Journal of Mediterranean Ecology **vol. 4, No.3-4, 2003: 25-35**
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Leaf xeromorphic adaptations of some plants of a coastal Mediterranean macchia ecosystem

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Key words: Drought, leaf anatomy, histology, ecophysiology, gas exchanges.

Abstract

Water appears as the major environmental constraint in Mediterranean climate, and global change effects are likely to provide more frequent and longer drought periods. Water shortage has significant demographic effects on ecosystem composition, being some plant species thriving in arid environments because of a combination of several anatomical and physiological adaptations.

In this study, co-occurring species of a typical thermo-Mediterranean macchia ecosystem were studied to survey the main anatomical features. Observations performed confirmed the presence of several adaptive properties for the macchia ecosystem to couple with the high temperature and solar radiation and the lack of summer rainfall typical of the Mediterranean climate. Most of the species examined show the presence in their internal leaf tissues of ergastic substances, mainly tannins and calcium oxalate, with defensive functions, crucial in the adaptive resistance of plants to water stress. Nearly all the species present adaptations for protection against the photo damage possibly induced from the strong UV-B solar irradiance in the summer. The more significant anatomical features are the trichomes, covering the abaxial surfaces of leaves. Such structures are able to regulate the water budget of the plants both by influencing the diffusion boundary layer of the leaf surface and by regulating leaf optical parameters and, hence, leaf temperature. In many species, when trichomes or wax layers reduce radiation absorbance, two or three layers of palisade parenchyma are present, presumably concurring to provide a better efficiency in utilizing the photosynthetic light. In almost every plant examined, stomata are sunken or well protected.

This paper reports also the annual courses of the leaf water potential, net photosynthesis and transpiration rates of the four dominant species of the population (*Juniperus phoenicea L., Pistacia lentiscus L., Phillyrea angustifolia L. and Chamaerops humilis L.*) and the ecosystem flux.

Introduction

The ecosystems typically associated with Mediterranean climates with warm, dry summers are constituted mostly by low-stature sclerophyll evergreen shrubs. According to the five different regions of the world characterized by a Mediterranean climate, these ecosystem are named in different ways (Cody & Mooney, 1978). In California, this community is known as “chaparral”, while its name is “fynbos” in the Cape Province of South Africa, “matorral” in Chile, “malle” in Australia and “macchia” or “maquis” in the European Mediterranean basin (Specht, 1969; Valiente-Bonuet et al., 1998).

Mediterranean ecosystems are fragile and sensitive environments. Their soils have been severely eroded and natural vegetation dramatically modified through several thousand of years of human activities and land use. The concern about the safeguard of these ecosystems is reinforced by environmental, ecological and biological argumentations since the Mediterranean-type plant communities are a recognized reservoir of biodiversity.

Water appears as the major environmental constraint in Mediterranean climates, and the global change effects are likely to provide more frequent and longer droughts (Hulme, 1996; Penuelas et al., 1998; Chen, 2002). Water shortage has significant demographic effects on ecosystem composition, being some species thriving in arid environments because of a combination of several physiological and anatomical xeromorphic (Greek xeros = dry) adaptations.

In this study, co-occurring species of a typical thermo-mediterranean macchia ecosystem were studied to survey the main leaf anatomical features in relation to local meteo-climatology. The annual CO_2 flux of the ecosystem and the main ecophysiological processes of the four dominant species were also observed and analyzed.

Methods

Site description

The experimental site is located within a nature reserve named “Arca di Noè” in a coastal area of North-Western Sardinia, Italy (40°36’N, 8°9’E 74 m above sea level) (Fig.1). The area represents one of the 114 natural habitats included in the framework of the European network NATURA 2000.

The climate is typical of mid-latitude Mediterranean islands. Most of the rainfall occurs in winter and autumn and summers are hot and dry (Chessa et al., 1999; Delitala et al., 2000). Temperatures are those of the subtropics moderated by maritime influence. The variability of annual rainfall often causes severe drought periods with water deficit also in spring and winter (Duce et al., 1999).

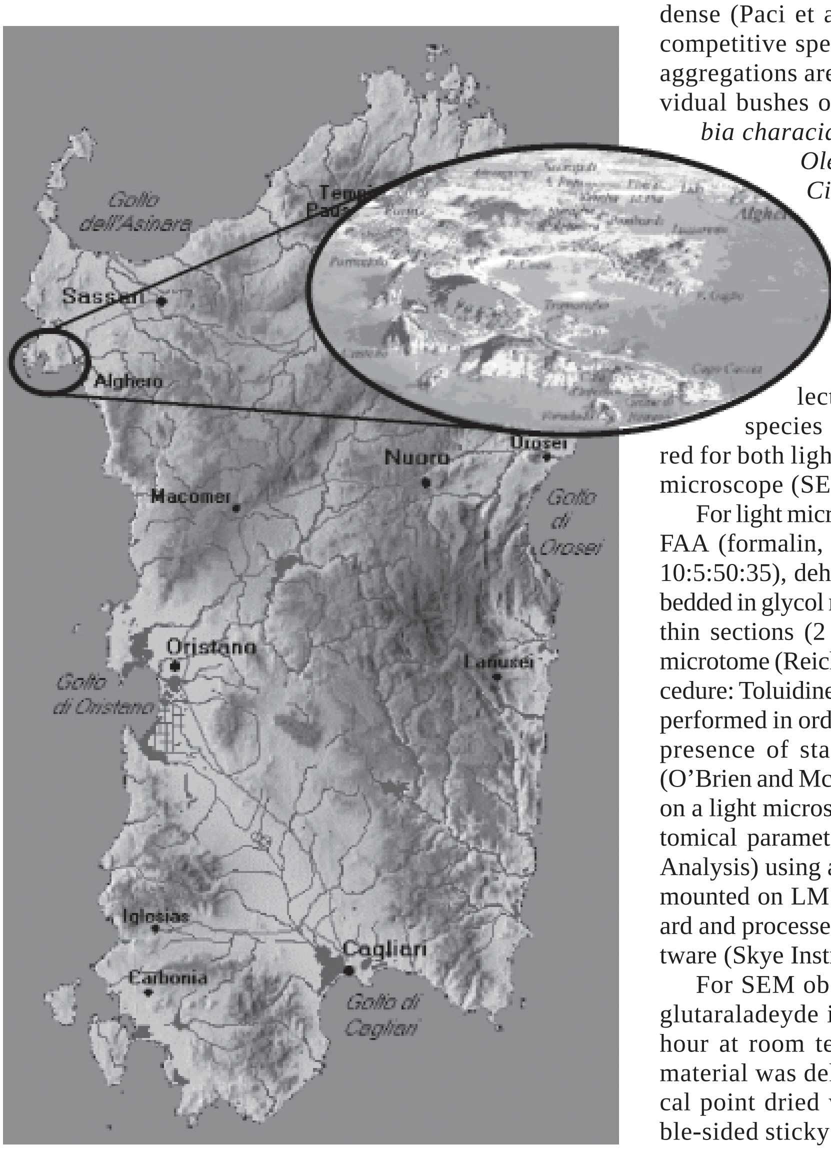

Fig.1 - The location of Arca di Noè site (in the circle on the left).

According to the USDA classification (1997), the prevailing soils are Lithic- Xerorthent and Typic Rhodoxeralfs, with an average depth of about 0.30 m.

The vegetation of the area can be distinguished in two main zones: a typical Mediterranean macchia and a coastal garigue, where stones and calcareous rocks surround small shrubs and herbaceous species. In the coastal zone, many important endemic species can be found: *Astragalus massiliensis* L., *Centaurea horrida Bad*., *Helichrysum italicum* (Roth) Don subsp. *Microphyllum* (Willd) Nyman, *Genista acanthoclada* DC subsp. sardoa (Bèg et Landi) Valsecchi.

The dominant species of the macchia vegetation are: juniper (*Juniperus phoenicea* L.), lentisk (*Pistacia lentiscus* L.), tree phillyrea (*Phillyrea angustifolia* L.) and dwarf fan palm (*Chamaerops humilis* L.). These species form a sparsely vegetated plain shrubland with plants aggregated in patches variable in size. A study on the spatial distribution and the competition relationship between the dominant species conducted where the canopy is more dense (Paci et al., 1999) shows that juniper is the most competitive specie, determing pure aggregate. The main aggregations are represented by juniper and lentisk. Individual bushes of *Rosmarinus officinalis* L. and *Euphorbia characias* L. are also present. *Olea europea* var. *sylvestris* Hofmgg et Link, *Cistus monspeliensis* L., *Myrtus communis* L. and *Arbutus unedo* L., cover an area opposite to the coastal line.

Leaf morphology and histology

Five fully-developed leaves were collected during the autumn season from all the species populating the nature reserve and prepared for both light microscope (LM) and scanning electron microscope (SEM) observations.

For light microscope observations, leaves were fixed with FAA (formalin, acetic acid, ethanol 95%, distilled water, 10:5:50:35), dehydrated in a graded ethanol series and embedded in glycol methacrylate resin (GMA). Transverse semithin sections (2 μm) were made with an autocut rotative microtome (Reichert-Jung, Germany). A double staining procedure: Toluidine Blu and Periodic Acid-Schiff’s reagent was performed in order to evidence leaf structures, as well as the presence of starch grains and poliphenols and tannins (O’Brien and McCully, 1981). Observations were carried out on a light microscope (Leitz DM RB, Germany). Main anatomical parameters were measured by VIA (Video Image Analysis) using a CCD camera (JVC model NO.TK-880 E) mounted on LM Images were digitized with an ELVIS board and processed with the Chameleon Image Analysis Software (Skye Instruments, UK).

For SEM observations, leaves were fixed with 2,5 % glutaraladeyde in 0,1 M phosphate buffer (pH 7,2) for 1 hour at room temperature. After washing in buffer, the material was dehydrated in a graded ethanol series, critical point dried with CO_2 , mounted on stubs using double-sided sticky tape and coated with a thin layer of gold. Observations were carried out on Philips SEM 515 scanning electron microscopy operating at 10 kV. The description of anatomical characters has been done according to Westerkamp and Demmelmeyer (1977).

Ecophysiological observations

The ecophysiological behavior of the four dominant species (juniper, lentisk, tree phillyrea and dwarf fan palm) was also studied.

Leaf assimilation rate (A), leaf transpiration rate (E), stomatal conductance (gs) and water potential (ψ) were measured in 1998 using a portable infrared gas analyzer (LCA2, ADC, UK) and an electronic pressure chamber. The gas exchange and water potential measurements were taken twice a month on four plant per species using two samples per plant. Samples were taken on mature leaves for dwarf fan palm, and 1-year old shoots for lentisk, juniper and phillyrea.

Local meteo-climatology and flux measurements

Climate data were obtained from a meteorological station located at 5 km from the experimental area.

Meteorological and carbon dioxide flux data collected by stations located in the same site in the frame of the E.U. project MEDEFLU.

Flux data were taken by eddy covariance technique using a triaxial sonic anemometer (Gill 3D, Solent, UK) and a fast H_2O / CO_2 gas-analyzer Li-6262 (Li-Cor, Nebraska, USA).

Results

Leaf morphology and histology

Leaf histological parameters, stomata and trichome densities are respectively summarized in Tables 1 and 2. The main anatomical leaf features of each species are reported together with the light and electron microscope images.

The leaf internal structure of *Pistacia lentiscus* L. (Fig. 2a, magnification 100x) evidences a upper epidermis formed by a single-layer of cells, covered by a thick cuticle (Fig. 2b, 400x).

The palisade parenchyma is organized in 2-3 layers of elongated cells and the spongy tissue is characterized by wide lacunas and by one layer of elongated cells bordering the lower epidermis, reminding a typical isolateral structure. Isodiametric convex epidermal cells form a coarsely undulated abaxial surface, where stomata are unaligned, leveled with surface, elliptic with flat guard cells (Fig.2c, 430x). Starch grains and phenolic substances were present in the palisade tissue. Obvious wax crystalloids layers cover completely the adaxial epidermis. The few uncovered areas show isodiametric convex epidermal cells (Fig. 2d, 1200x).

The leaves of *Phillyrea angustifolia* are equifacial (Fig. 3a, 100x): the palisade tissue occurs both at adaxial and abaxial surfaces, being the upper palisade formed of more tightly-packed and elongated cells. The spongy parenchyma is located between the two palisade layers. Both epidermises consist of one layer of isodiametric cells (larger and more convex in the adaxial) covered by a very thick cuticle. In the lower epidermis the recurrence of deep crypts which contain pluricellular, unbranched capitate hairs with smooth surface is remarked together with obvious, irregularly distributed crystalloids (Fig. 3b, 400x). Numerous unaligned sunken stomata, with convex guard cells are present (Fig. 3c, 600x). In the adaxial surface the hairs are rare and confined in less deep crypts, the stomata are absent and the wax crystalloids overall distributed (Fig. 3d, 1200x).

Juniperus phoenicea has squamiform and imbricate leaves (Fig 4a, 50x). Both the epidermises are constituted of

Tab. 1 Leaf histological parameters. Values expressed in µm reported the thickness of different tissue.

Species	Total lamina	Upper cuticle	Upper epidermis	Upper palisade	Spongy tissue	Lower palisade	Lower epidermis	Lower cuticle	Hypodermis	Palisade/total mesophyll
Pistacia l.	459	11	12	193	221	-	13	9	-	0,42
Phillyrea a.	372	13	14	137	112	72	8	16	-	0,56
Chamaerops h.	378	5	7	355^	n.p.	-	7	-	-	0,94
Juniperus p.	252	n.m.	10	70	160	-	12	n.m.	11	0,35
Olea e.	330	6	16	114	181	-	11	2	-	0,35
Cistus m.	259	-	28	86	128	-	17	-	-	0,33
Rosmarinus o.	345	16	14	141	95	-	9	-	70	0.41
Myrtus c.	291	2	18	51	204	-	14	2	-	0,18
Arbutus u.	387	9	27	168	156	-	21	6	-	0,43
Euphorbia c.	501	10	43 ++	211	201	-	31 +++	5	-	0,42
Helichysum i.	233	4	22	75	119	-	13	-	-	0,32
Astragalus m.	381	9	21	191	33	103	16	8	-	0,77
Centaurea h.	343	5	23	149	143	-	19	4	-	0,42
Genista s.	874 §	22	8	225*	-	-	-	-	-	0,26

Legend:
- not present n.m. not measurable * = photosynthetic tissue § = thorn diameter ^ = uni-facial structure
++ = upper papillae included +++ = lower papillae included

Tab. 2 Leaf surface parameters.

	Upper surface		**Lower surface**		
Species	Stomatal density (N/mm^2)	Hair density (N/mm^2)	Stomatal density (N/mm^2)	Hair density (N/mm^2)	Max diameter Stomata (µm)
Pistacia lentiscus	-	-	*254*	-	*3.64*
Phillyrea angustifolia	-	*n.m.*	*213*	*8*	*1.19*
Chamaerops humilis	*n.m.*	-	*n.m.*	-	*0.79*
Juniperus phoenicea	*n.m.*	-	*n.m.*	-	-
Olea europea	-	*n.m.*	*n.m.*	*58*	*2.02*
Cistus monspelliensis	-	-	*n.m.*	*n.m.*	-
Rosmarinus officinalis	-	-	*n.m.*	*n.m.*	-
Myrtus communis	-	-	*342*	-	*1.67*
Arbutus unedo	-	-	*177*	-	*2.08*
Euphorbia characias	-	*n.m.*	*28*	*31*	*2.0*
Helchrysum italicum	*n.m.*	*n.m.*	*n.m*	*n.m.*	-
Astragalus massiliensis	*160*	*33*	*94*	*31*	*1.08*
Centaurea horrida	*126*	*n.m.*	*152*	*n.m.*	*1.0*
Genista sardoa*	*81*	-	-	-	*0.95*

- not present
* = thorn surface

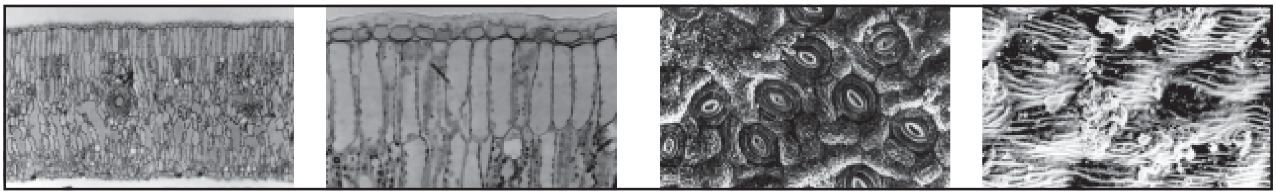

Fig. 2a - 2b - 2c - 2d.

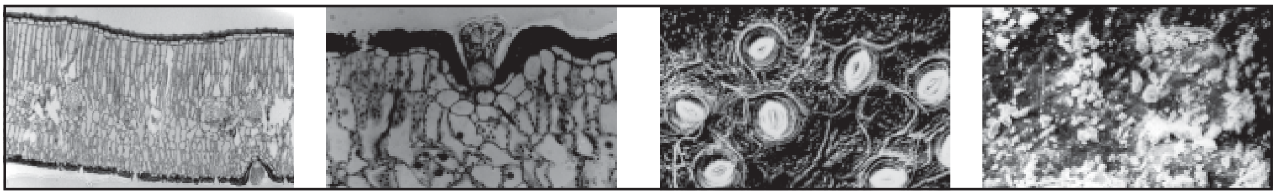

Fig. 3a - 3b - 3c - 3d.

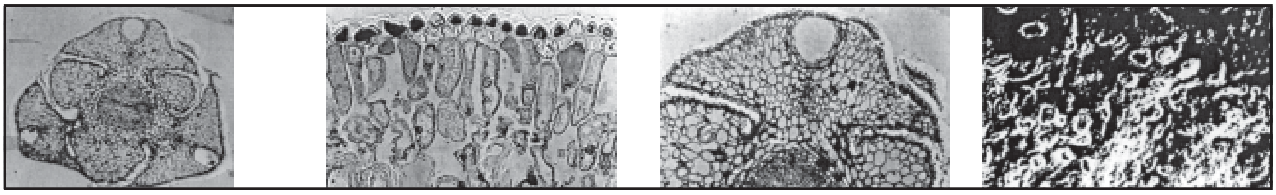

Fig. 4a - 4b - 4c - 4d.

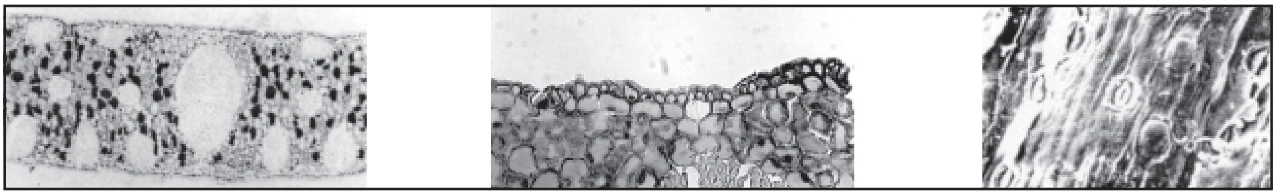

Fig. 5a - 5b - 5c.

one-layer of cells with the outer walls provided by cuticular ornamentation similar to papillae (Fig. 4b, 400x). The hypodermal layer of sclerenchyma beneath the upper epidermis is interrupted by stomata, present in both surfaces. In the lower side, stomata are more numerous, sunken and covered by a cuticular extroversion. Palisade and spongy parenchymas are plicate, and vertical invaginations from the main walls protrude into the protoplast. Resinous ducts present in the upper side of the mesophyll are delimitated by three cell layers, mechanical, parenchymatous and secretory respectively (Fig. 4c, 100x). The external surface observation evidences clear wax crystalloids and unaligned stomata visible when the wax layer is interrupted (fig 4d, 1200x). The resinous ducts are hidden by a longitudinal relief on the upper side of the leaves.

The unifacial structure typical of monocotyledons characterizes the leaves of *Chamaerops humilis* (Fig. 5a, 100x). Mesophyll is formed by both elongated and isodiametric cells filled with protein grains and phenolic substances. Both epidermises are constituted by one layer of cells bordering at the internal side with an hypodermic layer of larger cells and sclerenchymatic cell groups. Only one epidermis has the cuticular layer (Fig. 5b, 400x). Both surfaces are covered by evident platelets and rods of wax which hide numerous elliptic stomata aligned and parallel to the longitudinal axis of the leaf. The stomata are lightly sunken, formed by dumbbell-shaped guard and large subsidiary cells, and surrounded by a raised ring of wax (Fig. 5c, 600x).

Bounches of 5 linear and coriaceous leaves, with glossy dark-green upper surface and light-green lower surface characterize the specie *Rosmarinus officinalis.* The traverse section of the lamina shows markedly revolute margins and the semi-square sections of the main vein. The upper epidermis is covered by a very thick cuticular layer and the two-layered palisade parenchyma is interrupted by the protrusions of hypodermal layer. Spongy parenchyma represents only few percent of the entire lamina (Fig. 6a, 100x). The palisade cells are heavily packed with abundant starch grains (Fig. 6b, 400x). The lower epidermis is formed by one layer of small-size cells and the stomata are markedly raised above the surface. This surface is covered by a dense *indumentum* of long, dendritic, pluricellular, tip-pointed hairs mixed with glandular hairs with large oleiferous heads and capitate trichomes (Fig. 6c, 300x). The upper epidermis, scabrous and corrugated, is characterized by many pedestals of very fragile hairs, probably collapsed during the sampling treatment (Fig. 6d, 300x).

Both surfaces of the leaves of *Euphorbia characias* are bluish-green colored and markedly covered with hairs. The total lamina is very thick and is constituted by one or two layers of highly compacted cells of palisade parenchyma and a spongy mesophyll with very wide lacunae. Cells of both epidermises are filled up with phenols and protein grains (Fig. 7a, 50x). Abaxial epidermis is covered by cuticular papillae with striate surface very raised above the surface. Stomata, recessed into surface, are surrounded by 6 papillae (fig 7b, 1200x). Numerous long tip-pointed hairs with large pedestal are inserted among the papillae (Fig. 7c, 300x). The indumentum of the upper surface is characterized by sporadic and short hairs with a large base inserted in the papillae that are less raised above the surface (fig. 7d, 300x).

The internal structure of *Olea europaea* shows the one-layered upper epidermis formed by cells with the external wall thickened and covered by very thick cuticle. The palisade parenchyma is formed by 2-3 layers of very elongated and tightly-packed cells. The spongy parenchyma is characterized by wide lacunae and filiform sclerenchymatic cells (Fig. 8a, 100x). T-shaped sclerenchymatic cells are frequently distributed under upper epidermis (Fig. 8b, 400x) and both in palisade and spongy tissues The one-layered lower epidermis is interrupted by numerous stomata and peltate hairs sunken under the surface; these hairs consist of a short stalk cell and a large head organized in 5-10 cel-

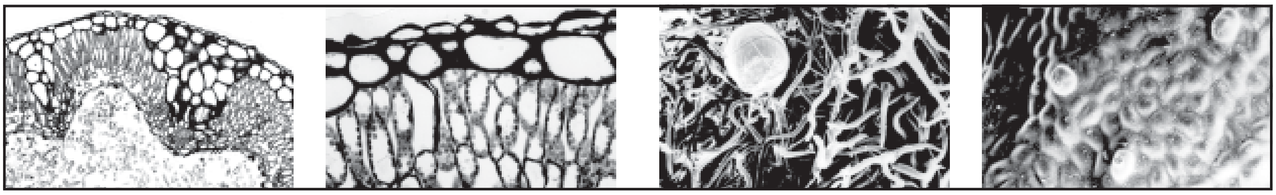

Fig. 6a - 6b - 6c - 6d.

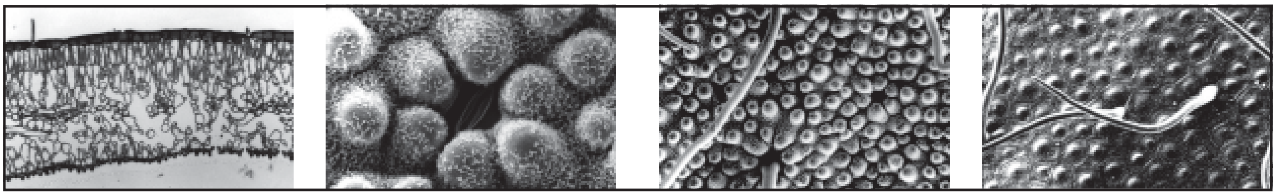

Fig. 7a - 7b - 7c - 7d.

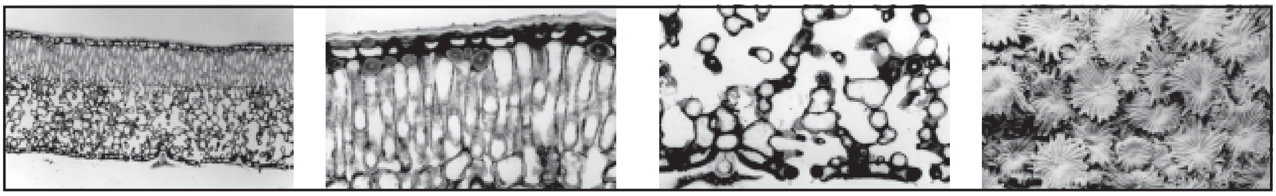

Fig. 8a - 8b - 8c - 8d.

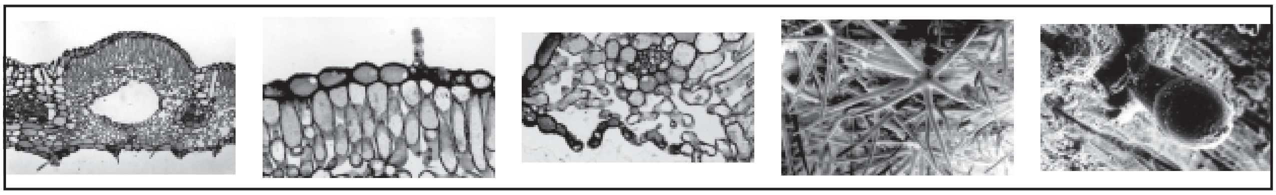

Fig. 9a - 9b - 9c - 9d - 9e.

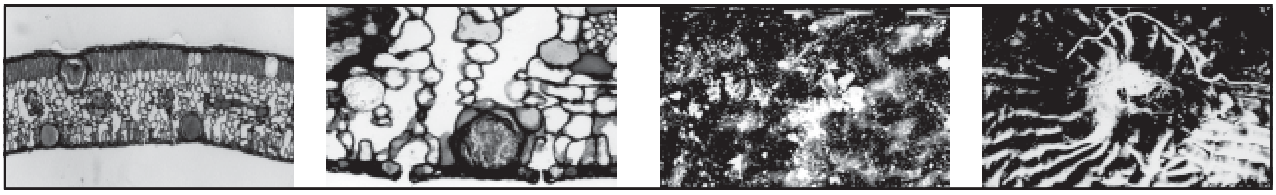

Fig. 10a - 10b - 10c - 10d.

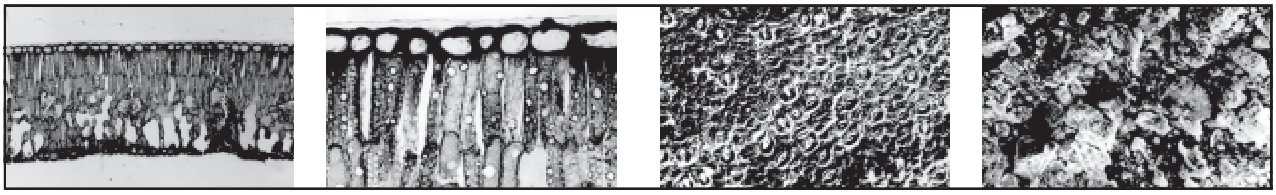

Fig. 11a - 11b - 11c - 11d.

ls. (Fig. 8c, 400x). Numerous peltate hairs overlap completely the abaxial surface, and hide elliptical-shaped stomata (Fig.8d, 170x). Many areas show large plates of wax entirely coating the peltate hairs. Adaxial surface is completely covered by a wax layer organized in platelets and crystalloids. Rare and less developed hairs are present.

The lanceolate leaves of *Cistus monspeliensis* are triveined with the main vein very raised above the surface. Both surfaces are rough and lobated with the abaxial one dull-green colored because of the presence of hair *indumentum*. The total lamina thickness is very irregular, being double in some areas. Spongy parenchyma is characterized by the presence of wide crypts completely coated with stomatal openings. In some cases, the crypts are totally inserted in the mesophyll without any external communication (Fig. 9a, 50x). Upper epidermis is formed by one layer of cells, rare capitate hairs with a short stalk cell and a pluricellular head are observed. Two or three layers of elongated and very packed cells form the palisade parenchyma. (Fig. 9b, 400x). The lower epidermis shows numerous stomata very raised above surface (Fig. 9c, 400x) and numerous pluri-cellular insertions of dendritic hairs. The abaxial surface is deeply incised forming a multi-lobated structure covered by two kinds of hairs: very dense dendritic (Fig. 9d, 300x) and capitate trichomes with a large head and smooth surface (Fig. 9e, 2100x). On the main vein surface long simple hairs with pointed-tip are evident. Also the adaxial surface has a multi-lobated surface covered by both short- and long-stalked capitate glandular trichomes, characterized by a large head. Obvious wax crystalloids are overall distributed.

Spongy tissue fills almost all the total mesophyll of *Myrtus communis*, while the palisade parenchyma is formed by one-layer of regular palisade cells. Both one-layered epidermises are constituted by regular cells filled up of phenolic substances and covered by a lightly thickened cuticle. Internal glands border the inner sides of both epidermis, more frequently the upper one (Fig. 10a, 100x). These globose structures are delimited by two layers of cells strongly stained as well as the lumen of the glands. In front of internal glands, epidermal cells become hollow and melt with the outer layer of internal glands. Under the upper epidermis globose structures containing oxalate crystals are observable. Very numerous stomata, circular and randomly distributed, are lightly raised above the abaxial surface (Fig. 10b, 400x). Both surfaces are completely covered with differently-shaped wax structures (Fig. 10c, 1200x). Epidermal cells assume a radial organization sinking into surfaces that probably identify internal glands (Fig. 10d, 600x).

Arbutus unedo leaves are coriaceous, lanceolate, dark-green with crenate margins. The internal leaf structure shows both epidermises formed by one cellular layer with cell walls very thickened covered with a dense cuticular layer. Mesophyll is constituted by two layers of very regular palisade cells and by the spongy parenchyma characterized by wide lacunae (Fig 11a, 100x;). All tissue are filled up with phenolic substances (Fig.11b, 400x). Convex epidermal cells, irregularly shaped, form the abaxial surface, in which numerous circular stomata are sunken (Fig. 11c, 170x). Differently-shaped waxes cover both surfaces, particularly the surface of the major veins (Fig. 11d, 2400x).

Very small, linear and filiform leaves characterize *Helichrysum italicum.* The internal structure evidences very revolute margins and the main veins are very raised above surface (Fig. 12a, 100x). The palisade parenchyma is organized in two layers of elongated and packed cells filled up with starch grains (Fig.12b, 400x), the spongy tissue is composed by elongated cells surrounded by large lacunae. In both one-layered epidermises very raised stomata form big underlying stomatal chambers (Fig. 12c, 400x). Abaxial and adaxial surfaces are entirely covered by a thick layer of interwoven long, collapsed hairs hiding numerous pluricellular pedestals with large collapsed globose structures, probably oleiferous glands (Fig. 12d, 300x).

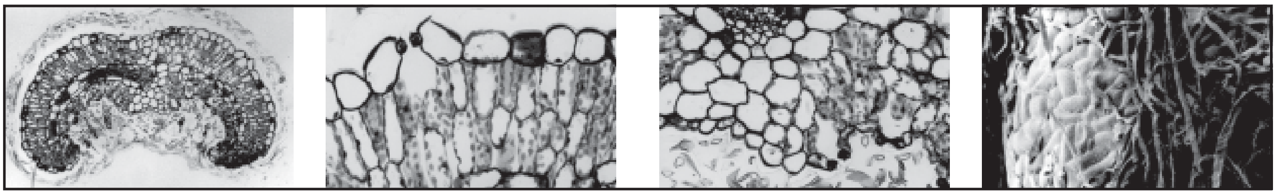

Fig. 12a - 12b - 12c - 12d.

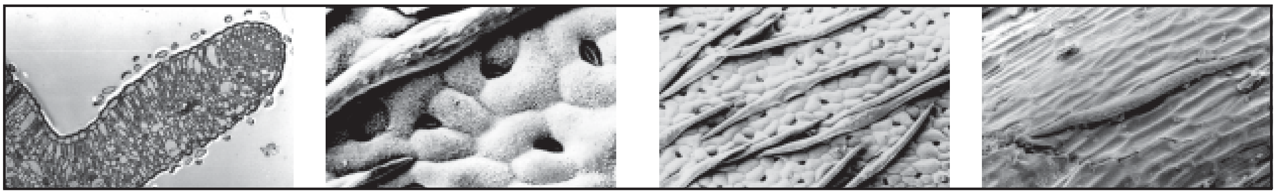

Fig. 13a - 13b - 13c - 13d.

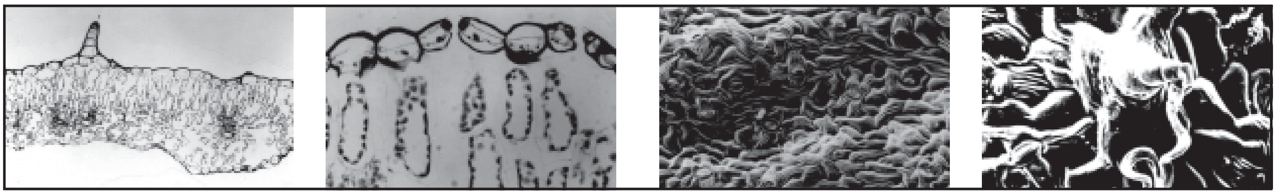

Fig. 14a - 14b - 14c - 14d.

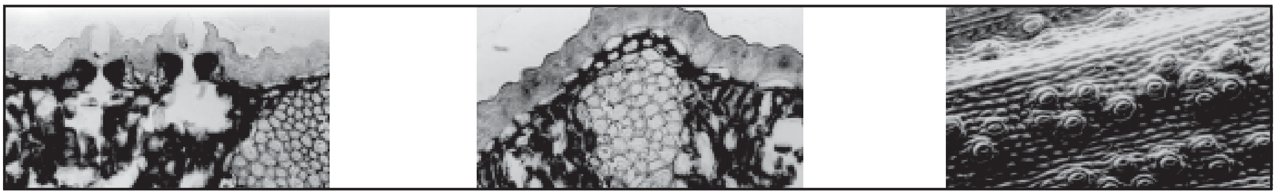

Fig. 15a - 15b - 15c.

Plants of *Astragalus massiliensis* have composed leaves constituted by 5-8 pairs of leaflets alternatively inserted on persistent rachis which becomes spinous in the growing season. The apical leaflet at the bloom stage is replaced by a thorn. Internal leaf tissues are organized in isolateral structure: two layers of palisade, one of which more developed, border with both epidermises and very narrow spongy parenchyma is situated in the medial part of mesophyll (Fig. 13a, 100x). Both epidermises are one-layered and covered by a thickened cuticle layer. In abaxial surface numerous stomata are very sunken and hidden by marked convexity of epidermal cells (Fig. 13b, 600x). Dense T-shaped hairs with equal, tip pointed, arms with verrucose surface are observed, particularly on major vein surface (Fig.13c, 170x). The upper side is very similar to the lower one with epidermal cells less convex, stomata less sunken and T-shaped hairs less dense. The surface of leaf modified in thorn shows rectangular, elongated and concave epidermal cells with T-shaped hair less developed and less numerous oriented along the longitudinal axis of the epidermal cells. Longitudinal relief with rounded stomata level with surface are evident, and the whole surface is covered by a dense wax layer organized in platelets (Fig. 13d, 600x).

Typical composed leaves, with major veins which lignifies becoming branched thorns during the season, characterize the species *Centaurea horrida*. The internal leaf architecture reveals a very heterogeneous thickness, with both epidermises constituted by one-layer of differently-sized cells with lightly thickened outer walls (Fig. 14a, 100x). Palisade and spongy tissues are organized in poorly packed cells with very thin walls. Numerous stomata, very raised above the surface, form sub-stomatic very wide chambers (Fig. 14b, 400x). Both surfaces are characterized by a puzzle of epidermal cells organized to form a surface with subsidences and reliefs at the top of which stomata are present (Fig. 14c, 300x). Sporadic circumvolute trichomes with large septate pedestal characterize both sides of the leaf (Fig. 14d, 600x).

The species *Genista sardoa* has typical trifoliate leaves, with lignify becoming thorns during the season. Here, the thorns are observed. The transverse section shows a one-layered epidermis covered by a cuticular thickening which becomes double-layered in correspondence of stomata (Fig. 15a, 400x). Presence of collenchymatic cell groups under the epidermis gives a ribbed organization. Photosynthetic tissue characterized by abundant chloroplasts is alternated to mechanical tissue (Fig. 15b, 400x). The thorn surface has epidermal cells elongated and concave, structured in longitudinal reliefs, at the basis of which are present circular stomata whose guard cells are surrounded by a double-thickened outline very raised above the surface (Fig. 15c, 170x).

Ecophysiological observations

The climatic diagram of the Arca di Noè site (Fig. 16) shows a evident alternation of two seasonal patterns: a cold

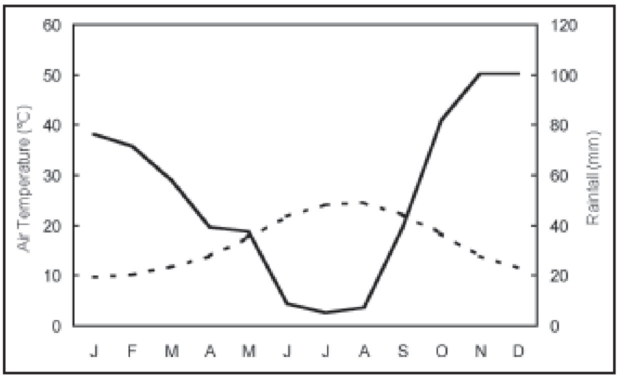

Fig. 16 - Climate diagram at the Arca di Noè site (40 years of observations). Continuous line represents the precipitations (643 mm/year). Dashed lines is the temperature (16.8 °C).

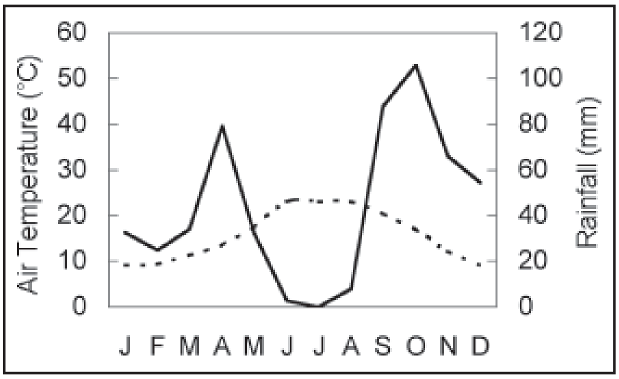

Fig. 17 - Air temperature and rainfall during 1998.

and rainy period from October to April and a hot, dry period from May to September. The mean annual value of precipitation is 643 mm, and the mean temperature 16.8°C.

The year in which these observations were carried out (1998), was characterized by winter rainfalls lower than the climatic values, leading to a annual precipitation of 578 mm, while the temperature pattern was congruent to that of the climatic analysis (Fig.17).

Active physiological processes and growth of evergreen sclerophyllous Mediterranean vegetation in Arca di Noè site take place mainly during autumn and spring when favorable environmental conditions occur. In summer, when soil moisture content decrease and atmospheric evaporative demand increases due to high solar radiation conditions, physiological activity becomes limited.

Examples of seasonal changes in the physiological activity for the four dominant species are shown in Fig 18, 19, and 20. Concurrently to the absence of rainfall, water potential values during summertime were very low, in particular for juniper and phillyrea, with values close to –5 Mpa. Lentisk and dwarf fan palm showed greater water potential values, thus indicating a better tolerance to water deficit (Fig.18).

During the summer, all the four species showed low photosynthesis. Juniper and lentisk had the higher level of leaf assimilation rate in the spring, while phillyrea and dwarf fan palm were characterized by a higher photosynthetic activity in autumn and winter (Fig 19).

Phillyrea, *Pistacia* and *Chamaerops* showed similar trends of leaf transpiration rate (E), low in the spring and progressively increasing till to the autumn, when transpiration was at its maximum value. On the contrary, *Juniperus* showed a different behavior, being E maximum during spring and decreasing in summer and autumn (Fig. 20).

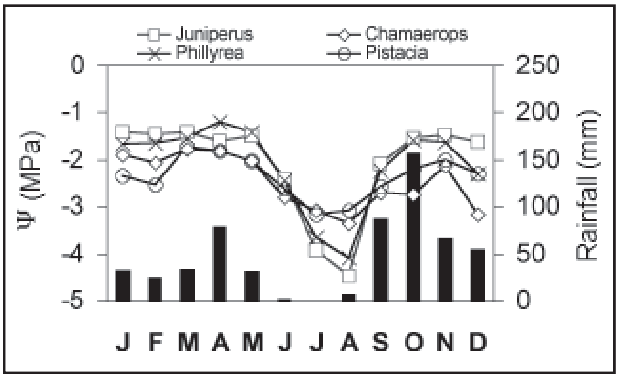

Fig. 18 - Leaf water potential of lentisk, phillyrea, dwarf fan palm and juniper and monthly precipitations during 1998.

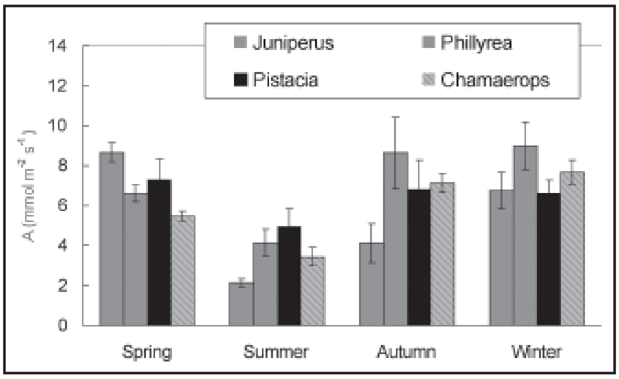

Fig. 19 - Net photosynthesis of the same species during the different season.

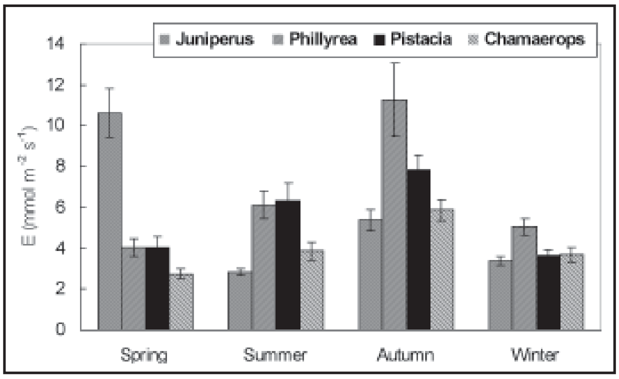

Fig. 20 - Seasonal leaf transpiration rates in the same four species.

All the species evidence a continuous, although variable, physiological activity allowing the maintenance of active growth processes also during autumn and winter, when rainfalls provide water availability and temperatures are lower.

The daily CO_2 fluxes reported in Fig. 21 also indicate that the macchia ecosystem acts as a atmospheric carbon source during the dry late summer, when drought is maximum, and early autumn, when same rain already occurred. The calculated ecosystem balance is –70 g $CO_2\ m^{-2}\ y^{-1}$, direct index of a very low efficiency of this coastal ecosystem as sink of atmospheric carbon dioxide, and indirect indicator of a delicate vitality of the ecosystem itself.

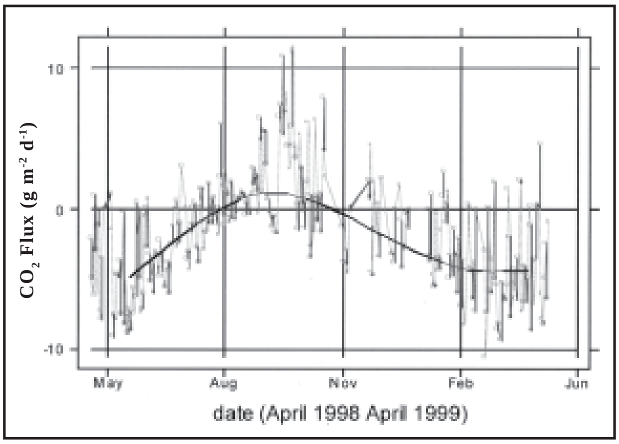

Fig. 21 - Daily CO_2 measurement at the Arca di Noè during 1998/1999. Data points and the interpolating line are shoown.

Discussion

Several studies have outlined how xeromorphic features are prominent in withstanding drought and that water deficit, high light intensity and nitrogen deficiency are the main factors causing a lower ratio of leaf surface to volume (Field & Mooney, 1986; Castro Diez et al., 1997). The reduction of leaf area exposed to the external environment leads to primary structural modifications of the leaf anatomy, such as decreased cell size, thicker walls and cuticles, a more compact network of veins, a higher stomatal frequency, and palisade strongly developed at the expense of spongy mesophylls (Shields, 1950; Gratani & Bombelli, 1999). Compact and more differentiated palisade parenchyma and generally well-developed sclerenchymatic tissue provide the leaves with a certain degree of protection against irreparable damage during severe drought (Kummerow, 1973; Stephanou & Manetas, 1997). Sclereids may also aid in water transport through the palisade (Heide-Jorgensen, 1990).

The evergreen sclerophylls (hard-leaved) seem to have in common the fact that their genetic potential allows a high degree of plasticity, producing structures and metabolic adaptations which ensure a high photosynthetic efficiency as long as water is available (Flexas et al., 2001).

The vegetation populating our Mediterranean macchia community proved to be able to survive to strong summer drought through several anatomical and physiological adaptations. The four dominant species appear in fact able to maintain a significant level of photosynthetic activity, although summer drought play the major role in limiting their growth (Spano et al., 1998, 1999; Asunis et al., 2000). The anatomical observations confirm widespread adaptive properties to couple with the high temperature and solar radiation and the lack of summer rainfall typical of the Mediterranean climate as observed (Christodoulakis & Mitrakos, 1987, Aronne & DeMicco, 2001).

Although the function of crystals in the plants is not yet completely understood (Pritchard et al., 1997), a relation with ecological processes including some plant/environment and plant/pathogen interactions has been postulated (Northup et al., 1995). A function as defensive compounds and against foraging animals have been attributed to terpenes, and tannins, and oxalates seem also to play a substantial role in the control of cellular ionic and osmotic balance, crucial in the adaptive resistance of plants to water stress (Franceschi & Horner, 1980). Many common adaptations act to increase osmotic pressure under water deficit, similarly to the accumulation of solutes due to the conversion of starch into simple carbohydrates (Shields,1950).

Because of their spatial arrangement in both vacuoles and walls of epidermal cells, the phenolic compounds can maximize their UV-B radiation-absorbing function, and most UV-B radiation can be depleted before it can reach the internal tissues, as observed for conifers from Nagel et al., (1998). Nearly all the genera observed here present such an adaptation for protection against the photodamage possibly induced from the strong UV-B solar irradiance in the Mediterranean summer. The first line of defense in this regard are the epidermis and the cuticle and their effective screening of UV-B can reduce the penetration to more sensitive mesophyll tissues in the interior of the leaves (Day et. al., 1993). *Euphorbia, Cistus, Arbutus* in particular have a thickened epidermal layer and many of the other species are characterized by thickened wall of the epidermal cells. Also, the generally high leaf tissue thickness has to be interpreted as a protective mechanism such that the penetration of UV-B radiation to lower regions of the leaf is decreased (Baldini et al., 1997).

In many of the species examined, when trichomes or wax layers reduce radiation absorbance, two or three layers of palisade parenchyma are present, presumably concurring to a better efficiency in utilizing the photosynthetic light. *Phillyrea* and *Astragalus* have palisade tissues in both sides of the leaves. The palisade, because of its structure, greatly enhances the inner free surface of the leaves, thus enabling more rapid and efficient gas exchanges (Fahn & Cutler, 1992, Kyparissis & Manetas, 1993). A strategy common to almost all the species populating the macchia is the increase of cuticular resistance to water loss: waxes, arranged in rods, platelets or crystalloids often cover both epidermises. Among the xeromorphic features is the presence of trichomes, covering the abaxial surfaces of the leaves of some of the species. *Astragalus* evidences hairs also on the upper surface of the leaves, while *Olea, Euphorbia, Cistus, Rosmarinus, Helichrysum* have dense pubescent layers giving a high refractive, silvery appearance to the lower leaf surface. The function of trichomes has been widely discussed in literature since the first papers of Haberlandt (1894). They may directly affect transpiration by influencing the diffusion boundary layer of the leaf surface and, in addition, supervise the water economy of plants through temperature regulation. Trichome layers substantially increase leaf reflectance for all the wavelengths of solar radiation between 400 and 3000 nm and restrict radiation absorbance, which results in a reduction of the heat load (Ehleringer et al., 1976; Ehleringer & Mooney, 1978, Vogelman, 1993). The papillae visible on some surfaces (*Euphorbia*) and the deeply lobed surface of *Cistus* may also be very effective in convective heat loss, as remarked by Vogel (1970).

As the majority of xerophytes, the species of the coastal macchia show in many cases stomata sunken to various depths, overarched by papillae (as in *Euphorbia*), surrounded by rings of waxes (as in *Chamaerops*), hidden by epidermal convexities (as in *Astragalus*). Stomata are protected by water losses in *Cistus*, being located inside deep crypts similarly to those descript in some species of the genus *Banksia* (Proteaceae) (Ewart, 1930) as well as in *Ceanothus gloriosus* (Kummerow, 1973). Stomata are present also on the upper leaf surface in four of the species growing in the garigue (*Helichrysum, Genista, Centaurea, Astragalus)*. The amphistomaty may have evolved in response to increasing aridity during Tertiary, as it is considered to be a highly xeromorphic trait (Parkhurst, 1978). Stomatal density appears generally high, in agreement with typical xeromorphism. High temperature and drought are two important carbohydrate-depleting stresses encountered regularly by the vegetation (Cowling, 2001). Morphological xeromorphic adaptations, as the observed small, thick, layered leaves of many of the species of the mediterranean macchia are likely to favour the carbon gain profits over transpirational losses (Turner, 1994). However, the most recurrent species appear to suffer long period of drought, and to recover their photosynthetic activities only after hot and dry summer months have gone. The same well defined seasonality with prolonged drought effects appears also from the ecosystem carbon flux that, as in other Mediterranean ecosystems, evidences that the photosynthesis following rainfall events in early fall is accompanied by enhanced soil respiration (Miglietta & Peressotti, 1999). This concomitant increase in photosynthesis and respiration leads to a ecosystem net carbon loss over several months of the year. The Mediterranean coastal macchia studied here is hence a very small sink of carbon and thus appears very sensitive to degradation, even with very small climatic changes. A decrease in precipitation around the Mediterranean could in fact easily cause substantial carbon losses from these systems, with potential positive feedbacks on the atmospheric balance of greenhouse gases (Agnew & Warren, 1996; Hulme, 1996, Le Houerou, 1996). Moreover, changes in climate may lead to severe land degradation and finally to desertification, causing an overall reduction in plant biomass and biodiversity, and the loss of many autochthon species.

Acknowledgement

This paper contributes to the EU- Environment and Climate Programme (MEDEFLU, contract ENV4-CT97-0455).

Thanks are due to Gianfranco Filippini, Mafalda Govoni and dr. Silvia Loreti for the helpful assistance in SEM and VIA analyses.

References

Agnev, C. & Warren A. 1996. A framework for takling drought and land degradation. Journal of Arid Environments 33: 309-320.

Aronne G., De Micco V. 2001. Seasonal dimorphism in the Mediterranean Cistus incanus L. subsp. Incanus. Annals of Botany 87(6): 789-794.

Asunis, C., Cesaraccio, C., Spano, D., Duce, P., Sirca, C. & Motroni A. 2000. Ecofisiologia di alcune specie della macchia mediterranea nella Sardegna centro-meridionale. Atti del II Congresso della Società Italiana di Selvicoltura ed Ecologia Forestale (SISEF): 467-472.

Baldini, E, Facini, O, Nerozzi F, Rossi F. & Rotondi, A 1997 Leaf characteristics and optical properties of different woody species Trees 12: 73-81.

Castro Diez, P., Villar Salvador, P., Perez-Rontome, C., Maestro Martinez, M.& Montserrat Marti, G. 1997. Leaf morphology and leaf chemical composition in three Quercus (Fagacee) species, along a rainfall gradient in NE Spain. Trees 11: 127-134.

Chessa, P.A., Cesari, D. & Delitala, A. 1999. Regimes of precipatation and temperature in Sardinia (Italyt) and the related synoptic sitations. Theoretical and Applied Climatology 63: 195-222.

Christodoulakis, N.S. & Mitrakos, K.A. 1987. Structural analysis of sclerophylls in eleven evergreen phanerophytes in Greece. pp 547-551. In: J.D. Tenhuen et al (eds) Plant response to stress. NATO ASI Ser. G15.

Cody, M.L. & Mooney, H.A. 1978. Convergence versus nonconvergence in Mediterranean-climate ecosystem. Annual Review of Ecology and Systematics 9: 265-321.

Cowling, S.A. 2001. Plant carbon balance, evolutionary innovation and extinction in land plants. Global Change Biology 7: 231-239.

Day, T.A., Martin, G. & Vogelman T.C. 1993 Penetration of UV-B radiation in foliage: Evidence that the epidermis behaves as a non-uniform filter. Plant Cell Environment 16: 735-741.

Delitala, A.M., Cesari, D., Chessa, P.A. & Ward, N.M., 2000. Precipation regimes over Sardinia (Italy) during the 1946-1993 and associated large-scale climatic variations. International Journal of Climatology 20: 519-542.

Duce, P., Spano, D., Cesaraccio, C. & Asunis C. 1999. Descrizione del test-site Arca di Noè. In: Monitoraggio di un ecosistema a macchia mediterranea: misure ecofisiologiche e micrometeorologiche. Quaderno n. 6 collana tecnico-scientifica Istituto Nazionale per l'Analisi e la Protezione degli Agroecosistemi. Firenze .

Ehleringe,r J., Bjorkman, O. & Mooney H.A. 1976 Leaf pubescence: effects on absorbance and photosynthesis in a desert shrub. Science 192: 376-380.

Ehleringer, J. and Mooney H.A. 1978 Leaf hairs: effect on physiological activity and adaptive value to a desert shrub Oecologia 37:183-200.

Ewart, A.J. 1930 Flora of Victoria, Univ. Press, Melbourne, Australia.

Fahn, A. & Cutler D.F. 1992 Xerophites. Gebruder Borntraeger. Berlin pp176.

Field, C. & Mooney H.A. 1986. The photosynthetic-nitrogen relationship in wild plants pp 25-55. In: Givnish T.J. (Ed.) On the economy of plant form and function. Columbia University press, Cambridge.

Flexas, J., Gulias, J., Oanasson, S., Medrano I. & Mus, M., 2001. Seasonal patterns and control of gas exchange in local populations of the Mediterranean evergreen shrub *Pistacia lentiscus*. Acta Oecologica 22: 33-43.

Franceschi, V.R. & Horner, H.T. 1980. Calcium oxalate crystals in plants. Botanical Review 46: 361-427.

Gratani L., Bombelli A. 1999. Leaf anatomy, inclination, and gas exchange relationship in evergreen sclerophyllous and drought semidecidous species. Photosynthetica 37 (4) 573-585.

Haberlandt, G. 1894 Über Bau und Funktion der Hydatoden. Ber Dt Bot Ges 12: 367-378.

Heide-Jorgensen, H.S. 1990. Xeromorphic leves of *Hakea suaveo-*

lens R.Br. IV. Ontogeny, structure and function of the sclereids. Australian Journal of Botany 38: 25-43.

Hulme, M. 1996. Recent climatic change in the world’s drylands. Geophysical Research Letters 23: 61-64.

Kyparissis A., & Manetas 1993 Seasonal leaf dimorphism in a semideciduous Mediterranean shrub: ecophysiological comparison between winter and summer leaves. Acta Oecologica-Oecologia Plantarum 14 (1): 23-32.

Kummerow, J. 1973 Comparative anatomy of sclerophylls of Mediterranean climatic areas pp 157-167. In: F. DiCastri and H.A. Mooney (eds.) Mediterranean type ecosystem. Ecological Studies.

Le Houerou, H.N. 1996 Climate change drought and desertification. Journal of Arid Environments 34: 133-185.

Miglietta F, Peressotti A., 1999. MEDEFLU: Summer Drought reduces carbon fluxes in Mediterranean “Forest” . Global Change newslette, 39:15-19.

Nagel L., Bassman, J.H., Edwards, G.E., Robberecht, R. & Franceschi, V. 1998 Leaf anatomical changes in *Populus trichocarpa*, *Quercus rubra*, *Pseudotsuga meziesii* and *Pinus ponderosa* exposed to enhanced ultraviolet-B radiation. Physiology Plantarum 104: 385-396.

Northup, R.R. , Yu, Z., Dahlgreen, R.A. & Vogt, K.A. 1995 Polyphenol control of nitrogen release from pine litter Nature 377: 227-229.

O’Brien, T.P. & McCully, M.E. 1981 The study of Plant Structure. Principle and Selected Methods. Termacarphy PTY. LTD Melbourne Australia.

Paci, M., Pellizzaro, G., Spano, D., Asunis, C., Cesaraccio C.& Sirca C. 1999. Analisi della distribuzione spaziale della vegetazione: un esempio di applicazione alla macchia mediterranea nella Penisola di Capo Caccia (Sassari), Monti e Boschi 5: 17-25.

Parkrust, D. F. 1978 The adaptative significance of stomata occurrence on one or both surfaces of leaves. Journal of Ecology 66: 367-383.

Penuelas, J., Filella, I., Llusia, L., Siscart, D. & Pinol, J. 1998 Comparative field study of spring and summer leaf gas exchange and photobiology of the mediterranean trees *Quercus ilex* and *Phillyrea latifolia*. Journal of Experimental Botany 319: 229-238

Pritchard, S., Peterson, C., Runion, G.B., Prior, S. & Rogers, H. 1997 Atmospheric CO_2 concentration, N availability and water status affect patterns of ergastic substance deposition in longleaf pine (*Pinus palustris Mill.*) foliage. Trees 11: 494-503.

Shields, L.M. 1950 Leaf xeromorphy as related to physiological and structural influences. The Botanical Review vol. XVI N.8: 306-447.

Spano, D., Duce, P., Cesaraccio, C., Francescani, A. & Asunis C. 1998. Rilievi ecofisiologici su alcune specie della macchia mediterranea, Atti IV Giornate Scientifiche S.O.I., Sanremo, 1-3 Aprile 1998: 379-380.

Spano, D., Duce, P., Cesaraccio, C., Francescani, A., Rossi, F. & Rotondi A. 1999. Rilievi ecofisiologici e caratteristiche morfo-istologiche di alcune specie della macchia mediterranea. In: Monitoraggio di un ecosistema a macchia mediterranea: misure ecofisiologiche e micrometeorologiche. Quaderno n. 6 collana tecnico-scientifica Istituto Nazionale per l’Analisi e la Protezione degli Agroecosistemi. Firenze 21-29.

Specht, R.L. 1969. A comparison of the schlerophyllous vegetation characteristic of Mediterranean type climates in France, California and Southern Australia. I. Structure, morphology and succession. Australian Journal of Botany. 17: 277-292.

Stephanou M., & Manetas Y 1997. The effect of season, exposure, enhanced UV-B radiation, and water stress on leaf epicuticular and internal UV-B absorbing capacity of Cistus creticus: a Mediterranean field study. Journal of experimental Botany 48: 1977-1985.

Turner, I.M. 1994. Scherophylly: primarly protective? Functional ecology, 8: 669-675.

Valiente-Banuet, A., Flores-Hernàndez, N., Verdù, M. & Davila P. 1998 The chaparral vegetation in Mexico under nonmediterranean climate : the convergence and Madrean Tethyan Hypotheses reconsidered. American Journal of Botany 85(10):1398-1408.

Vogel, S. 1970 Convective cooling at low air speed and the shape of broad leaves. Journal of Experimental Botany 21: 91-101.

Volgelman, T.C. 1993 Plant tissue optics. Annual Review of Plant Physiology and Plant Molecular Biology 44: 231-251.

Westerkamp, C. & Demmelmeyer, H 1997 Leaf surfaces of the Central Europen Woody Plants. Atlas and Keys. Gebruder Borntrager, Berlin Stuttgart pp A0-A25.