cruz RE, Garcia MU. Nitrogen fixation and mycorrhizae in acacias on degraded grasslands. In: Kamiset A, Taylor DA, editors. Tropical Acacias in East Asia and the Pacific. Bangkok: Winrock Intenational Institute for Agriculture Research; 1992. pp. 59-71

[45] Barea JM, Azcon R, Azcon-Aguilar C. Mycorrhizosphere interactions to improve plant fitness and soil quality. Antonie van Leeuwenhoek International Journal of General and Molecular Microbiology. 2002;**81**(1–4):343-351. DOI: 10.1023/a:1020588701325

[46] Rillig MC. Arbuscular mycorrhizae and terrestrial ecosystem processes. Ecology Letters. 2004;**7**:740-754. DOI: 10.1111/j.1461-0248.2004.00620.x

[47] Nannipieri P, Kandeler E, Ruggiero P. Enzyme activities and microbiological and biochemical processes in soil. In: Burns RG, Dick RP, editors. Enzymes in the Environment. Activity, Ecology and Applications. New York: Marcel Dekker; 2002. pp. 1-33

[48] Gil-Sotres F, Trasar-Cepeda C, Leiros MC, Seoane S. Different approaches to evaluating soil quality using biochemical properties. Soil Biology and Biochemistry. 2005;**37**: 877-887. DOI: 10.1016/j.soilbio.2004. 10.003

[49] Martínez-García L, Miranda J, Pugnaire FI. 2011. Impacts of changing rainfall patterns on mycorrhizal status of a shrub from arid environments. European Journal of Soil Biology. 2011; **50**:64-67. DOI: 10.1016/j.ejsobi.2011. 12.005