A. Scialabba, M. R. Melati & F. M. Raimondo

Taxonomic studies on the Sicilian elms: leaf structure of their species and hybrids

Abstract

Scialabba, A., Melati, M. R. & Raimondo, F. M.: Taxonomic studies on the Sicilian elms: leaf structure of their species and hybrids. – Bocconea 5: 493-504. 1997. – ISSN 1120-4060.

Leaf features of *Ulmus canescens, U. minor, U. glabra,* and their hybrids are described in detail. All taxa have asymmetrical leaf bases, either due to the absence of secondary veins in one basal lobe or because of inflexion of the basal leaf margin. Leaf shapes in hybrids are more or less intermediate between those of the putative parents, except for the presence of one or more distinct leaflets at the base, on the shorter side of the leaf blade, indicating a tendency of the leaf to become compound.

Introduction

Elm taxonomy has recently been the object of much discussion and revision (Ciferri & Zangheri 1947, Melville 1959, 1978, Browicz & Zieliński 1977, Heybroek 1976, 1979, Gambi & al. 1980, Richens 1955, 1976, 1977, 1980, Davis 1982, Rinallo & al. 1983, Jalas & Suominen 1986, Turland & al. 1993). Elm species are interfertile and produce hybrid swarms, which makes their delimitation very difficult.

In Sicily the genus *Ulmus* is represented by *U. minor* Mill. (= *U. carpinifolia* Gled.), *U. glabra* Huds. and, allegedly, *U. canescens* Melville (Pignatti 1982). Only *U. glabra* (Raimondo 1977) and *U. minor* have been found to grow in pure stands locally. Most of the Sicilian elm population consist of trees variously combining the characteristics of two or three taxa and cannot be identified with any one of them (Raimondo & Schicchi 1989). In Sicily, *U. glabra* and *U. minor* are at the south-western limit of their area; *U. canescens,* which was recently considered a subspecies of *U. minor* (Browicz & Zieliński 1977) and grows in Greece, Yugoslavia, S. Italy, Cyprus, Israel, and Turkey, is here at its south-western limit (Jalas & Suominen 1986). Melville (1959) reports *U. canescens* for Sicily on the basis of an unlocalized Sicilian specimen of Todaro kept at K and BM. Having examined our Sicilian specimens, sent between 1978 and 1984, he considered them to be hybrid derivatives of the three species mentioned above (Melville in sched., K and PAL), none of them being typical *U. canescens.* These facts, as well as the recurrent presence in such hybrids of atypical foliar structures, prompted us to seek to clarify the taxonomy of the Sicilian elm populations.

Elm flowers and fruits are small and simply built, offering few characters of taxonomic value. Leaves, on the contrary, are highly diversified, and the analysis of leaf features is of undisputed value in elm taxonomy.

In this study, the Sicilian species and their hybrids, *U. canescens* × *U. minor*, *U. canescens* × *U. glabra* and *U. minor* × *U. glabra* were analysed so as to determine the effect of hybridization on leaf features.

Material and methods

Short shoots of adult trees were collected in various Sicilian localities, as specified in Table 1. As no herbarium samples of *Ulmus canescens* were available, the morphological characteristics of that species were derived from the illustrations of Melville (1959), based on material from the E. Mediterranean area and, allegedly, Sicily (specimens at BM and K, not seen).

Leaf measurements were taken from the third leaf counting downward from the end of a brachyblast, and sometimes from adjacent leaves. Values given represent the average of measurements made on 10 different leaves. To observe venation pattern, whole adult leaves were fixed in FAA (formalin-acetic acid-alcohol), then cleared in 5 % sodium hydroxide and stained by fuchsin (Fuchs 1963). In some cases a squash was made after staining. Leaf tissues were observed by light microscopy.

We use the term "leaf architecture" to denote the position and shape of those elements that constitute the outward expression of leaf structure, including venation pattern, marginal configuration, and leaf shape. The terminology for leaf architecture is that of Hickey (1973). Anatomical terms are those of Esau (1965).

Results (Table 2)

Ulmus canescens. – Leaves have an average length of 3-7 cm and a mean maximum width of 2.5-4 cm. The surface is tomentose, particularly underneath. The leaf lamina is ovate-elliptic in shape, with an acute apex, and is asymmetrical at the base due to the

Table 1. Provenance of the studied material (vouchers at PAL).

	Locality (and area)	collecting date
Ulmus minor	Petralia Sottana (Madonie Mts)	July 1984
U. glabra	Pisciarotto (Madonie Mts)	August 1977
U. canescens × *U. minor*	Montaspro (Madonie Mts)	June 1978
	Fiume Torto	October 1990
	San Leonardo	May 1981
U. canescens × *U. glabra*	San Giuseppe Jato	May 1985
U. minor × *U. glabra*	Tassita (Caronia Mts)	June 1992

suppression of the basal lobe on one side. The base is semicordate on the long side and rounded to semicordate on the short side. The margin is serrate or coumpound-serrate, with serrations of two or three definite size categories (according to the classification of Hickey 1973). The petiole is 0.5-1 cm long.

Venation is pinnate, with 12-18 secondary veins per side which all, as well as their branches, end at the margin (simple craspedodromous venation).

Ulmus minor. – Leaves have an average length of 5-12 cm and a mean maximum width of 3-7 cm. The surface is scabrous on both faces. The leaf lamina is elliptic, with an acute or acuminate apex and an obtuse base, and is almost always asymmetrical at the base due to the suppression of the basal lobe on one side, or sometimes because of an inflexion of the margin of the basal lobe without corresponding suppression of secondary veins. The basal margin is straight or concave on one side. In some leaves the curvature of the margin is more accentuated than in others. The margin is serrate with straight-convex serrations in young leaves, or compound-serrate in adult leaves. The petiole is 0.7-1.8 cm long.

Venation is pinnate, with 12-15 secondary veins per side which either all, as well as their branches, end at the margin (simple craspedodromous venation), or about half of which terminate at the margin but the remainder end differently (mixed craspedodromous venation). The primary vein is moderately thick, straight. The secondary veins diverge at an acute angle. The tertiary veins form a random reticulate pattern (Fig. 1A). The quaternary veins form orthogonal meshes. The ultimate marginal venation is incomplete. Veinlets originate from tracheids which dichotomize two or three times, giving rise to an asymmetrically branched vascular skeleton. Lightly stained vein endings consist of two or more xylem elements, often with large tracheids (Fig. 1B). Areoles are imperfect, sometimes incomplete, with a random arrangement and an irregular shape. Long and short simple trichomes are present on the leaf surface and form dense clusters in the axes of the secondary veins (Fig. 1A). Druse crystals are scattered over the whole leaf surface, and prismatic crystals are found inside elongated cells, so arranged as to form strands that run parallel to the bundles. Fibres are joined to the vascular strands of all orders.

Ulmus glabra. – Leaves have an average length of 7-15 cm and a mean maximum width of 4-9 cm. The surface is slightly scabrous. The leaf lamina obovate or elliptic, with an obtuse or occasionally acuminate apex, or with an acuminate accessory tip vascularized by a secondary vein; it is symmetrical (only exceptionally asymmetrical) at the base, which is rounded or obtuse or sometimes cordate. The margin is compound-serrate, with groups of serrations of different types and sizes; it may be revolute and with simple serrations in young leaves. The petiole is frequently absent.

Venation is pinnate, with 12-18 secondary veins per side forming a simple craspedodromous venation since they all end at the margin. The moderately thick primary vein (vein width = 1.25-2 % of leaf width) is straight; the secondary veins diverge at an acute angle and are forked near the margin. The tertiary veins are thick at their point of origin (Fig. 1C) and form an orthogonal reticulate pattern. The veins of 4th and 5th order form orthogonal meshes. The marginal veins are incomplete, as the freely-ending veinlets terminate at the margin. Vein endings are usually simple and linear, though sometimes

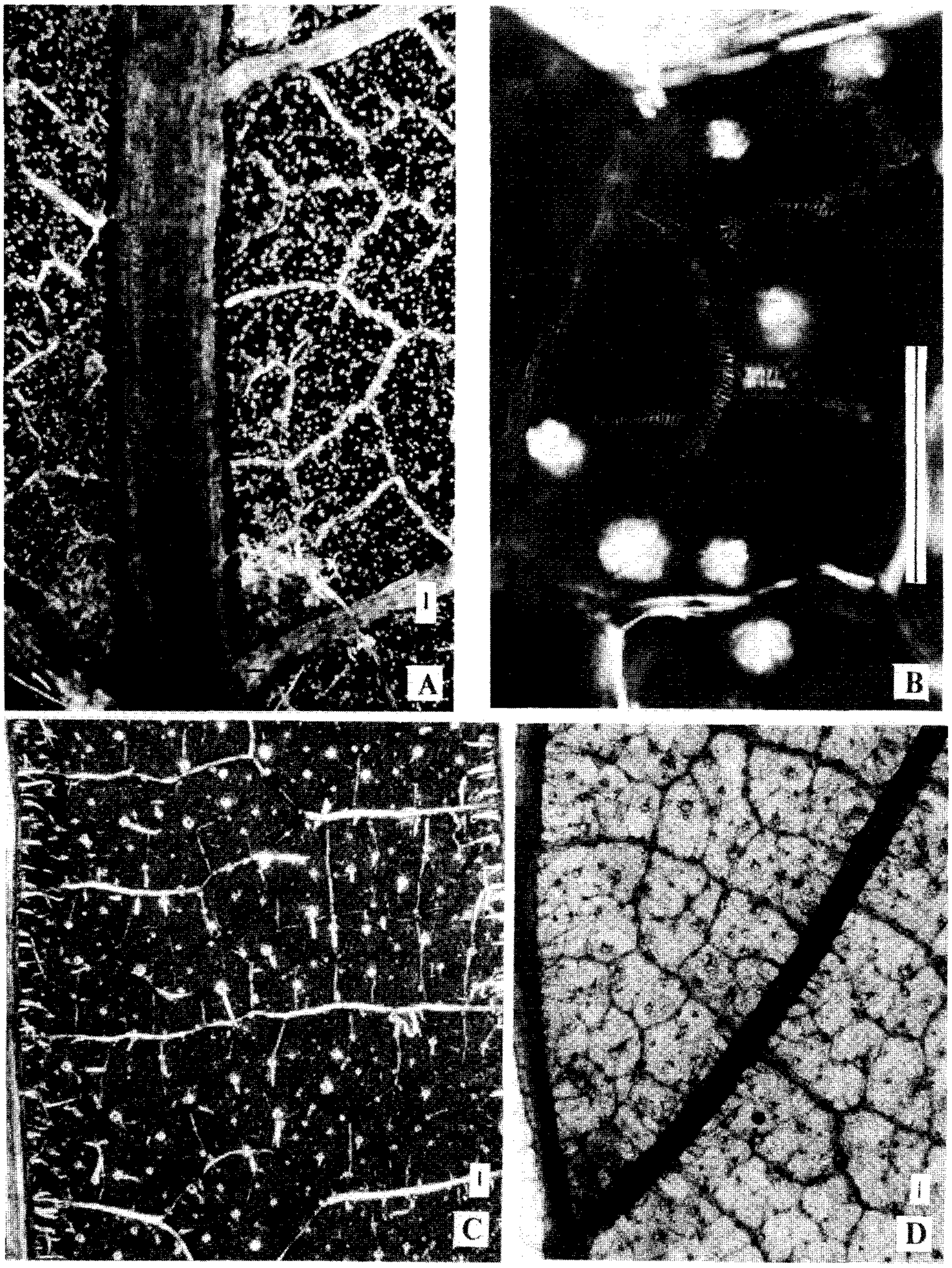

Fig. 1. Leaf architecture of Sicilian *Ulmus* species: A-B, *U. minor;* C-D, *U. glabra.* – A, C: middle portion of the leaf, showing indumentum; B, D: areoles (note druses in B). – Scale bar = 100 µm.

branched or absent. The areoles are well developed, with an orientated arrangement and a quadrangular, pentagonal or irregular shape (Fig. 1D). The trichomes, which are found both along the veins and on the leaf surface, are normally short or of medium length (Fig. 1C); trichome clusters occur in the axils of the secondary veins (Fig. 1D). Druses and a few crystals are scattered over the entire leaf surface. Fibres are present along all veins.

Ulmus canescens × *U. minor.* – Leaves have an average length of 4-7 cm and a mean maximum width of 3-4 cm. The leaf lamina is obovate-elliptic, with an acute apex and a cordate base that is asymmetrical owing to suppression of the basal secondary veins on one side, or in some cases to inflexion of the margin on the basal lobe without corresponding suppression of secondary veins. The curvature of the basal margin is concave on the short side and very convex on the long side, where the primary and secondary basal veins are connected to veins of higher order in a vascular pattern similar to that of a leaflet. Probably due to necrosis of leaf tissue, leaflets are formed on both sides of the leaf base. On the concave side, up to three leaflets can be seen, which are petiolulate in the San Leonardo population but are usually sessile in the Montaspro population. The leaf margin is serrate, of a convex-convex, straight-convex, straight-concave or rarely straight-acuminate type. In some leaves, the margin is compound-serrate, with serrations of two definite size groups (especially in the Montaspro population). The 0.5-1 cm long petiole is not noticeably thickened.

Venation is pinnate, with 12-18 secondary veins per side, simple craspedodromous (they all, as well as their branches, end at the margin). The primary vein is moderately thick and almost straight. The secondary veins diverge at an acute angle, are straight, sometimes branched toward the margin. The tertiary veins form a random reticulate pattern. The quaternary veins form orthogonal meshes. The ultimate marginal venation is incomplete, since free ending veinlets terminate directly at the margin. Veinlets are absent or branched twice, three or more times, and the xylem elements are well lignified (Fig. 2B). Some vein endings consist of two or more strands of xylem elements which at the apex form nodules (Fig. 2D) that stain red with fuchsin. The areoles are imperfect and form randomly arranged meshes of irregular shape and variable size (Fig. 2A). Both short and long trichomes (Fig. 2C) are scattered over the leaf surface and clustered in the axils the secondary veins. Druses are spread over the whole leaf surface while elongated crystals are arranged in strands running parallel to the bundles. The bundle sheath is parenchymal. Fibres are associated with veins of all orders.

The three populations examined (Montaspro, Fiume Torto, San Leonardo) are very similar. In the population of Fiume Torto the leaves are relatively small and the asymmetry is often caused by an inflexion of the margin of the basal lobe, while in the population of Montaspro the asymmetry is due to suppression of the basal lobe on one side in most of the cases. In the Montaspro population, the nodules stain only slightly with the stains used.

Ulmus canescens × *U. glabra.* – Leaves have an average length of 9-12 cm and a mean maximum width of 3-8 cm. The leaf lamina is elliptic, with an acuminate apex and an asymmetrical base that is cordate on the long side (Fig. 4) but concave on the short side due to suppression of the basal lobe and deletion of secondary veins. In some

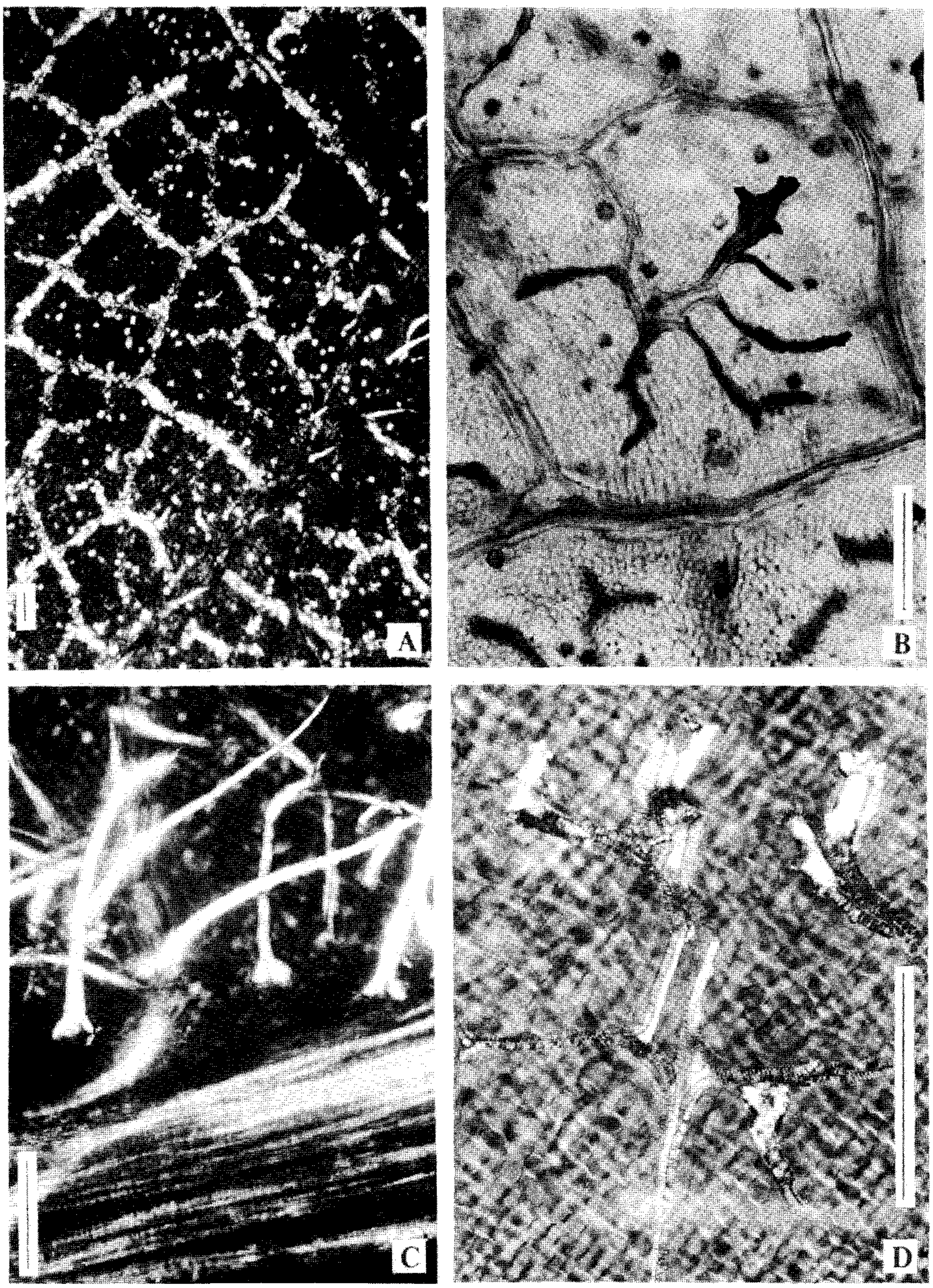

Fig. 2. Leaf architecture of *Ulmus canescens* × *U. minor*. – A, Imperfect areoles, with and without veinlets; B, areole with lignified vein endings and random branches; C, fibres accompanying the vascular strands, and trichomes; D, vein endings showing xylem elements that form nodules. – Scale bar = 100 μm.

leaves, small leaflets exist on the short side. The revolute margin is compound-serrate, with serrations of two definite size groups, sometimes simply serrate at the apex and base as well as on the leaflets. The petiole is mostly 1-1.5 cm long.

Venation is pinnate, with 10-14 secondary veins per side all terminating at the margin (simple craspedodromous venation). The primary vein is weak and straight, but its spiral xylem elements are often very thick. The secondary veins diverge at an acute angle and are sometimes branched toward the margin. The tertiary veins branch off at a right angle and form an orthogonal reticulate pattern (Fig. 3A). The quaternary veins form orthogonal meshes. Veins of the 5th order are present. The ultimate marginal veins are incomplete. The veinlets are branched once or twice and are made up of a strand of xylem elements. The areoles are well developed, with an orientated arrangement and polygonal or pentagonal shape. Short and long trichomes are present. Druses are scattered over the whole leaf surface, while crystals, inside elongated cells, are organized in strands associated with the bundles. Parenchymal bundle sheaths are present in the veinlets (Fig. 3B). The fibres run parallel to the veins of higher and lower order.

Ulmus minor × *U. glabra*. – Leaves have an average length of 9-12 cm and a mean maximum width of 4-7 cm. The leaf lamina is elliptic or elliptic-ovate, with an acuminate or obtuse apex (Fig. 4) and 16-22 secondary veins per side; it is very asymmetrical at the base due to the lack of one basal lobe. In some leaves the intercostal bands on the shorter side become narrower toward the base of the leaf, so that the number of secondary veins on the short side is the same as on the long side. One side is convex, the other very concave or straight. Leaflets are present on the short side. The compound-serrate margin bears serrations of two different size categories, sometimes replaced by convex-concave serrations. The leaflet margins have convex-convex serrations, their margin is at times revolute. The petiole is absent or very short (< 0.5 mm).

Venation is pinnate, with 16-22 veins per side, craspedodromous. The primary vein is weak (vein width < 1.25 % of leaf width) and straight. The simple secondary veins diverge at an acute angle and mostly fork toward the margin. The tertiary veins are thickened at their origin and form a random reticulate pattern, with varying angles of anastomosis, or an orthogonal pattern (Fig. 3C). The veins of 4th and 5th order form orthogonal meshes. The ultimate marginal venation is incomplete, since the free-ending veinlets reach the margin. The veinlets are branched, dichotomizing (twice or) three times. The areoles are either well developed, with meshes of relatively consistent size and shape, or imperfect with meshes of irregular shape (Fig. 3D). The xylematic skeleton consists of a few rows of xylem elements that do not stain with fuchsin or with safranin. The arrangement of the areoles is random, showing no preferred orientation, and their shape is polygonal. Short and long trichomes are found mainly along the veins, and are clustered in the axils of the secondary veins. Crystals have been observed under polarized light. The bundle sheath is parenchymal. Fibres run parallel to the veins of all orders.

Conclusions

The distribution of leaf architectural elements is known to provide a consistent set of characters that are of potential use, defining the type of venation, shape of margin, tertiary venation pattern, areole shape, marginal venation, etc. (Hickey 1971). We have paid special attention to those characters that remain relatively constant within each species or hybrid and may have ecological or taxonomic value.

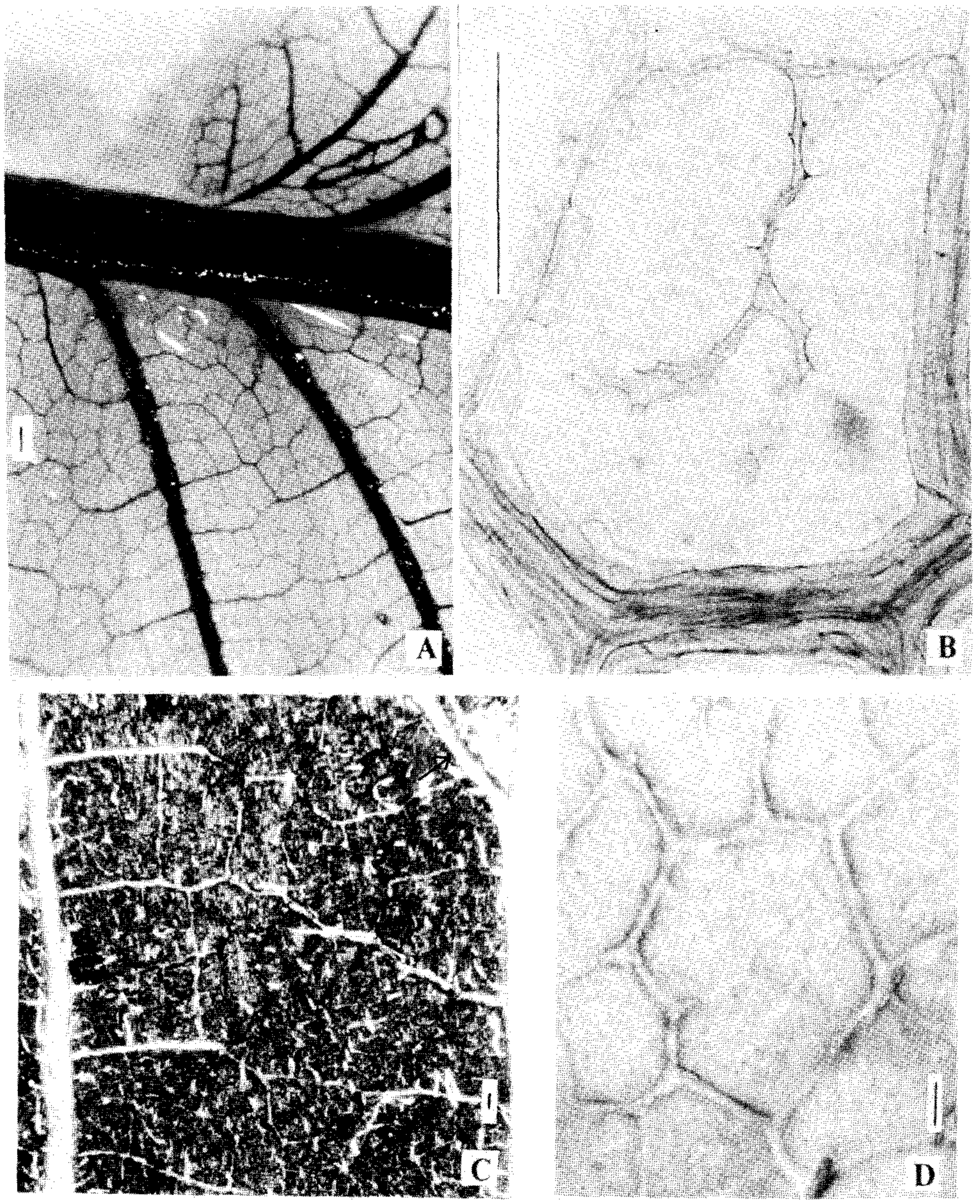

Fig. 3. Leaf architecture of Sicilian *Ulmus* hybrids: A-B, *U. canescens* × *U. glabra;* C-D, *U. minor* × *U. glabra.* – A, basal and C, middle portion of the leaf, showing areoles and indumentum; B, D, well developed areoles. – Scale bar = 100 μm.

Table 2. Summary of leaf morphological and anatomical observations on Sicilian elms. – ++, frequent; +, present; (+), rare; ±, present or absent, –; absent; ?, unknown.

Ulmus	*canescens*	*minor*	*glabra*	*can.* × *minor*	*can.* × *glabra*	*minor* × *glabra*
Petiole	+	+	–	+	+	–
Deletion of lobe	+	±	(+)	+	+	+
Leaflets	–	–	–	++	+	+
Lamina shape	elliptic-ovate	elliptic	elliptic, obovate	obovate-elliptic	elliptic	ellipt., ellipt.-ovate
Lamina apex	acute	acute/acuminate	acumin./obtuse	acute	acuminate	acumin./obtuse
Serration comp.	±	±	+	±	±	+
Tertiary reticulate	?	random	orthogonal	random	orthogonal	random, orthog.
Branched veinlets	?	+	(+)	++[1]	+	++
Fibres	?	+	+	+	+	+
Trichomes	long	short/(long)	short-medium	short/long	short/long	short-medium
Druses	?	+	++	+	+	(+)
Crystals	?	+	+	+	+	(+)

[1] With lignified nodules

As a first step, we tried to define the limits of the species morphologically. Hybrids were then identified as such by their leaf characters. It became evident that whole leaf sectors may be added or suppressed, a potentiality that sheds light upon the evolution of leaf asymmetry in the genus (Melville 1978). Leaf asymmetry may result from the lack of one or two secondary veins in one basal leaf lobe, or from the tissues of one basal lobe being undeveloped (probably due to differential growth or necrosis of tissues). During ontogenesis, the formation of secondary leaf venation starts in the basal part of the leaf and extends progressively upward. Secondary and intercostal venation together determine the venation pattern of the leaf. The areoles of *Ulmus minor* show an imperfect development and a random arrangement, the vein endings are very branched. The areoles of *U. glabra* are well developed with an orientated arrangement, the vein endings are simple or, rarely, branched.

The leaf shapes of the hybrids are more or less intermediate between those of the parents, but can be more asymmetrical than those of either parent species. The most common condition is for one parent to be partially dominant over the other. In some hybrids the basal lobe on one side of the leaf is suppressed and, in parallel, small leaflets may develop in its place that have the same vascular pattern as the leaf.

Areolation and vein endings differ in the different hybrids. Areoles of *Ulmus canescens* × *U. minor* have an imperfect development and a random arrangement. The areoles of *U. canescens* × *U. glabra* are well developed or imperfect, with an orientated arrangement. The areoles of *U.* minor × *U. glabra* are prevalently well developed, with random arrangement. The veinlets of *U. canescens* × *U. minor* are branched repeatedly and end in lignified xylem nodules, perhaps indicating an ecological adaptation. In *U. canescens* × *U. glabra* the veinlets are normally lignified, while in *U. minor* × *U. glabra* they are much branched and only slightly lignified.

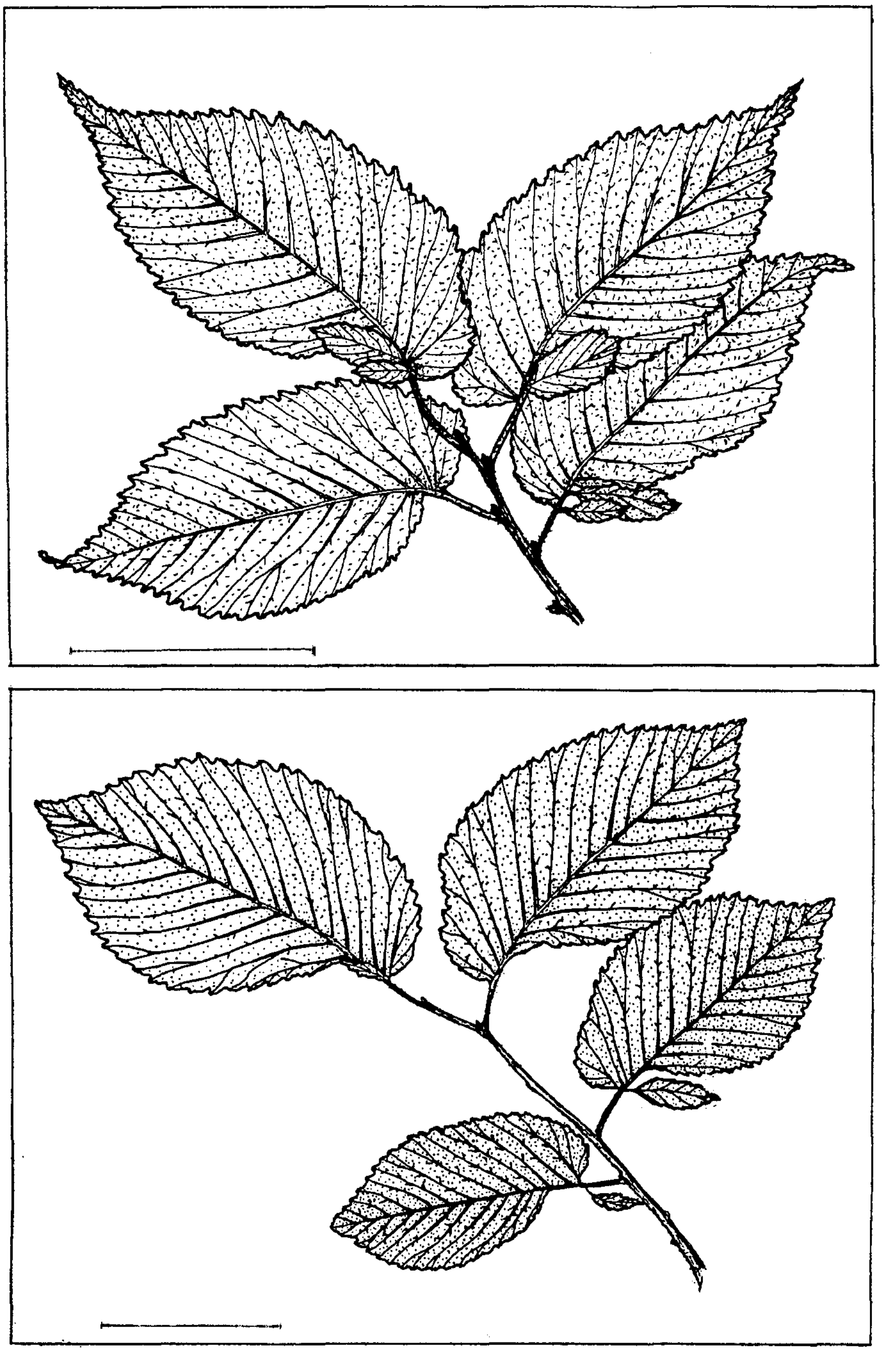

Fig. 4. Leafy long shoots of Sicilian *Ulmus* hybrids: *U. canescens* × *U. glabra* (above), *U. minor* × *U. glabra* (below). – Scale bar = 5 cm.

In almost all the elms examined, the xylem skeleton is thin and fragile because it is consists of few rows of loosely spiral or annular xylem elements. It is the fibres which run parallel to the first, second, third and in some cases fourth order veins that give support and resistance to the leaf.

Morphological trends observed by us in elm leaves include: a tendency to becoming compound, acquisition of craspedodromous secondary venation, of regularity in the course of secondary veins, of uniformity in the size and form of intercostal areas, of consistency in tertiary venation pattern, of higher order venation, and of serrate or compound-serrate margins.

The presence and distribution of crystals may be a useful character for taxonomic classification (Küster 1956). In the elms examined, the presence of crystals organized in strands and joined to the bundles, and of druses scattered over the entire leaf surface, is a constant feature. *Ulmus glabra* is particularly rich in crystal druses. Presence and distribution of hairs, often a variable feature linked with xeromorphy, may nevertheless be useful for taxonomic purposes (Metcalfe & Chalk 1950). Two types of trichomes prevail in *Ulmus*, short (except in *U. canescens*) and long (in *U. canescens* and its hybrids).

In the adult leaves of the hybrids examined, the majority of leaf characteristics are intermediate between those of the parents. In some cases, however, the interaction of the parental genes results in an additive effect (complete disappearance of one basal lobe and ramification of the basal secondary veins in *Ulmus canescens* × *U. minor*) or in obliteration of characteristics expressed in both parents, or in the manifestation of features not expressed in the parents (the appearance of leaflets at the base of the leaf). The latter observation is at first surprising, but may perhaps be explained as the result of interactions between genetically controlled growth systems. Melville (1978) observed that, in elm hybrids, the basal leaves of short shoots may be closer in shape to one parent but the distal leaves closer to the other, a fact that may help determining the parentage of individuals in a hybrid swarm.

The primordial stages of leaf formation goes through a stage of determination which confers to it a considerable degree of autonomy for its later development. This means that all of the diverse morphology of the leaves must be thought of as originating through a process of determination at a relatively undifferentiated stage (Steeves & Sussex 1989). The application of Turing's diffusion reaction theory of morphogenesis to problems of leaf ontogeny and morphology, which has previously proved fertile in accounting for hybrid leaf shapes in *Quercus* (Melville 1960) and for fenestration in the leaf of *Monstera* (Melville & Wrigley 1969), could prove useful in the explanation of the mechanisms of leaflet formation, and deletion of basal lobes, in Sicilian elm trees.

Acknowledgements

Financial support by the Consiglio Nazionale della Ricerca and the Ministero dell'Università e della Ricerca Scientifica is gratefully acknowledged.

References

Browicz, K. & Zieliński, J. 1977: Two new taxa within the *Ulmaceae* family for the flora of Bulgaria and their geographical distribution. - Fragm. Florist. Geobot. **23:** 141-150.

Ciferri, R. & Zangheri, P. 1947: Qualche dato per una revisione degli olmi italiani. – Arch. Bot. (Forlì) **7:** 67-72.

Davis, P. H. 1982: Flora of Turkey and the East Aegean Island, **7.** – Edinburgh.

Esau, K. 1965: Plant anatomy. – New York.

Fuchs, C. 1963: Fuchsin staining with NaOH clearing for lignified elements of whole plants or plant organs. – Stain Technol. **38:** 141-144.

Gambi, G., Gellini, R. & Brogi, L. 1980: Aspetti botanico-forestali del genere *Ulmus*. – Inform. Fitopatol. **1:** 27-47.

Heybroek, H. M. 1976 : Sistematiek en nomenclatuur van het geslacht *Ulmus*. - Meded. Rijksinst. Onderz. Bos- Landschapsbouw "De Dorschkamp" **8:** 237-240.

– 1979: Elms in agroforestry. – Wageningen.

Hickey, L. J. 1971: Leaf architectural classification of the angiosperms. – Amer. J. Bot. **58:** 450.

– 1973: Classification of the architecture of dicotyledonous leaves. – Amer. J. Bot. **60:** 17-33.

Jalas, J. & Suominen, J. 1986: Atlas florae europaeae, **3.** – Helsinki.

Küster, E. 1956: Die Pflanzenzelle, ed. 3. – Jena.

Melville, R. 1959: *Ulmus canescens:* An eastern Mediterranean elm. – Kew Bull. **12:** 499-502.

– 1960: A metrical study of leaf-shape in hybrids II. Some hybrid oaks and their bearing on Turing's theory of morphogenesis. – Kew Bull. **14:** 161-177.

– 1978: On discrimination of species in hybrid swarms with special reference to *Ulmus* and the nomenclature of *U. minor* Mill. and *U. carpinifolia* Gled. – Taxon **27:** 345-351.

– & Wrigley, F. A. 1969: Fenestration in the leaves of *Monstera* and its bearing on the morphogenesis and colour pattern of leaves. – Bot. J. Linn. Soc. **62:** 1-16.

Metcalfe, C. R. & Chalk, L. 1950: Anatomy of the dicotyledons, **1-2.** – Oxford.

Pignatti, S. 1982: Flora d'Italia, **1.** – Bologna.

Raimondo, F. M. 1977: Primo rinvenimento di *Ulmus glabra* Hudson sulle Madonie, Sicilia settentrionale. – Webbia **31:** 261-277.

– & Schicchi, R. 1989: Note sulla flora forestale di Sicilia: variabilità in *Ulmus* L. – Giorn. Bot. Ital. **123,** Suppl. **1:** 124.

Richens, R. H. 1955: Studies on *Ulmus*. 1° The range of variations of east Anglian elms. – Watsonia **3:** 138-154.

– 1976: Variation, cytogenetics and breeding of the European field elms (*Ulmus minor* Miller sensu latissimo = *U. carpinifolia* G. Suckow). – Anali Šumarstvo **7:** 107-133.

– 1977: New designation in *Ulmus minor* Miller. – Taxon **26:** 583-584.

– 1980: On fine distinctions in *Ulmus* L. – Taxon **29:** 305-320.

Rinallo, C., Grossoni, P., Gellini, R. & Brogi, L. 1983: Contributo alla revisione sistematica del genere *Ulmus* in Italia. – Ann. Accad. Ital. Sci. Forest. **32:** 1-27.

Steeves, T. A. & Sussex, I. M. 1989: Pattern in plant development. – Cambridge.

Turland, N. J., Chilton, L. & Press, J. R. 1993: Flora of the Cretan area. – London.

Address of the authors:
Prof. Anna Scialabba, Prof. Maria Rita Melati & Prof. Francesco M. Raimondo, Dipartimento di Scienze botaniche dell'Università, Via Archirafi 38, I-90123 Palermo, Italy.