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Oleaceae Pollinosis: A Review

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Key Words

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Abstract

In the Oleaceae family, the most allergenic pollen is produced by *Olea europaea*, the olive tree, which in the Mediterranean area has been recognized as one of the most important cause of seasonal respiratory allergy. The olive pollination season lasts from May to the end of June and sometimes causes severe symptoms (ocularrhinitis and/or bronchial asthma). Olive pollinosis is quite rare in the form of monosensitization. Although adults are affected the most, olive sensitization can be recognized in children, too. However, it is not easy to be sure about the clinical relevance of allergic sensitization to olive, even in patients with positivity to this pollen, as results from allergological tests in vivo and/or in vitro, because positivity to *Olea* pollen is not equivalent to clinical responsibility, above all in patients without seasonal respiratory symptomatology. Studies on the allergenic content of *O. europaea* pollen are currently in progress. In Northern and Central Europe, where there are no olive trees, there are two commonly occurring genera of the Oleaceae family, namely *Fraxinus* and *Ligustrum*, but these have a low frequency of allergic sensitization. A fourth anemophilous member of the Oleaceae, *Phillyrea*, has a more regional distribution in some parts of the Mediterranean. Other nonanemophilous and usually not allergenic Oleaceae genera are *Forsythia*, *Jasminum* and *Syringa*.

The Oleaceae family includes at least 400 species of woody, arboreal or shrubby, evergreen, deciduous or also lianose plants.

They can be spontaneous (*Olea europaea*, variety Oleaster, *Fraxinus*, *Phillyrea*, etc.) or cultivated for ornamental, officinal or food purposes (privet, lilac, jasmine, olive). Among various genera other than *Olea*, we find *Forsythia*, *Jasminum*, and *Syringa*. The flowers vary according to the genus, they are often curtailed, but sometimes they are numerous and reunited in bunches.

The olive (*Olea europaea*) is a long-lived tree which has been grown by man for more than 5,000 years for its fruit, oil and wood [1]. It probably originally came from Asia mi-

nor and then spread across the whole Mediterranean basin, either cultivated or wild (Oleaster) [2–6].

About four centuries ago the *Olea* was introduced into the Americas (in North America prevalently in California [7] and Arizona [8]; in South America mainly in Chile). Besides, the olive is cultivated in Australia and South Africa too [1]. A wild form of olive is referred to as 'Oleaster', but there are other arboreal forms that vary according to the country in which they grow; for instance, in Spain there are now about ten varieties [9] and many more are to be found in Italy [10]. Still now, it is not clear whether all these varieties of olive trees are able to produce different allergens. In Italy varieties of *Olea*, which are different in the northern

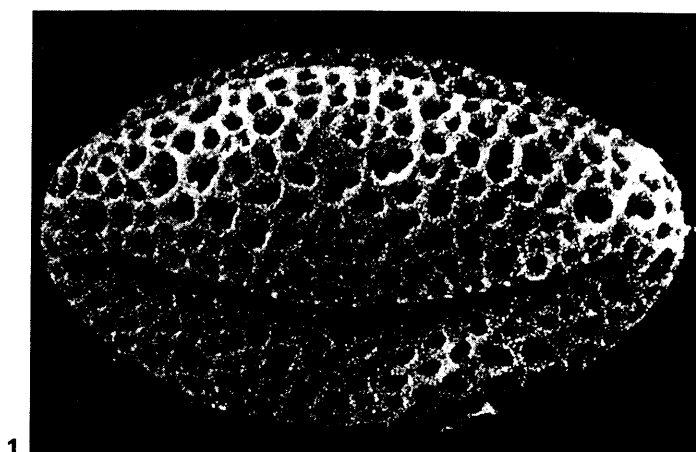


Fig. 1. *O. europaea* pollen. SEM. $\times 2,300$.

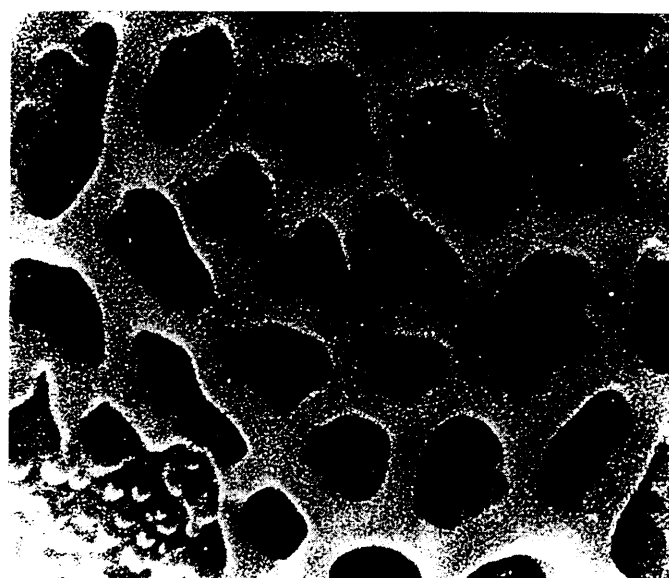


Fig. 2. Detail of exine of *O. europaea* pollen. Exine of medium thickness, reticulum with wide meshes. SEM. $\times 7,500$.

and southern parts of the country, can induce different IgE-mediated reactions [11].

As concerns the genus *Ligustrum*, the *Ligustrum vulgare* species (common privet) is a shrub about 2–3 m high, growing in light woods of lowlands and hills. It has lance-shaped leaves and small white sweet-smelling flowers united in a thick panicle. *Ligustrum ovalifolium* is instead a very popular garden-separating hedge. The *Fraxinus ornatus* tree (manna ash, flowering ash) grows spontaneously in woods from the sclerophylles evergreen Mediterranean zones to those submountains of oaks and chestnut. In some areas the *Fraxinus* is cultivated.

Aerobiology

O. europaea pollen is small (mean diameter 17–21 μm), spheroidal, isopolar and tricolporate; the equatorial contour is subtriangular while the meridian one is circular or lightly elliptic (fig. 1). The exine is about 3 μm thick and shows a reticulation (fig. 2) with dense cocumelles. The intine is thin, making onci at the opening level.

The pollination of the Oleaceae in Europe lasts from late January to early April according to latitude and local climatic conditions, which are moreover variable from year to year.

The pollination season in Italy varies from one region to another. In Campania and Apulia (Southern Italy) it lasts from early April to late June with peaks from late May to the first 2 weeks of June [12] (fig. 3). In Umbria (Central Italy)

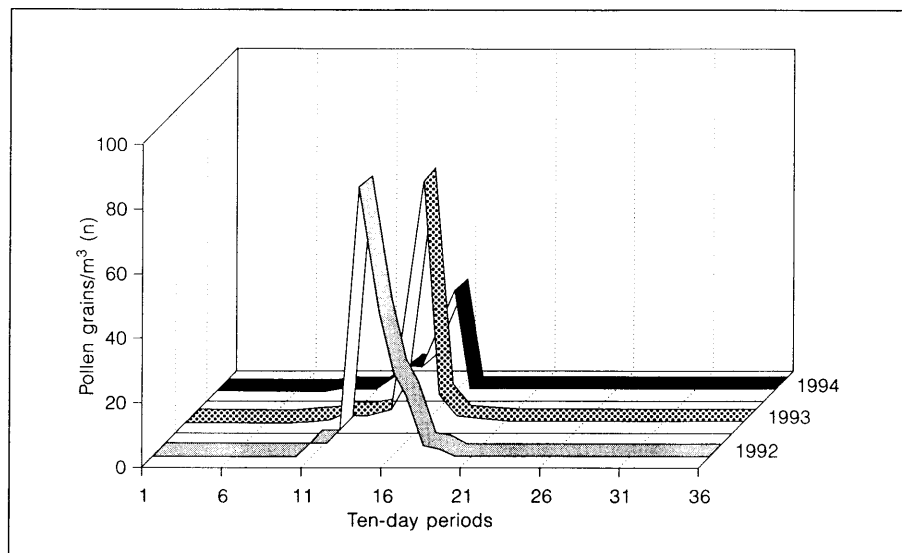
and Liguria (Northern Italy) it lasts until July [13]. However, the pollination period of the *Olea* is not very long, lasting about 1 month.

A remarkable variety of environmental factors can affect the pollen productivity of the olive plant.

Among the main external variables are light, temperature, atmospheric pressure, wind direction and speed, availability of water and minerals [14, 15]. In particular, the anthers of the *Olea* open in favorable weather, above all when it is warm and dry. They tend to open during daylight hours, from early morning to late afternoon. The correlation between the course of temperature during the months preceding the flowering of the tree and the beginning of pollination has also been studied [16]. A lower mean temperature delays the start of pollination while a higher temperature hastens it. A precocious start of the pollination season is frequently characterized by a high pollen productivity. This trend, coupled with the records of mean temperatures of months preceding the flowering season, could represent, if confirmed, a means to forecast the start and severity of the pollen season of the *Olea*. An agronomic phenomenon typical of the *Olea*, as well as a number of other trees, is the rotation of productivity with the succession of seasons with higher and others with lower pollen production [17–19].

The highest atmospheric pollen concentration varies between 100 and 200 grains/ m^3 air, which is the most frequent level in the Mediterranean area [12] and 2,000–3,000 in some areas such as Apulia, where the olive is intensely cultivated [16]. The pollination of the *Olea* is entomophilous but also anemophilous when pollen production is abundant.

Fig. 3. Airborne concentrations of Oleaceae pollens in Naples during 1992–1994. Results are expressed as 10-day running means based on daily observations.



Pollen of *Ligustrum* (mean diameter 25 μm) is subellipsoid with a circular or subtriangular equatorial contour and an elliptic meridian contour (fig. 4). Pollen grains are tricolporate with large furrows and pointed extremities. The exine is very thick, with a reticulation provided with apertures on a level with furrows. As to reticulation, the high and thick cucumellas exhibit a characteristic pattern (fig. 5).

The intine is thickened under the apertures. Pollination of the *Ligustrum* is mainly entomophilous and consequently the atmospheric concentration of pollen grains per cubic meter of air is low (usually $<50/\text{m}^3$), sometimes causing hay fever symptoms only under conditions of local exposure (e.g. when cutting *Ligustrum* hedges) [12].

Pollen of the *Fraxinus* is small (from 15 to 21 μm mean diameter) (fig. 6) with a reticulated exine (fig. 7).

Pollination is mainly anemophilous; starting in April or May in Central Europe [20] and in late March or in the first 2 weeks of April in Southern Italy.

Atmospheric concentration varies through the year, usually reaching only 10–20 pollen grains per m^3 of air [21].

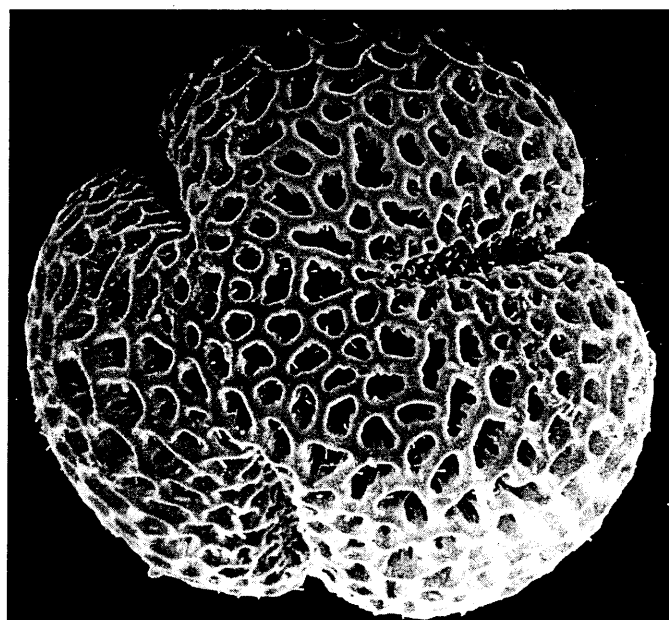


Fig. 4. *L. ovalifolium* pollen. SEM. $\times 1,000$.

Allergens

Studies on the allergenic content of *O. europaea* pollen are currently in progress. Early studies on the allergen characterization of *Olea* pollen are those by Blanca et al. [22], who found 20 fractions with allergenic activity using IEF, RAST and RAST inhibition. The major allergenic protein that was found at pI 6 through the use of SDS-PAGE was

constituted by two subunits of 15 and 17 kD with a MW of about 32,000 kD.

Using gel cromatography, Vela et al. [23] identified and described 10 different antigens and a major allergenic content of about 65,000 kD, probably made up of aggregated basic and acid proteins.

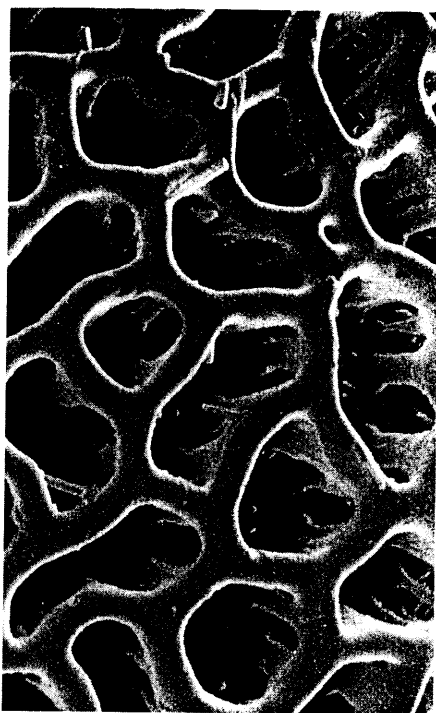


Fig. 5. Detail of exine of *L. ovalifolium* pollen. Exine thick (about 2.1 μm). Ectexine about four times as thick as endexine. Surface reticulate. SEM. $\times 7,500$.

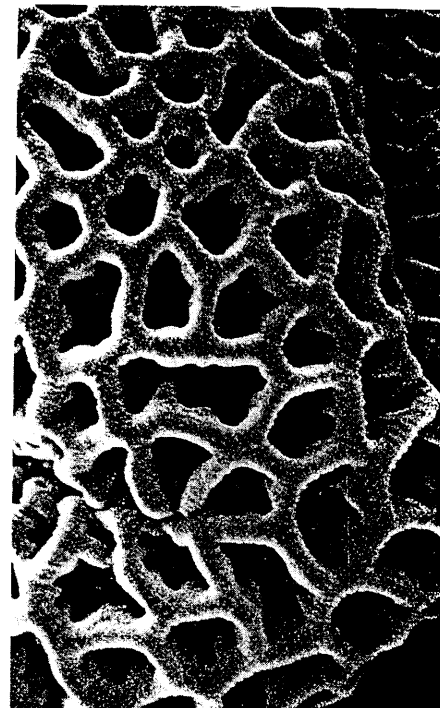


Fig. 7. Detail of exine of *F. excelsior* pollen. Detail of exine (thickness about 1 μm). Sexine thicker than nexine. SEM. $\times 4,000$.



Fig. 6. *F. excelsior* pollen. SEM. $\times 2,500$.

Likewise, studies by the Lahoz group showed in olive pollen raw extract a major allergenic glycoprotein of about 60,000 kD consisting of different subunits of minor MW [24]. Subsequently, several other authors [25–28, 31–33, 35] identified the major allergens of *Olea* pollen as proteins of 17, 19 and 42 kD separated by SDS-PAGE, which were able to bind specific IgE in high percentage when put together with serum of atopic subjects with *Olea* sensitization. A monoclonal antibody obtained from the immunization of mice with whole *Olea* extract was able to react with proteic fractions of 17 and 19 kD separated by SDS-PAGE.

Studies of Villalba et al. [30] have shown the results of fractionization of allergenic proteins of 19 kD until amino acid analysis showing that *Ole e* I is characterized by glycosylated proteins of 18 and 20.5 kD with similar epitopes and different isoallergenic shapes.

Barber et al. [31] showed that allergenic proteins of *O. europaea* are highly variable, probably as a result of physical factors affecting the plant's maturation [31]. These authors identified the major allergenic activity in one multimeric unit of 55,000 kD with a subunit of 20,000 kD. Also free proteins were found of MW 20,000 kD with arcs of

RAST inhibition like those of multimeric unit. On the basis of such results we can surmise that the allergenic activity of *Olea* pollen is probably associated with a protein family of polymers with a high degree of homology [31].

Lombardero et al. [32] showed that monoclonal antibodies OL-2 and OL-7 are able to identify two different epitopes of *Ole e* I. These results are in agreement with those obtained by Villalba et al. [30] who identified two fractions of about 18 and 20 kD by SDS-PAGE.

Monoclonal antibodies were also used to study the effects of major allergen depletion on the biological capacity of an extract. It was shown that *Ole e* I constitutes almost the totality of the biological capacity of a whole extract and the nearly total disappearance of the biological activity of an allergenic extract is due to its removal. Only 10% of atopic subjects with allergic sensitization to olive are in fact not allergic to *Ole e* I [32].

A similar experiment with a *Dermatophagoides pteronyssinus* allergenic extract showed that the depletion of major allergen *Der p* I results in the reduction of only 30% of its biological activity, because of the presence of other major allergens such as *Der p* II [33].

Moreover, De Cesare et al. [34] showed that the 19,000 MW component of *Ole e* I represents the glycosylated form of the 17,000 component.

Recently, the major allergen of *O. europaea* pollen as been amplified, cloned and sequenced with cDNA technology by Villalba et al. [35] and Lombardero et al. [36]. The amino acid sequence of the *Ole e* I was deduced; the whole protein contains 145 amino acids with a calculated molecular weight for the main sequence of 16,272 daltons. Many nucleotide substitutions were found by both groups.

Villalba et al. [37] found six cysteine residues determining three disulfide bridges which may explain the demonstrated stability of the molecule [38].

Studies of Lahoz et al. [39] analyzed the epitope mapping of *Ole e* I and other allergens of Oleaceae pollens by using monoclonal antibodies against *Ole e* I and demonstrated that the major allergen of *O. europaea* has at least four epitopic determinants with the possibility of an overlap of these epitopes.

Cross-Reactivity

Bousquet et al. [40] observed a high degree of cross-reactivity among allergens of *Olea*, *Ligustrum* and *Phillyrea*.

The results are consistent with those of other authors [41]. In a group of 103 atopic subjects living in Michigan, Kernermann et al. [42] observed an allergenic cross-reactivity

among extracts of olive, *Fraxinus*, privet and Russian olive. Even though the *O. europaea* does not grow in that geographic area, 19 subjects presented a skin test positivity to the allergens of olive pollen. The results suggest that a skin test positivity to this allergen could be also stimulated by another cross-reactive allergen.

Baldo et al. [43], examining allergenic components of *Ligustrum* pollen, showed that in the context of 15 allergens the most frequent binding with specific IgE is associated with components of MW ~19, ~20, ~40, ~70 kD. These components are therefore very similar to those of the *Olea*. An allergenic cross-reactivity between extracts of Oleaceae and some species of the Poaceae family was shown, too [44, 45].

Obispo et al. [46] showed that the main allergen of *O. europaea* (*Ole e* I) is also present in other species of the Oleaceae family (*Fraxinus excelsior*, *Ligustrum vulgare* and *Syringa vulgaris*). Similar results were obtained by Batanero et al. [47], who demonstrated 85.5–89.6% identity between *Ole e* I and the major allergen of *Syringa vulgaris* (*Syr v* I). On the contrary, the studies of inhibition performed by Cardaba et al. [48, 49] showed some differences in the recognition pattern and reactivity of the anti-*Ole e* I mAbs toward the allergens of other pollens of the Oleaceae family. The authors concluded that the cross-reactivity among these pollens is due to a common well-defined epitope recognized by anti-*Ole e* I mAbs.

Genetics

Lahoz's group studied the relationship between HLA class II haplotypes and alleles and the IgE antibody response to *Ole e* I in patients with *Olea* pollinosis [50, 51]. They found an increase of the frequency of phenotype DR7 and DQ2, indicating an association between the DRB1* 0701/2, DQB1* 0201 alleles and the IgE antibody response to *Ole e* I.

On the contrary, DR4 frequency correlates to that of the non-allergic control group, suggesting the possibility that the HLA-DR4 antigen or the haplotypes may have a protective role.

Clinical Aspects

Ole e I pollinosis is clinically characterized by oculorhinitis and/or bronchial asthma with a changing frequency, since olive forming shows variation in the Mediterranean area [52] (table 1).

In Southern France Bousquet et al. [53] found a 15% frequency of skin positivity to Oleaceae on atopic patients.

In Greece, this frequency is very high (37.2%) [54]. In Spain and Israel, various authors observed the sensitizing properties of *Olea* allergens [55, 56]. In Italy there are several studies of epidemiologic and clinical interest on the frequency of positivity in diagnostic tests to *Olea* pollen in various regions.

In some areas of Southern Italy frequencies of 30% have been found in Apulia [19], 12.22% in Sicily [57], and 16.80% in Latium [58].

All such studies showed a higher prevalence of rhinoconjunctival symptomatology in comparison with bronchial asthma. Moreover, the polysensitized subjects are more frequent than those with monosensitization to olive pollen.

In a group of 4,142 patients living in the Naples area and examined consecutively in our center from January 1, 1988 to December 30, 1989, we found a frequency of skin prick test positivity to *Olea* pollen allergens of 13.49% in adults and 8.53% in children, considering all skin-positive patients [59].

The prevailing clinical manifestations in both groups of patients (adults and children) are the association of rhinitis and bronchial asthma followed by rhinitis and bronchial asthma as isolated manifestations.

In our patients allergic sensitization to olive rarely occurs isolated (1.33% in children and 2.28% in adults). We find it more frequently associated with sensitization to other allergens, above all pollens [59].

However, we can assert that, after *Parietaria* and *Gramineae*, *O. europaea* is the third agent of pollinosis in the Mediterranean area [60–65].

Table 1. Percent of skin test positivity to *O. europaea* pollen allergens in subjects from the Mediterranean area with suspected atopic respiratory allergy

Country	%	Ref.
Southern France	15.2	53
Greece	37.2	54
Spain	36.3	56
Portugal	17.0	72
Italy		
Apulia	30.0	19
Sicily	12.2	57
Latium	18.5	58
Campania (Naples)	13.4	59

A special pattern of behavior of patients (either adults or children) with monosensitization to olive and living in the Naples area is characterized by symptoms that are frequently present throughout the year without an apparent increase during the olive pollination season [66]. This special aspect of our patients was recently described in Spanish patients too [38, 67].

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