

Mycorrhizal status of main spontaneous or introduced forest trees in El Tarf province (Algerian North-east)

Arifa Beddiar¹, Mériem Adouane², Imène Boudiaf³ and Adem Fraga⁴

^{1,2,3,4}Laboratory of Plant Biology and Environment, Department of Biology, Badji Mokhtar University, PB12, 23000 Annaba, Algeria

fragbed@yahoo.fr

Abstract: In Algeria, often, serious imbalances of the forest ecosystems caused by the badly led exploitations, the repeated fires, the overgrazing involve the disappearance of the timbered state on vast territories. Within the framework of the plantation and restoration programs of these degraded forests, we plan to contribute to it by tests of controlled mycorrhization of forest trees in nursery. But to realize these tests, it is necessary to know beforehand the natural symbiotic status of the studied species.

We present there, the first stage of our work which consisted in establishing the mycorrhizal status of main spontaneous or introduced forest species into Algerian North-east. The study was carried out in El Tarf province and related to all the species of pine, oaks and other leafy trees existing in this area.

The results show that all the species observed have either ectomycorrhizas (ECM) or arbuscular endomycorrhizas (AM) or both at the same time. In addition, the introduced species of the *Acacia* or *Casuarina* genus carry AM and also nitrogen fixing nodules. It should be noted the remarkable diversity of ectomycorrhizal morphotypes associated to the pines and to the oaks and also that of the endomycorrhizal fungi spores occur in the rhizosphere of other species.

Lastly, it is significant to note the omnipresence of *Cenococcum geophilum* mycorrhiza in all the stations and the majority of the ectomycorrhizal trees.

Keywords: ectomycorrhizas, arbuscular mycorrhizas, forest species, Algerian North-east

Introduction

In the Algerian North-East, the forest ecosystems play a socio-economic, ecological and entertaining role of first order (Messaoudène *et al.*, 1996). However, each year, the surface of these forests is reduced seriously because of many factors such as the long dry season, overgrazing, fires and overexploitation. Today, the reconstitution of these forests proves to be of need consequently to increase the possibilities of trees to fight against the external agents proves more than necessary. In addition, it was established that the majority of the forest trees contract symbiotic associations with some fungi and bacteria of the soil which gives opportunity to the tree to resist the drastic effects of the climate, the lack of nutrients provided from the soil and the parasitic attacks (Dommergues *et al.*, 1999; Smith and Read, 2008).

In this way, we foresee experiments of controlled mycorrhization in nurseries. But before, we think that the knowledge of the symbiotic status of the various species is an essential step. It is the subject of this present article. Our study proceeded in the El Tarf province (the extreme algerian North-east) and related to the trees or the shrubs of the forest ecosystems of plain, dune and mountain as well as the ripicolous ecosystems.

Materials and methods

-Presentation of the study zone and stations

The study zone is the area of Algeria limited by the Mediterranean Sea to north, the hills of the Tellian Atlas in the south and the west and by the Tunisian border in the east (Fig.1). It is characterized by an average rain fall of 910 mm/year and an average maximum temperature around 18°C. The coldest months are January and February whereas July and August are hottest. The dry season lasts nearly 4 months and the atmospheric humidity goes between 69 and 74%. According to the Emberger climagramm (1955), the area of El Tarf extends from the sub wet to wet bioclimatic stage of vegetation (de Belair, 1990) and stands on deposits and alluvia, numidians siliceous flyshs and dunes. In this area, altitude varies sea level up to 1202m.

The geological constitution of the basement gather only nummulitic grounds dating primarily from the tertiary sector and quaternary (Joleau, 1936).The most advanced soils are under the oaks and the pines, they are forest

brown soils. The hills are characterized by sandstones and Numidian clays, the depressions by alluvia and colluviums (Marre, 1992). The localization and the description of the study stations are presented in figure1 and table 1.



Fig. 1: Localization of sampling stations (scale : 1 /1000000)

Table 1: Localization and characteristics of the various sampling stations

Stations	Brief description of the station
S1	Maquis of kermes oak on littoral dune. Ben Mhidi county, 2 km after oued Mafragh, altitude: 20m
S2	Plantation of maritime pine, Righia, Berrihane county, alt.: 50m
S3	Forest of cork oak and ripisylvaie, Righia, Berrihane county, 18 km after the Righia village, in direction of El-Kala, alt.: 50m
S4	Forest of cork oak. El-Koursi county, in the West of the Mellah lake, alt.: 100m
S5	Degraded forest of cork oak. El Kala county, natural reserve of Braptia, full southern with Mellah lake, alt.: 87m
S6	Plantation of Eucalyptus and Acacia. El Kala county, alt.: 50m
S7	Mix introduced species Souarak county, arboretum of Tonga, alder grove, alt.: 50m
S8	Mixed forest of cork oak-zeen oak. Bougous county, 500m after the Bougous village, alt.: 260 m
S9	Forest of zeen oak, jebel Ghorra, Bougous county, 2 km after the Bougous village towards the Tunisian border, alt.: 850 m

-Estimate of the mycorrhizal colonization

For each station, we took root samples of ten individuals of each tree species present in the settlement. Part of the fine roots was observed directly with the dissecting microscope in order to seek the ectomycorrhizas (ECM), which made it possible to distinguish different morphotypes according Agerer (1987-1996) description. On the other part, the frequency of the arbuscular mycorrhizal (AM) colonization was evaluated by the method of Trouvelot *et al.* (1986) after staining with the acid fuschine or Chlorazol black according to the technique of Kormanik *et al* (1980). Lastly, the *Rhizobium* or *Frankia* infection was detected by the presence of nodules.

- Extraction of Glomalean fungi spores

This research was carried out on soil samples taken of from *Acacia decurrens* rhizosphere in mixture with *Eucalyptus camaldulensis* in the El kala station (S6). The arbuscular mycorrhizal fungi (AMF) spores or sporocarps were extracted according to the wet sieving technique (Gerdeman, 1963). Their identification was tried by means of Schenck & Perez (1986) key.

Results and discussion

The results show the existence of mutualist symbiosis in all the prospected species. Whatever were the species either main one (cork oak, zeen oak and maritime pine), the secondary species (alder, chestnut, merisier, laurel...) or the introduced species (eucalyptus, acacia, cypress...), they carry all either ECM or AM or both at the same time. We thus share the suggestion formulated by Trappe (1977) and then by Selosse and Le Tacon (1999) according to which the mycorrhizal state is a general state in the plant world.

Among the observed ectomycorrhizal morphotypes, *Cenococcum geophilum* mycorrhiza (Fig. 2), easily recognizable, is in the majority and omnipresent as well at the oaks and the pines or other trees (chestnut, eucalyptus). *Cenococcum geophilum* is a fungus which has a great capacity to infect trees of different species and under varied ecological conditions (Le Tacon, 1997). When the water stress established (in summer), much of mycorrhizas is destroyed, those of *Cenococcum geophilum* are however more resistant (Garbaye & Guehl, 1997).

Its omnipresence at the majority of the studied forest species reveals difficult environmental conditions. Indeed, the Mediterranean forest constitutes an ecosystem submitted to strong edaphic, climatic and economic constraints which stimulates *Cenococcum geophilum* to be proliferating in this medium type.

According to Selosse and Le Tacon (1999), this species which is met in all the planet natural ecosystems with ectomycorrhizas, would contribute probably universally, to the survival of the forest trees with ectomycorrhizas during the water stress periods. Secondary species of the oak forest, spontaneous ones (wild cherry tree, bay-tree, alder, poplar...) or are introduced (hazel-tree, eucalyptus...) present an AM frequency infection ranging between 25 and 60% in the majority of the cases.

Acacia and *Casuarina*, although introduced species in Algeria present an additional infection respectively by *Rhizobium* and *Frankia*. The research of the Glomalean spores in the rhizosphere of *Acacia mearnsii* and *Eucalyptus camaldulensis* revealed a very significant morphotypic diversity (Fig. 3). The *Glomus* genus seems prevalent.

Table 2: Mycorrhizal status of main studied forest species (ECM: ectomycorrhiza, AM: arbuscular mycorrhiza, R: *Rhizobium* nodules, Fr.: *Frankia* actinorhizas)

Family	Plant Species and sampling stations (S)	Type of symbiosis and % of colonisation	Number of ECM morphotype
Pinaceae	<i>Pinus maritima</i> Lam S2	ECM (82,12)	10
	<i>Pinus pinea</i> L., S8	ECM (64,22)	4
	<i>Pinus radiata</i> L., S8	ECM (33,35)	2
Cupressaceae	<i>Juniperus oxycedrus</i> L., S6	MA (19,22)	-
	<i>Cupressus sempervirens</i> L., S6	ECM (58,64), MA (29,2)	3
Taxodiaceae	<i>Taxodium disticum</i> L., S 8	MA (26,22)	-
Casuarinaceae	<i>Casuarina equisetifolia</i> Forst., S1	MA (58,02), F	-
	<i>C. cunninghamiana</i> Miq.	MA (50,66), F	-
	<i>C. obesa</i> Miq.	MA (44,35), F	-
Fagaceae	<i>Quercus suber</i> L., S3, S4, S5, S6.	ECM (68,71), MA (09,00)	14
	<i>Quercus faginea</i> L., S4, S7,	ECM (62,32)	9
	<i>Quercus coccifera</i> L., S1	ECM (49,13)	2
	<i>Castanea sativa</i> L., S7	ECM (50,42), MA (41,15)	7
Betulaceae	<i>Alnus glutinosa</i> L.Gaertn, S2, S8	ECM (61,43), MA (25,52)	8
Salicaceae	<i>Populus alba</i> L., S5	ECM (28,40), MA (31,86)	4
	<i>Salix pedicellata</i> L., S5	MA (25,03)	-
	<i>Salix babylonica</i> L, S2, S8	MA (23,62)	-
Corylaceae	<i>Corylus avellana</i> L., S7	MA (29,24)	8
Juglandaceae	<i>Juglans regia</i> L., S8	MA (33,15)	-
Rosaceae	<i>Cerasus avium</i> L., S7	MA (44,09)	-
	<i>Crataegus monogyna</i> Jacq., S5	MA (38,64)	-
	<i>Rubus ulmifolius</i> L., S5,	MA (58,37)	-
Lauraceae	<i>Laurus nobilis</i> L., S5	MA(63,05)	-
Oleaceae	<i>Fraxinus oxyphylla</i> L., S8	MA (40,42)	-
	<i>Olea europaea</i> L., S1, S5	MA (51,56)	-
	<i>Olea oleaster</i> Link., S1, S5	MA (44,32)	-
	<i>Phillyrea media</i> L., S1, S5, ,S4	MA (65,62)	-
Anacardiaceae	<i>Pistacia lentiscus</i> L., S1, S5	MA (35,39)	-
Myrtaceae	<i>Myrtus communis</i> L., S3, S5	MA (63,54)	-
	<i>Eucalyptus globulus</i> Labill., S6, S8	ECM (36,80), MA (12,96)	3
	<i>Eucalyptus camaldulensis</i> Dehnh, S6	ECM (30,72), MA (15,06)	3
Fabaceae	<i>Ceratonia siliqua</i> L., S7	MA(35,44)	-
	<i>Calycotome villosa</i> Link. S5,	MA (60,00)	-
	<i>Genista ferox</i> L., S6	MA (30,39)	-
	<i>Genista numidica</i> L., S6	MA (29,35)	-
	<i>Cytisus triflorus</i> L'Herit., S6	MA (29,12)	-
	<i>Acacia cyanophylla</i> Benth., S8	MA (68,20), R	-
	<i>Acacia mearnsii</i> De Wild., S8	MA (72,75), R	-
	<i>Acacia melanoxylon</i> R., S8	MA (61,23), R	-
	<i>Acacia retinoides</i> Willd., S8	MA (42,26), R	-
	<i>Retama monosperma subsp bovei</i> Spach (Maire) S1	MA (08,77), R	-
Cistaceae	<i>Cistus monspelliensis</i> L.S2,S3,S5	ECM (09, 75), MA (18,77)	1
	<i>Halimium halimifolium</i> (L) Wilk	MA (42,42)	-

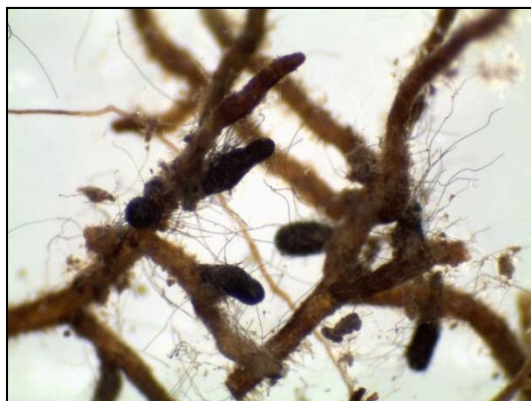


Figure 2: *Cenococcum geophilum* mycorrhiza colonizing the roots of *Quercus suber* (photo Adouane)

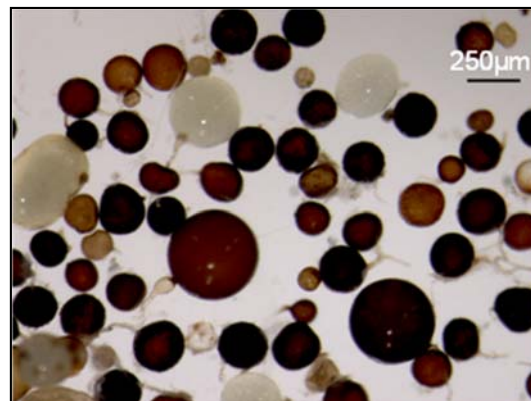


Figure 3: Various morphotypes of spores in the *Acacia mearnsii* rhizosphere (photo Meddad)

Conclusion

The results of this study allow us to give the following conclusions:

All the prospected trees present either an ECM or an AM colonization or both. Alder and filao carry actinorhizas, all Fabaceae are nodulated.

It is advisable to reinforce this study by identifying and by isolating the micro-organisms implicated in various symbiosis observed. These symbiotic microorganisms play certainly a significant role in the trees nutrition. But, do they act in an optimal way?

To make them profitable, intensifying the programs of biodiversity inventory of the whole knowledge is a requisite to any reliable biological or applied research, in order to select the most powerful strains by comparing them with known introduced strains. In this manner, the most performed strains will be selected. The exploitation of *Cenococcum geophilum* fungus would be also a significant way to explore.

To conclude, let us say simply that Algeria suffers from two flails, desertification and deforestation which dangerously accelerated after climatic changes and also under the effect of the intensive methods of exploitation and irrigation. Among the likely approaches susceptible to act against these two flails, one of most promising is that of to master the controlled mycorrhization in order to produce seedlings of good quality and to realize the successful afforestations.

References

- Agerer, R. (1987-1996). *Colour Atlas of ectomycorrhizae*. Einhorn-Verlag Eduard Dietenberger, Schwäbisch Gmünd.
- Bä, A., Duponnois, R., Diabaté, M. & Dreyfus, B. (2011). *Les champignons ectomycorhiziens des arbres forestiers en Afrique de l'ouest*. IRD Editions.
- Belair de, G. (1990). *Structure, fonctionnement et perspectives de gestion de quatre complexes lacustres et marécageux (El-Kala Est algérien)*. PhD Thesis U.S.T.I Montpellier University.
- Daniels, B. A. & Skipper, H. D. (1982). Methods for the recovery and quantitative estimation of propagules from soil. In: *Methods and principles of mycorrhizal research*. Schenck, .C. (ed). Amer. Phyt. Soc., (pp 29-35).
- Dommergues, Y. Duhoux, E. & Diem, H. G. (1999). *Les arbres fixateurs d'azote. Caractéristiques fondamentales et rôle dans l'aménagement des écosystèmes méditerranéens et tropicaux*. CIRAD, Espace, FAO et IRD (eds).
- Emberger, L. (1955). Une classification biogéographique des climats. *Rec. Trav. Lab. Océol. Zool., Fac. Sci. Montpellier Ser. Bot.*, 7, (pp 1-43).
- Garbaye, J. & Guehl, J. M. (1997). Le rôle des ectomycorhizes dans l'utilisation de l'eau par les arbres forestiers. *Revue Forestière Française*, n° spécial, (pp 106-120).

- Gerdemann, J. W. (1963). Spores of mycorrhizal endogone species extracted from soil by wet sieving and decanting. *Trans-Br-Mycol Soc* 46, (pp 234-235).
- Joleau, D. L. (1936). *Etude géologique de la région de Bône et de la Calle. Bull. Serv. Carte. Géol. De l'Algérie.* 2° série stratigraphique. Description régionale n° 2.
- Kormanik, P. P., Bryan, W. C. & Schultz, R. C. (1980). Procedures and equipment for staining large numbers of plant root samples for endomycorrhizal assay. *Can. J. Microbiol.* 26, pp 536-538.
- Le Tacon, F. (1997). Vers une meilleure prise en compte des champignons mycorrhiziens dans la gestion forestière. *Revue Forestière Française, n° spécial*, (pp 245- 255).
- Marre, A. (1992). *Le tell oriental algérien de Collo à la frontière tunisienne.* Etude géomorphologique. O.P.U. Alger, (pp 413-624).
- Messaoudène, M., Metna, B. & Djouaher, N. (1996). La régénération de *Quercus suber* (L) dans la forêt domaniale de Beni-gobri (Algérie). *Actes du séminaire méditerranéen sur la régénération des forêts de Chêne-liège.* Tabarka, Du 22 au 24 Oct. 1996. Annales de l'NRF.
- Schenck, N.C. & Perez, Y. (1986). *Manual of the identification VA mycorrhizal fungi.* INVAM
- Selosse, M. A. & Le Tacon, F. (1999). Les champignons dopent la forêt. *La recherche* 319, (pp 33-35).
- Trappe, J. M. (1977). Biogeography of hypogeous fungi: trees, mammals and continental drift. Abstr. 2nd Int. Mycol. Congr. Inc., Bigelow H E, Simmons E G (eds), Tampa, USA.
- Trouvelot, A. Kough, J. L. & Gianinazzi-Pearson, V. (1986). Mesure du taux de mycorrhization VA d'un système racinaire. Recherche d'une méthode d'estimation ayant une signification fonctionnelle. In: *Physiological and genetic aspects of mycorrhizal symbioses* (eds. V. Gianinazzi-Pearson et S. Gianinazzi): 217-221. INRA, Paris.