# Preliminary Pollen Study of the Oleaceae in Malesia

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## Abstract

Preliminary work on pollen of the Oleaceae shows that it is of limited diagnostic value at the generic level. Small pollen grains (10-18$\mu$ diameter) are recorded for Malesian species of *Chionanthus, Myxopyrum* and *Olea.* The size of pollen grain is associated with style length.

## 1. Pollen morphology

The pollen morphology of the Oleaceae is reticulate and tricolporate (Wodehouse, 1965; Erdtman, 1971). A preliminary survey of Malesian species using JEOL 35 SEM, where the pollen grains were acetolysed before coating with gold, confirms this and also shows that there is variation between species for characters of the muri (whether beaded or smooth, high or low) and for the size of the lacunae (Table 1). The large lacunae are conspicuous under light microscopy in several species of *Jasminum* and in *Nyctanthes arbor-tristis* (personal observation e.g., and in *Ligustrum* (Wodehouse, *ibid).*

Some characters are constant for a genus e.g., all the species of *Olea* examined have large lacunae with spinules inside (Plate 1f); the muri of *Myxopyrum* are smooth (Plate 1d & e) and the muri of *Chionanthus* are high (Plate 1a & b). However, since only a single character is constant, a few species of different genera share the same characters, e.g. *Ligustrum confusum* and the pollen of male flowers of *Olea decussata* and the large grains of *Myxopyrum ovatum* all have large lacunae with high, smooth muri (Table 1). Pollen morphology cannot therefore be used infallibly to identify oleaceous pollen to genus. However, from this preliminary study certain character combinations do indicate generic identity e.g., small lacunae with low, smooth muri is characteristic only of *Myxopyrum.* A larger sample is needed to confirm this.

Variation of these characters may also occur within species, for example pollen from the male flower of *Olea decussata* has smooth, not beaded, muri, the large pollen grains of *Myxopyrum ovatum* have high muri instead of low (Plate 1e), and different samples of *Chionanthus ramiflorus* differ in the size of the lacunae and whether the muri are beaded or smooth (Table 1).

In addition, for the Oleaceae, pollen morphology also has limited value for identification at the family level as reported by Erdtman (1971): that several other families, such as Caprifoliaceae *(Viburnum)* and some species of the Celastraceae have very similar pollen to that of the Oleaceae. As such, pollen morphology is of limited taxonomic value above the species level in the Oleaceae.

Wodehouse (1965) suggested that the low muri of pollen grains of *Fraxinus,* compared with the other genera he examined, was related to its pollination by wind. *Myxopyrum* shows this same character but is very unlikely to be pollinated by wind as the anthers are almost sessile and are included within the corolla tube.

Table 1. Morphological characters of pollen of some Malesian Oleaceae

| Species | Muri | | | | Lacunae | |
|---|---|---|---|---|---|---|
| | + *beaded* | − *smooth* | + *high* | − *low* | + *large* | − *small* |
| *Olea brachiata* | + | | + | | + | |
| *O. paniculata* | + | | + | | + | |
| *O. decussata* (male flowers) | − | | + | | + | |
| (bisexual flowers) | − | | − | | + | |
| *Chionanthus enerve* | + | | + | | + | |
| *C. ramiflorus* | + & − | | + | | + & − | |
| *C. pluriflorus* | − | | + | | − | |
| *Ligustrum confusum* | − | | + | | + | |
| *Myxopyrum coriaceum* | − | | − | | − | |
| *M. ovatum* (small grain) | − | | − | | − | |
| (large grain) | − | | + | | + | |

## 2. Size of Pollen

Wodehouse (1965) recorded the range of pollen grain diameter in the Oleaceae as between 19.5 and 30 $\mu$ and Erdtman (1971) gave the range of the longest axis as between 20 and 63 $\mu$. These both fall within the medium-size class of pollen grains. Wodehouse *(ibid)* found that most angiosperm pollen grains fall within the 20 to 40 $\mu$ diameter range.

This study shows that while most of the genera of the Oleaceae do fall within this range, the pollen of *Chionanthus, Myxopyrum* and some species of *Olea* is much smaller, between 10 and 17 $\mu$ diameter (Table 2) i.e., fall within the small-size class of Wodehouse (1965) and Erdtman (1971).

Muller (1979) discussed possible causes for differences in size of pollen grains and reported cases where pollen size correlated with flower size, style length, anther length and latitude or altitude.

For the Oleaceae, the relationship is conspicuously between style length and pollen size (Table 2). Genera with sessile stigmas or styles up to 1 mm long produce pollen in the small-class size: *Chionanthus* (10-17.5 $\mu$ diameter), *Myxopyrum* (13-16 $\mu$) and some *Olea* species (12-15 $\mu$). Genera with styles 1 mm or more have in general pollen grains larger than 20 $\mu$. The exception is *Osmanthus scortechinii* with style length between 1 and 4 mm but with pollen grains 17.5-22.5 $\mu$ diameter.

Baker & Baker (1979) also found that for 147 species with style lengths of 10 mm or longer they investigated, the mean pollen diameter was significantly larger than that of their shorter-styled counterparts.

The relation between pollen size and style length is also found in heterostylous species. Ganders (1979) recorded that in 50 out of 55 heterostylous genera, the thrum anthers produce larger pollen than the pin anthers. (It is the thrum pollen that has to grow down the longer pin style). It was first suggested by Darwin (1877, cited by Ganders, 1979) that pollen grains that need to produce long pollen tubes to grow down long styles would need greater food reserves and would therefore be larger.

The relation between flower size (corolla length) and pollen grain diameter is less obvious and flowers longer than 10 mm *(Jasminum, Forsythia, Nyctanthes* and *Syringa)* also have longer styles. For *Chionanthus,* where all species have sessile or short stigmas, there is a considerable difference in corolla length between the temperate species, *C. retusus* and *C. virginicus* with flowers more than 10 mm long, and the tropical species with small flowers less than 5 mm long, but the range in pollen size overlaps between these species with large and small flowers (Table 2). In addition,

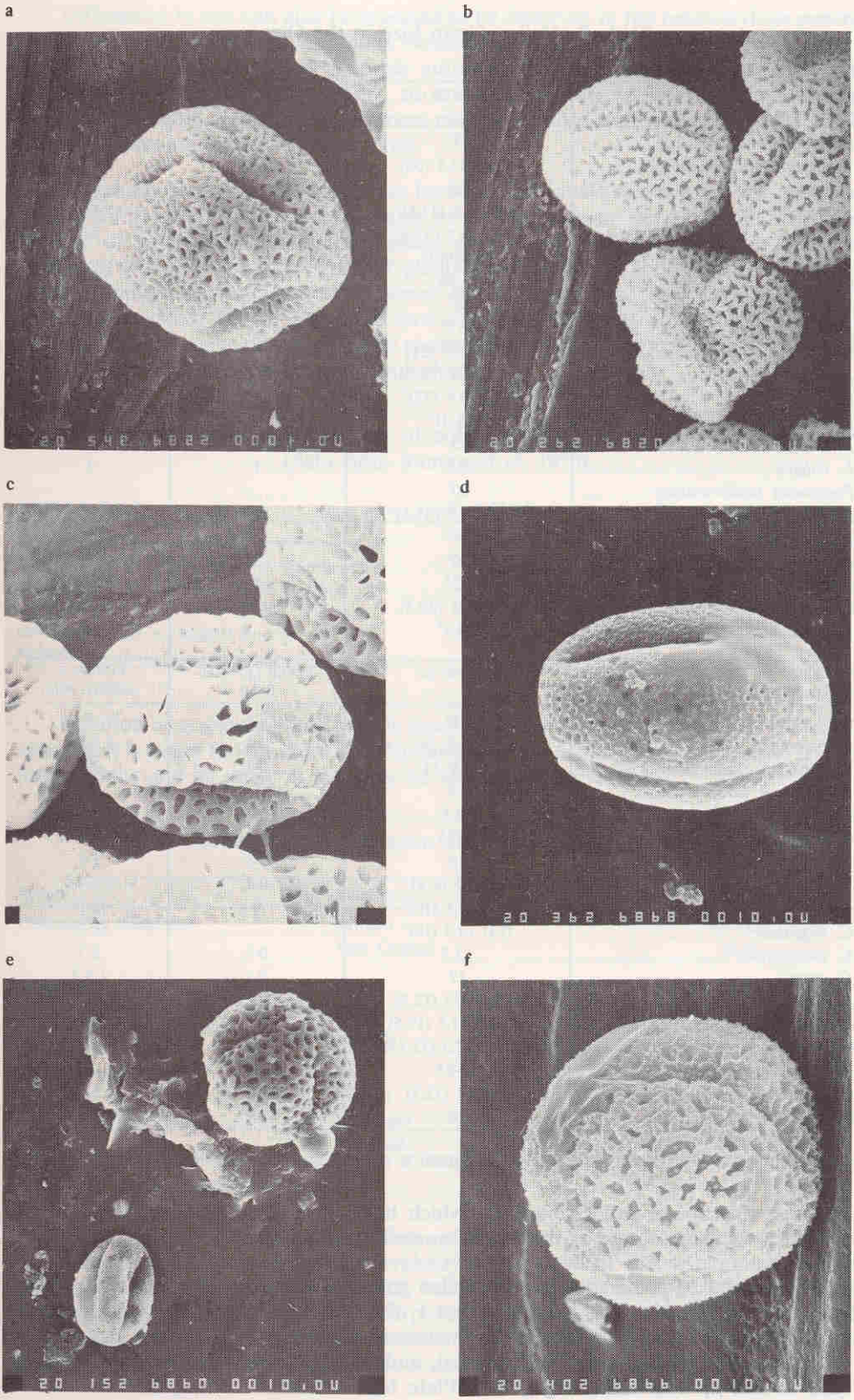


Plate 1. Pollen of Some Malesian Oleaceae
*a: Chionanthus pluriflorus; b: C. ramiflorus; c. Ligustrum confusum; d: Myxopyrum coriaceum; e: M. ovatum* (large and small grains); *f: Olea brachiata*

Table 2. Pollen Grain Size in the Oleaceae

| SPECIES MEDIUM-SIZED POLLEN | DIAMETER ($\mu$) | STYLE LENGTH (MM) | COROLLA LENGTH (MM) |
|---|---|---|---|
| *Menodora integrifolia* | 63[3] | ..... | ..... |
| *Jasminum crassifolium* | (37.5) 43.5 (50) | 10-20 | 35-55 |
| *J. humile* | 46[3] | ..... | ..... |
| *J. multiflorum* | (30) 34.5 (37.5)[1] | 13[1] | 33 |
| *J. sambac* | (27.5) 29.5 (32.8)[2] | 4[2] | 33 |
| | (33)-(90)[4] | ..... | ..... |
| *Nyctanthes arbor-tristis* | 35-58 | 9 | 27-30 |
| *Schrebera holstii* | 35[3] | ..... | ..... |
| *Syringa vulgaris* | 26.3[6] | ..... | 8-15 |
| *Ligustrum confusum* | (20) 22.5 (25) | 1 | 3-4 |
| *L. glomeratum* | (25) 28.3 (32.5) | 1-2 | 2-3 (4) |
| *L. ibota* | (28) 30.5 (32)[6] | ..... | ..... |
| *L. ovalifolium* | 28.5-31[6] | ..... | 8 |
| *L. sinense* | 25 | 1 | 2-3 |
| *L. vulgare* | 28.5[6] | 1 | 3 |
| *Fontanesia phillyraeoides* | 20[3] | ..... | ..... |
| *Forsythia suspensa* | (19.4) 28.5 (37.6)[6] | 9[1] | 19 |
| *Fraxinus americanus* | 24[6] | ..... | ..... |
| *F. coriacea* | 20[6] | ..... | ..... |
| *F. excelsior* | 22[3] | 1 | 2-3 |
| *Osmanthus scortechinii* | (17.5) 21 (22.5) | 1-4 | 1.5-2.5 |
| *Phillyrea angustifolia* | 20.5[3] | ..... | ..... |
| SPECIES SMALL-SIZED POLLEN | Diameter ($\mu$) | STYLE LENGTH (MM) | COROLLA LENGTH (MM) |
| *Olea europaea* | 22[6] | 0-1 | 5 |
| *O. brachiata* | 12.5 & 26 | 0-1 | 1.5-2.5 |
| *O. decussata* (male flower) | 7-9 & 13 | 0-1 | 2-4 |
| (bisexual flower) | 13 | ..... | ..... |
| *O. javanica* | 12.5 | 0-1 | 1-2 (3) |
| *O. paniculata* | 12-15 | 0-1 | 2-3 |
| *Myxopyrum coriaceum* | 16 | 0-1 | 3-5 |
| *M. ovatum* | 13-15.5 & 21 | 0-1 | 2.5-4.5 |
| *Chionanthus retusus* | (13) 14.1 (16)[5] | 0 | 12-18 |
| *C. virginicus* | (13) 14.3 (16)[5] | ..... | 18-30 |
| *C. curvicarpus* | 12.5 | 0-1 | 2-3 |
| *C. enerve* | 17 | 0-1 | 1.5-2 |
| *C. laxiflorus* | (10) 11.25 (12.5) | 0-1 | 2-3 |
| *C. pluriflorus* | (11.25) 13.5 (17.5) | 0-1 | 3-3.5 |
| *C. porcatus* | (11.25) 12.5 (13.75) | 0-1 | 2 |
| *C. ramiflorus* | 10-25 | 0-1 | 3 |
| *C. rubrovenia* | (10) 11 (12.5) | 0-1 | 1.5-2.5 |
| *C. rupicolus* | 10 | 0-1 | 2-4 |

(1: long-styled; 2: short-styled; 3: Erdtman; 4: Raman *et al;* 5: Sohma; 6: Wodehouse)

*Fraxinus, Ligustrum* and *Osmanthus,* which have small flowers, have medium-sized pollen in the same range as the large-flowered genera (Table 2).

Specimens of some species have pollen grains of two sizes, the difference in size being almost double. This phenomenom I observed in *Olea brachiata* (with pollen grains 12.5 and 26 $\mu$ in diameter), *O. decussata* (with the majority being larger, 13 $\mu$, than the other grains, 7-9 $\mu$ in diameter), and in *Myxopyrum ovatum* (the majority were 13-15.5 $\mu$, with some larger, 21 $\mu$ Plate 1e).

Size differences have also been observed by Devi (1975) for *Jasminum callophyllum* where he found the size difference corresponded to the state in which the grain was shed — the smaller grains were 2- or 3-celled, the larger grains were multi-celled.

Differences in size can also be expected to be observed in the heterostylous genera of the Oleaceae: *Forsythia, Jasminum, Nyctanthes* and *Schrebera* (see above). Unfortunately, for pollen sizes given by other authors it is not recorded whether these are for the long- or short-styled flowers, so it is not possible to ascertain whether the range of size within species of these genera may be ascribed to flower type or difference between plants of the same flower type.

Polyploidy can affect the pollen size between individuals of a species. For example, Sohma (1972) noted that tetraploid plants of *C. retusus* from Taiwan are almost twice the size of normal diploid grains. Raman *et al.* (1970) reported a similar case for *Jasminum grandiflorum* where triploid plants produced some grains twice the size of those of diploid plants (Table 3). However, between species of *Jasminum* there is no correlation between size of grain and level of ploidy (Table 3), the tetraploid species falling within the range of other diploid species. The much larger grain of cv. 'Iruvatchi' of *J. sambac* suggests that it is also polyploid.

Table 3. Level of ploidy and diameter of pollen grains in some species of *Jasminum* (data from Raman *et al.* 1970)

| *Jasminum* Species | Level of Ploidy | Diameter of grain ($\mu$) |
|---|---|---|
| *sambac* | 2n | 33-52.8 (42-85.8 cv. 'Iruvatchi') |
| *auriculatum* | 2n | 29-52.8 |
| *grandiflorum* | 2n | 33-46.2 |
| | 3n | 29-102 |
| *communis* | 3n | 33-52.8 |
| *rigidum* | 4n | 39.6-49.5 |

Variation on a geographical basis is small (Table 4) for wild *Chionanthus* material, although it is larger for the cultivated *Nyctanthes arbor-tristis.* This latter species is also heterostylous and its range in size may be ascribed to heterostyly.

Table 4. Variation of pollen grain diameter on a geographical basis

| Species | Locality | Pollen Grain Diameter ($\mu$) |
|---|---|---|
| *Chionanthus ramiflorus* | Borneo | 10-12.5 |
| | New Guinea | 10-15 |
| *C. retusus* | China | 13-15[3] |
| | Japan | 14-16[3] |
| | Taiwan | 13-15[3] |
| *Nyctanthes arbor-tristis* | Java | 42.5-52.5 |
| | Malaya | 35-55 |
| | Singapore | 39[2] |
| | Thailand | 58[1] |

(1: Erdtman, 2: Rao & Leong, 3: Sohma).

Within species, variation in size of the pollen grain in the Oleaceae can therefore be attributed either to heterostyly, polyploidy or the state of the development of the pollen grain when shed. Between species, variation in pollen size appears to be related to style length. The occurrence of small pollen grains in the Oleaceae is here reported for the first time and it is found in species of *Olea, Chionanthus* and *Myxopyrum,* all of which genera have sessile stigmas or styles less than 1 mm long.

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England, Australia, where the SEM study was carried out and to the late Dr Jan Muller of the Rijksherbarium, Leiden, for stimulating and helpful discussions on the pollen of the Oleaceae.

## APPENDIX

Material examined

1. *SEM. Chionanthus enerve* (Steenis) Kiew *S30433; C. pluriflorus* (Knobl.) Kiew *RK779; C. ramiflorus* Roxb. *NGF5969; Ligustrum confusum* Decn. *RK188; Myxopyrum coriaceum* Bl. *S30037; M. ovatum* Hill *NGF46864; Olea brachiata* (Lour.) Merrill *Ahmad s.n.* Kuala Lumpur; *O. decussata* (Heine) Kiew *S22516* (male flower), *Chew 939* (bisexual flower); *O. paniculata* R. Brown *NGF11948.*

2. *Light microscopy. Chionanthus curvicarpus* Kiew *SAN35826; C. laxiflorus* Bl. *SAN49386; C. pluriflorus* (Knobl.) Kiew *RK779; C. porcatus* Kiew *SAN43052; C. ramiflorus* Roxb. *SAN64322; C. rubrovenius* (Elmer) Kiew *Ramos & Edano 38584; C. rupicolus* (Lingels.) Kiew *Jacobs 9292; Jasminum crassifolium* Bl. *Elmer 20762; J. multiflorum* (Burm. f.) Andr. *RK1237; J. sambac* (L). Ait. *RK1238; Ligustrum confusum* Decn. *RK188; L. glomeratum* Bl. *SAN28544; L. sinense* Lour. *SA183; Myxopyrum ovatum* Hill *NGF46864; Nyctanthes arbor-tristis* L. *Junghuhn 33* (Java), *RK1001* (Malaysia); *Olea brachiata* (Lour). Merrill *T & P244; O. javanica* (Bl.) Knobl. *Kds10062; Osmanthus scortechinii* K. & G. *de*

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