

Andrea Maremmani · Stefano Bedini  
Ivica Matošević · Paolo E. Tomei  
Manuela Giovannetti

## Type of mycorrhizal associations in two coastal nature reserves of the Mediterranean basin

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**Abstract** We analysed the mycorrhizal types of two coastal Mediterranean plant communities. Plants belonging to 82 species of 46 families growing in two Mediterranean study sites located within a Regional Natural Park in Italy (Macchia Lucchese) and in a National Park in Croatia (Brijuni) were assessed for the type of their mycorrhizal associations. Mycorrhizas occurred in 83% and 90% of the plant species surveyed in Macchia Lucchese and in Brijuni, respectively. On the basis of macroscopic and microscopic characteristics, six different mycorrhizal types were found, and their co-occurrence in the same plant community showed the large mycorrhizal diversity in Mediterranean vegetation. Different occurrence patterns of mycorrhizal types have been recognised in diverse habitats within Macchia Lucchese, from sand dune plant communities to sclerophyllous woodland. Mycorrhizal type abundance calculated on a floristic basis was very different from that obtained using a vegetation cover index.

**Keywords** Mediterranean ecosystem · Plant associations · Mycorrhizal diversity · Sand dunes · Natural reserves

A. Maremmani · S. Bedini · M. Giovannetti (✉)  
Dipartimento di Chimica e Biotecnologie Agrarie,  
Università di Pisa, Via del Borghetto 80, 56124 Pisa, Italy  
e-mail: mgiova@agr.unipi.it  
Tel.: +39-50-571561, Fax: +39-50-571562

I. Matošević  
Dipartimento di Produzione Vegetale e Tecnologie Agrarie,  
Università di Udine, Via delle Scienze 208, 33100 Udine, Italy

P.E. Tomei  
Dipartimento di Agronomia e Gestione dell'agro-ecosistema,  
Università di Pisa, Via S. Michele degli Scalzi 2, 56124 Pisa, Italy

M. Giovannetti  
Istituto di Biologia e Biotecnologia Agraria, CNR,  
Sezione di Pisa, Via del Borghetto 80, 56124 Pisa, Italy

### Introduction

Studies on the distribution of mycorrhizal associations in nature have shown close relationships between major terrestrial biomes and predominant mycorrhizal types, and distinctive patterns of mycorrhizal symbiosis have been identified (Brundrett 1991; Read 1991). Moreover, mycorrhizal diversity is considered an important factor in the establishment, survival and maintenance of plant community diversity (Grime et al. 1987; Brundrett and Kendrick 1988; Read 1998; Van der Heijden et al. 1998). Despite the important role played by mycorrhizal diversity in natural plant communities, there is little information on the distribution and abundance of the different mycorrhizal associations in European Mediterranean areas (Lamont 1982; Puppi and Tartaglioni 1991; Read 1991; Lansac et al. 1995; Requena et al. 1996).

This study was carried out to detect the different types and occurrence patterns of mycorrhizal associations in their natural habitats in two plant ecosystems within the Mediterranean basin, chosen for their location in coastal nature reserves.

### Materials and methods

#### Description of the study sites

The study sites were: Macchia Lucchese, located in the Migliarino-San Rossore-Massaciuccoli Regional Natural Park, on the north-western coast of Tuscany, near Pisa (Italy) (approximately 43° 49' N, 10° 17' W) and Veli Brijuni, the central and largest island of the Brijuni archipelago (approximately 44° 55' N, 13° 43' E), an undisturbed site within the National Park of Brijuni (Croatia), near the Western coast of the Istria Peninsula. Soil characteristics, which are reported in Table 1, varied distinctly between the two sites, whereas they were relatively constant across each site.

In Macchia Lucchese, the average annual air temperature is 14°C and the total annual precipitation is 1,021 mm. The site consists of a succession of dunes and backdune depressions backed by a woodland composed of sclerophyllous and wetland forest communities. The dune vegetation is well preserved and includes many grass species such as *Ammophila arenaria* (L.) Link, *Cakile*

**Table 1** Chemical and physical soil characteristics in Macchia Lucchese, Tuscany, Italy and in Brijuni National Park, Croatia

Soil	Sand %	Silt %	Clay %	pH (H <sub>2</sub> O)	Organic carbon % (automatic 900C ELTRA)	Available phosphorus ppm (Olsen)
Woodland	93.6	0.8	5.6	8.6	0.41	5.3
Sand dune	94.5	0.1	5.4	9.1	0.16	2.8
Backdunes	93	1.4	5.6	8.7	0.29	6.7
Wet backdunes	93.8	0.8	5.4	8.7	0.56	3
Brijuni	32.2	26.2	41.6	6.6	3	27

*maritima* Scop., *Echinophora spinosa* L., *Eryngium maritimum* L., *Helichrysum stoechas* (L.) Moench, and some endemic species such as *Centaurea subciliata* DC. and *Solidago litoralis* Savi. Within the sand dune system, which extends approximately 400 m from the seashore, scattered patches of tree vegetation occur, where, within a few square meters, many different plant species co-occur, such as *Cistus incanus* L., *Juniperus oxycedrus* L., *Phillyrea angustifolia* L., *Pinus pinaster* Aiton, *Quercus ilex* L., *Spartium junceum* L. (Arrigoni 1990). Backdune depressions support hydrophytic plant species such as *Cladium mariscus* (L.) Pohl, *Phragmites australis* (Cav.) Trin., and *Typha angustifolia* L. In this habitat, five plant associations have been identified (Arrigoni 1990). The composition of the woodland is complex and characterised by great floristic and phytocoenotic diversity (Arrigoni 1990). The overstorey vegetation is composed of evergreen and deciduous species. *Q. ilex* is the most highly represented plant, mixed with coniferous species, such as *Pinus pinea* L. and *P. pinaster*, which determine the overall appearance of a pine-oak forest. Within the woodland some depression patches are present where the soil is waterlogged depending on seasonal variations of the groundwater level, and where wet forest species such as *Quercus robur* L., *Alnus glutinosa* (L.) Gaertn., *Fraxinus oxycarpa* Bilbb. ex Willd., *Populus alba* L. occur. The understorey vegetation is dominated by Mediterranean shrubs and creepers such as *C. incanus* L., *Hedera helix* L., *P. angustifolia* L., *Smilax aspera* L., *Ulex europaeus* L., and *Ruscus aculeatus* L.. In Brijuni, the mean temperature is 13.9°C and the mean annual precipitation is 817 mm. The predominant plant species belong to typical sclerophyllous Mediterranean vegetation (Macchia), such as *Arbutus unedo* L., *Erica arborea* L., *Myrtus communis* L., *Phillyrea latifolia* L., *Pistacia lentiscus* L., *Q. ilex* and *S. aspera* (Matosevic 1996).

#### Field sampling

The most common plant species occurring in the two sites were sampled (Arrigoni 1990; Matosevic 1996). Plants of Macchia Lucchese were sampled from three different habitats, characterised by different vegetation types: maritime sand dunes, backdune depressions and woodland (Table 2). Root systems with soil were carefully excavated intact, taking care that only roots definitely belonging to root systems of the sampled species were collected. For this reason young seedlings of tree species were uprooted whenever possible. Each specimen was placed in a polyethylene bag, and stored at 4°C upon return to the laboratory, until processed. The mycorrhizal status was determined on root samples of at least three individuals of each species.

#### Assessment of mycorrhizal status

Macroscopic characterisation of the mycorrhizal status was carried out on the basis of morphological features of the fine roots. The root system of each plant was thoroughly washed in running tap water to remove adhered soil, and immediately observed under a Wild (Leica) dissecting microscope. Roots showing external morphology of the ectomycorrhizal type were microscopically characterised after sectioning fresh material with a Leitz Kryostat 1720

freezing microtome. Sections (10–30 µm thick) were mounted in lactic acid and observed under a Reichert-Jung (Vienna, Austria) Polyvar light microscope, equipped with Nomarski interference contrast optics and with epifluorescence optics (HBO 200 mercury lamp). At least ten root samples were examined for each specimen. The presence of fungal mantle and Hartig net was used to define ectomycorrhizal status. Semithin sections were obtained from fresh ectomycorrhizal root tips, after fixation and embedding as described in Filippi et al. (1998).

Root samples devoid of any fungal mantle were cleared in KOH and stained with Trypan Blue in lactic acid (0.01%) and observed under the Polyvar microscope for assessment of endomycorrhizal infection. Some roots of each sample were sectioned with a freezing microtome and observed as described. All root samples were screened for the presence or absence of the different mycorrhizal types. Only plants showing arbuscules in the roots were classified as arbuscular mycorrhizal (AM) species.

Percentages of vegetation cover in Macchia Lucchese were calculated on the basis of Braun-Blanquet abundance-dominance indices reported by Arrigoni (1990).

#### Fluorescence microscopy

To detect the occurrence of intracellular hyphae within root cells, fresh roots showing morphological ectomycorrhizal features were sectioned (10–30 µm thick) with the freezing microtome and labelled with wheat germ agglutinin conjugated with fluorescein isothiocyanate (FITC-WGA), a specific probe for β-linked polymers of *N*-acetylglucosamine (Sbrana et al. 1995). Sections were incubated in a solution containing 50 µg/ml FITC-WGA in 10 mM phosphate buffer, pH 7.5, for 30 min, then washed, mounted in the same buffer, and observed under the fluorescence microscope with the filter combination B1 (BP 450–495, LP 520, DS 510). A positive reaction was indicated by emission of yellow-green fluorescence.

## Results

### Mycorrhizal types in Brijuni National Park

In the Mediterranean Macchia of Brijuni, mycorrhizal plants were present in 28 of the 31 species examined (90%) (Table 2). Five different types of mycorrhizal associations were found. AM occurred in 71% of plant species, while ectomycorrhizas (ECM) were detected in 13%. Arbutoid and ericoid mycorrhizas occurred in *A. unedo* and *E. arborea*, respectively (3%). Non-mycorrhizal plant species were represented by *Atriplex halimus* L., *Juncus maritimus* L. and *Dorycnium hirsutum*.

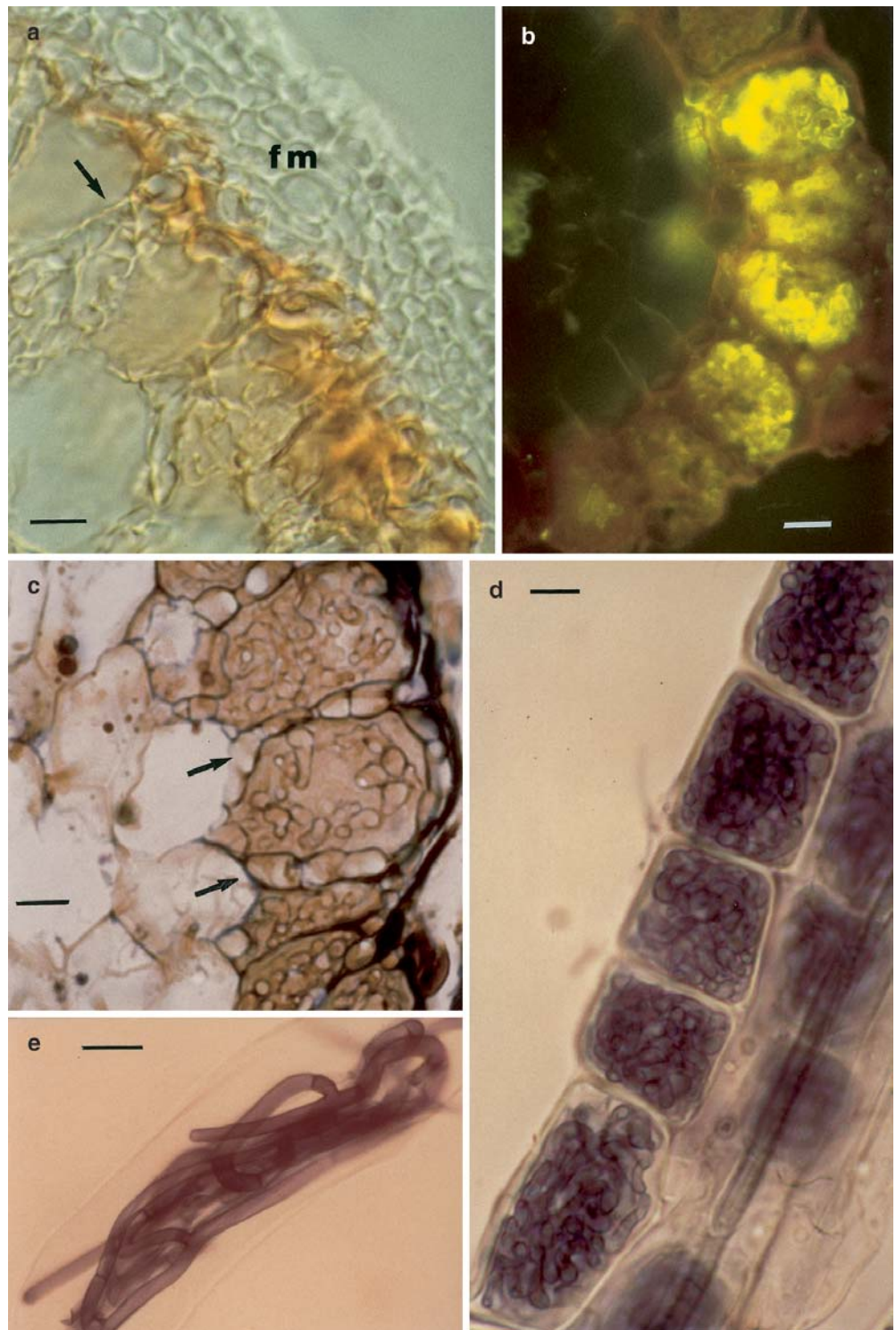
**Table 2** Plant species and types of mycorrhizal associations occurring in two Mediterranean ecosystems, located in Brijuni National Park, Croatia and in Macchia Lucchese Tuscany, Italy. Within Macchia Lucchese, plant species sampled from the different habitats are indicated in parentheses: *W* woodland, *BD* backdune depressions, *SD* sand dunes. *AM* Arbuscular mycorrhizas, *ABT* arbutoid mycorrhizas, *ECM* ectomycorrhizas, *ER* ericoid mycorrhizas, *NM* non-mycorrhizal plants, *OR* orchid mycorrhizas, *ECM+AM* ecto- and arbuscular mycorrhizas in the same root system

Plant species	Mycorrhizal associations		
	Brijuni	Macchia	Lucchese
Alismataceae <i>Baldellia ranunculoides</i> (L.) Parl.		AM	(BD)
Anacardiaceae <i>Pistacia lentiscus</i> L.	AM		
Apiaceae <i>Echinophora spinosa</i> L.		AM	(W, SD)
<i>Eryngium maritimum</i> L.		AM	(W, SD)
<i>Hydrocotyle vulgaris</i> L.		AM	(BD)
<i>Seseli tortuosum</i> L.		AM	(W, SD)
Araliaceae <i>Hedera helix</i> L.	AM	AM	(W)
Asclepiadaceae <i>Periploca graeca</i> L.		NM	(W, BD)
Asparagaceae <i>Asparagus acutifolius</i> L.		AM	(W, SD)
Asteraceae <i>Bidens frondosa</i> L.		AM	(W, BD)
<i>Centaurea subciliata</i> DC.		AM	(W, SD)
<i>Helichrysum italicum</i> G. Don.	AM		
<i>Helichrysum stoechas</i> (L.) Moench		AM	(W, SD)
<i>Otanthus maritimus</i> (L.) Hoffmanns. et Link		AM	(SD)
<i>Senecio bicolor</i> (Willd.) Tod.	AM		
<i>Solidago litoralis</i> Savi		AM	(SD)
Betulaceae <i>Alnus glutinosa</i> (L.) Gaertn.		ECM+ AM	(W, BD)
Brassicaceae <i>Cakile maritima</i> Scop.		NM	(SD)
Caprifoliaceae <i>Lonicera implexa</i> Ait.	AM		
<i>Viburnum tinus</i> L.	AM		
Caryophyllaceae <i>Silene colorata</i> Poir. ssp. <i>canescens</i> (Ten.) Cif. et Giac.		AM	(SD)
Chenopodiaceae <i>Atriplex halimus</i> L.		NM	
Cistaceae <i>Cistus creticus</i> L.	ECM		
<i>Cistus incanus</i> L.		ECM	(W, SD)
<i>Cistus monspeliensis</i> L.	ECM		
Convolvulaceae <i>Calystegia soldanella</i> (L.) R. Br.		NM	(SD)
Cupressaceae <i>Juniperus oxycedrus</i> L. ssp. <i>macrocarpa</i> (Sm.) Ball		AM	(W, SD)
Cyperaceae <i>Carex pendula</i> Huds.		NM	(W, BD)
<i>Cladium mariscus</i> (L.) Pohl		NM	(BD)
<i>Scirpus holoschoenus</i> L.		NM	(W, BD, SD)
Dipsacaceae <i>Pycnocomon rutifolium</i> (Vahl) Hoffmanns. et Link		AM	(W, SD)
Equisetaceae <i>Equisetum ramosissimum</i> Desf.		NM	(W, BD, SD)
Ericaceae <i>Arbutus unedo</i> L.	ABT	ABT	(W)
<i>Erica arborea</i> L.	ER	ER	(W)
Euphorbiaceae <i>Euphorbia peplis</i> L.		AM	(SD)
Fabaceae <i>Coronilla emerus</i> L. ssp. <i>emeroides</i> (Boiss. et Spun) Hayek	AM		
<i>Dorycnium hirsutum</i> (L.) Ser.	NM	NM	(W, SD)
<i>Medicago littoralis</i> Rohde		AM	(BD, SD)
<i>Medicago marina</i> L.		AM	(SD)
<i>Spartium junceum</i> L.	AM	AM	(W)
<i>Ulex europaeus</i> L.		AM	(W)
Fagaceae <i>Quercus ilex</i> L.	ECM	ECM	(W, SD)
<i>Quercus robur</i> L.		ECM	(W)
Gentianaceae <i>Blackstonia perfoliata</i> (L.) Huds.		AM	(BD)

Table 2 (continued)

Plant species	Mycorrhizal associations	
	Brijuni	Macchia Lucchese
Iridaceae		
<i>Iris pseudoacorus</i> L.		AM (W, BD)
Lamiaceae		
<i>Mentha aquatica</i> L.		AM (W, BD)
<i>Stachys maritima</i> Gouan		AM (SD)
<i>Stachys recta</i> L. var. <i>psammophila</i> Fiori		NM (SD)
Lauraceae		
<i>Laurus nobilis</i> L.	AM	AM (W)
Myrtaceae		
<i>Myrtus communis</i> L.	AM	
Oleaceae		
<i>Fraxinus oxycarpa</i> Bieb. ex Willd.		AM (W, BD)
<i>Phillyrea angustifolia</i> L.		AM (W)
<i>Fraxinus ornus</i> L.	AM	
<i>Phillyrea latifolia</i> L.	AM	
<i>Olea europaea</i> L. spp. <i>sylvestris</i> (Mill.) Rouy	AM	
<i>Ligustrum vulgare</i> L.	AM	
Orchidaceae		
<i>Cephalanthera rubra</i> (L.) Rich.		OR (W)
<i>Spiranthes spiralis</i> (L.) Koch.		OR (BD)
Plantaginaceae		
<i>Plantago major</i> L.		AM (BD)
Poaceae		
<i>Ammophila arenaria</i> (L.) Link ssp. <i>Arundicea</i> Lindb.		AM (SD)
<i>Phragmites australis</i> (Cav.) Trin. ex Steud.		AM (BD)
Polygalaceae		
<i>Polygala nicaeensis</i> Risso ex Koch ssp. <i>mediterranea</i> Chodat		AM (W)
Pinaceae		
<i>Pinus pinaster</i> Aiton		ECM (W, SD)
<i>Pinus pinea</i> L.		ECM (W, SD)
Primulaceae		
<i>Lysimachia vulgaris</i> L.		AM (W, BD)
<i>Samolus valerandi</i> L.		AM (BD)
Ranunculaceae		
<i>Clematis vitalba</i> L.	AM	
Rosaceae		
<i>Crataegus monogyna</i> Jacq. emend. Lindm.	ECM	
<i>Malus domestica</i> Borkh		AM (W)
<i>Rosa sempervirens</i> L.	AM	
<i>Rubus ulmifolius</i> Schott	AM	AM (W)
Rubiaceae		
<i>Rubia peregrina</i> L.	AM	
Ruscaceae		
<i>Ruscus aculeatus</i> L.		AM (W)
Salicaceae		
<i>Populus alba</i> L.		ECM+ AM (W)
Scrophulariaceae		
<i>Odontites lutea</i> (L.) Clairv.		AM (W, SD)
Smilacaceae		
<i>Smilax aspera</i> L.	AM	AM (W, SD)
Tamaricaceae		
<i>Tamarix gallica</i> L.	AM	
Thymelaeaceae		
<i>Daphne gnidium</i> L.		AM (W, SD)
Typhaceae		
<i>Typha angustifolia</i> L.		NM (BD)
Verbenaceae		
<i>Vitex agnus – castus</i> L.	AM	
<i>Teucrium flavum</i> L.	AM	
Juncaceae		
<i>Juncus maritimus</i> L.	NM	

**Fig. 1a–e** Light micrographs of different mycorrhizal types occurring in Mediterranean areas. **a** Cross section of *Cistus incanus* ectomycorrhiza showing a well developed fungal mantle (*fm*) surrounding the root and intercellular hyphae (arrow). Scale bar 10  $\mu$ m. **b** Cross section of arbutoid mycorrhiza of *Arbutus unedo*, after treatment with wheat germ agglutinin conjugated with fluorescein isothiocyanate (FITC-WGA), showing intense green labelling of intracellular fungal coils. Scale bar 12  $\mu$ m. **c** Semithin cross section of *A. unedo* mycorrhiza showing a well developed Hartig net (arrows) and intracellular colonisation of epidermal root cells. Scale bar 8  $\mu$ m. **d** Whole mount of a hairy root of *Erica arborea* showing root cells completely invaded by the ericoid fungal symbiont. Scale bar 10  $\mu$ m. **e** Individual root cell of *Cephalanthera rubra* showing intracellular hyphal coil. Scale bar 13  $\mu$ m.



### Mycorrhizal types in Macchia Lucchese

In Macchia Lucchese, 83% of the plant species surveyed were mycorrhizal. On the basis of macro- and micro-sopic characteristics, six morphotypes of mycorrhizal associations were classified. Of the 60 species examined, AM were present in 41 (68%), ECM in 7 (12%), and orchid mycorrhizas in 2 (3%). Ericoid and arbutoid mycor-

rhizas occurred in only one species each (2%) (Table 2). All individuals of the same species showed the same mycorrhizal type. Two species, *A. glutinosa* (both in woodland and backdune depressions) and *P. alba*, showed both ECM and AM, though in different portions of the root system.

Although the aim of the present study was not the description of ectomycorrhizal morphological types, obser-

vations under the dissecting microscope showed many differences among ECM occurring in different plant species. For example, *P. pinea* and *P. pinaster* roots showed characteristic dichotomic mycorrhizal structures, whereas *Q. ilex* and *Q. robur* showed irregularly branched mycorrhizas. A peculiar feature of *C. incanus* mycorrhizas was represented by single, unbranched roots showing elongated finger-like root apices surrounded by a well-developed fungal mantle (Fig. 1a). The occurrence of *Cenococcum*-type mycorrhizas, characterised by a thick, black, crusty mantle, from which straight, black hyphae radiated outward, was detected in *C. incanus* and *Q. robur*.

Sections of fresh *A. unedo* roots showed arbutoid ectoendomycorrhizas with typical cruciform morphology, a fungal mantle, a visible Hartig net and highly coiled intracellular hyphae, which were confirmed by the fluorescence FITC-WGA labelling (Fig. 1b). Observations on semithin sections evidenced the occurrence of intercellular Hartig net hyphae and intracellular infection of epidermal cells (Fig. 1c).

Root sections of *P. alba*, and *A. glutinosa* showed both ectomycorrhizas and intracellular fungal colonisation. The staining of such sections with Trypan Blue showed many hyphae devoid of septa infecting cortical cells and forming arbuscular structures.

Ericoid mycorrhizas occurred in *E. arborea* and orchid mycorrhizas were found in *Cephalanthera rubra* (L.) Rich. and *Spiranthes spiralis* (L.) Koch. (Fig. 1d, e).

Some species described in the literature as lacking mycorrhizas, such as *Carex pendula* Huds., *Scirpus holoschoenus* L. (Cyperaceae) and *D. hirsutum* (L.) Ser. (Fabaceae), had vesicles and intraradical hyphae in the roots (Harley and Harley 1987; Smith and Read 1997). Seven plant species did not have any intraradical fungal colonisation.

All AM plants showed *Arum*-type colonisation, except *S. aspera*, an evergreen climbing species. Detailed observation on stained roots and on fresh root sections showed a *Paris*-type infection: the fungus colonised roots by growing intracellularly from cell to cell and forming many coils and terminal arbuscules (Bedini et al. 2000).

#### Mycorrhizal types in different habitats of Macchia Lucchese

Different occurrence patterns of mycorrhizal types could be recognised in different habitats within Macchia Lucchese. The greatest diversity of mycorrhizal types occurred in the woodland, where all the described mycorrhizal types were represented (except monotropoid): ECM, arbutoid, ericoid, orchid mycorrhizas and AM (both *Arum*- and *Paris*-type). On the whole, in the woodland, ECM occurred in 17% of plant species, arbutoid mycorrhizas in 2%, ericoid mycorrhizas in 2%, orchid mycorrhizas in 2%, and AM in 68% (Table 2). Two plant species showed both ECM and

AM infection and non-mycorrhizal species represented 12% (Table 2). The distribution of the different mycorrhizal types on a vegetation cover basis (versus the floristic basis hitherto used) was calculated by using the percent covers of the plant species surveyed occurring in the main associations, as reported in Arrigoni survey tables (1990). In the *Viburno-Quercetum ilicis* (Br. Bl.) Riv Martin., within the woodland, data on the percent occurrence of mycorrhizal types on a floristic basis – 29% ECM and 57% AM – are different from data calculated on a vegetation cover basis – 74% ECM and 26% AM. In the wet forest association *Alnion glutinosa* Meijr.Drees within the woodland, data calculated on a floristic basis showed that ECM occurred in 12% of plant species, AM in 63% and plants with both mycorrhizal types represented 12%; in contrast, on a vegetation cover basis, ECM occurred in 0.4% plants, AM in 12%, whereas double ecto- and arbuscular colonisation occurred in 86% of plants. Likewise, in the other wet forest association, *Alno-Fraxinetum oxycarpae* (Br. Bl.) Tchou, 15% of plants showed ECM and 62% AM on a floristic basis, whereas the frequency was 4% and 65%, respectively, on a vegetation cover basis. In the mixed pine forest, AM occurred in 61% and ECM in 19% of plant species on a floristic basis, whereas on a vegetation cover basis AM occurred in 14% and ECM in 86% of plants.

The lowest mycorrhizal richness was found in maritime sand dunes, where AM was present in 66% of plant species and ECM in 14%, whereas 21% of plants were nonmycorrhizal (Table 2). In the plant association *Echinophoro-Ammophiletum arenariae* (Br. Bl.) Gehu within the sand dune system, AM and nonmycorrhizal plant species were 83% and 17%, respectively, on a floristic basis, and 71% and 3% on a vegetation cover basis.

In backdune depressions, AM occurred in 65% of plants surveyed, species showing both ECM and AM represented 5%, and 30% of species were non-mycorrhizal species (Table 2). In *Cladietum marisci* association within backdune depressions, AM occurred in 67% of plant species and non-mycorrhizal plants represented 33%, whereas data calculated on a vegetation cover basis showed that non-mycorrhizal plants represented 96% and AM only 4%.

#### Discussion

We have investigated the mycorrhizal types of the most thoroughly sampled Mediterranean plant communities to date by sampling 82 species of 46 families. Six different mycorrhizal types were classified, confirming previous reports on the co-occurrence of diverse mycorrhizal associations in the same plant community (Moyersoen et al. 1998) and on mycorrhizal diversity in Mediterranean ecosystems (Read 1991). Moreover, our results showed that the abundance of mycorrhizal types in diverse plant associations within Macchia Lucchese calculated on a

floristic basis were very different from those obtained using a vegetation cover index.

The mycorrhizal type of most plant species surveyed confirmed data reported by other authors. AM was found in 68% and in 71% of plant species in Macchia Lucchese and Brijuni Macchia, respectively, showing the extensive occurrence of this mycorrhizal type in Mediterranean ecosystems (Lamont 1982; Trappe 1987). Most forest trees such as *Pinus* spp. and *Quercus* spp., had ECM, *A. unedo* had arbutoid and *E. arborea* ericoid mycorrhizas (Smith and Read 1997). Our study confirmed the occurrence of arbutoid mycorrhizas in *A. unedo* growing in natural sites (Giovannetti and Lioi 1990). We described for the first time the mycorrhizal status of some endemic plant species such as *C. subciliata* and *S. litoralis*, which had AM colonisation.

A group of species belonging to families generally considered non-mycorrhizal, such as Cyperaceae and Typhaceae (Harley and Harley 1987; Tester et al. 1987; Peat and Fitter 1993), were devoid of any mycorrhizal symbiosis according to our findings, although intraradical aseptate hyphae and/or vesicles were present in some roots of *C. pendula* and *S. holoschoenus*. In our report, we used the occurrence of arbuscules as the only positive indication, by definition, of the establishment of an AM symbiosis, though other authors attributed AM status to species showing only hyphae and/or vesicles in their roots (Stenlund and Charvat 1994; Miller et al. 1999). The absence of arbuscules and appressoria, and the occurrence of hyphae and/or vesicles in the roots of non-host plants, in dead roots, pieces of organic matter, and organs other than roots have been considered by some authors as an indication of the ability of passive colonisation by AM fungi (Park and Linderman 1980; Malajczuk et al. 1981; Stasz and Sakai 1984; Giovannetti and Lioi 1990; Giovannetti et al. 1994; Giovannetti and Sbrana 1998).

The low incidence of non-mycotrophic species in Macchia Lucchese (17%) and in Brijuni Macchia (10%) suggests the inability of non-mycotrophic species to compete successfully with all the diverse mycorrhizal species (Peat and Fitter 1993).

We compared the distribution of different mycorrhizal types in relation to the floristics and vegetation cover of six plant communities (Arrigoni 1990). Our results showed that mycorrhizal type abundance calculated on a floristic basis was very different from that obtained on a vegetation cover basis. As an example, in the *Viburno-Quercetum ilicis* association, ECM occurred in 29% of plant species and AM in 57%, on a floristic basis. This result does not represent the actual relative abundance of these two mycorrhizal types in the plant community surveyed; in fact, when calculating their occurrence on a vegetation cover basis, ECM was present in 74% of plants and AM in 26%. Accordingly, both floristic and vegetation cover data should be considered when investigating how different mycorrhizal types influence the occurrence of distinctive patterns of mycorrhizal associations in different biomes and the success of host plants

(Brundrett and Kendrick 1988; Peat and Fitter 1993; Moyersoen et al. 1998).

Our work showed the occurrence of both ECM and AM in *A. glutinosa* and *P. alba*. Multiple infection has been described as a general phenomenon in the genus *Eucalyptus* and in *A. glutinosa* (Lapeyrie and Chilvers 1985; Chilvers et al. 1987; Harley and Harley 1987) and a succession over time of two overlapping mycorrhizal systems has been described in *Helianthemum chamaecistus* (Read et al. 1977). Only a few studies have compared the effects of the two mycorrhizal associations on host growth, and the relative importance of the two types of symbiosis under experimental conditions (Chatarpaul et al. 1989; Osonubi et al. 1991; Jones et al. 1998). Further studies could reveal the role played by plant hosts of both AM and ECM in the distribution of resources within the forest community, given the ability of ECM and AM fungi to associate with multiple hosts and to translocate nutrients between plants via mycorrhizal hyphae (Francis and Read 1984; Finlay and Read 1986; Molina et al. 1992; Simard et al. 1997a, b; Horton and Bruns 1998).

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