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Pollen and Seed Morphology of Some *Ophrys* L. (Orchidaceae) Taxa

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Morphologies of pollen and seed surfaces were investigated with SEM technology for eight natural *Ophrys* taxa collected from southeastern Mediterranean countries and Turkey. The morphometric characteristics of *O. transhyrcana*, *O. sphegodes*, *O. epirotica*, *O. mammosa*, *O. pseudomammosa*, *O. oestrifera* subsp. *oestrifera*, *O. cornuta*, and *O. apifera* were recorded and compared statistically. In general, the current species could be separated, in detail, according to those characteristics. These findings were also compared with those from recent studies, and were examined in terms of the present neighbouring taxa.

Keywords: morphology, *Ophrys*, pollen, seed, Turkey

Amongst the abundance of plant life, orchids can easily be identified by their unique characteristics. For example, most monandrous orchid pollen grains occur as tetrads that are united into a large structure, the pollinium (Dressler, 1981). The seeds of orchids are small and consist of an oval embryo enclosed within generally transparent, often fusiform, testa (Beer, 1863; Arditti, 1967, 1979; Barthlott, 1976). Nevertheless, individual species of orchids can be difficult to distinguish because of variations in their flowers. Unfortunately, researchers have tended to concentrate on macromorphology (leaves, roots, and especially the flower-labellum), but not micromorphology (pollen or seed surfaces), which has led to mostly inadequate species identifications. Whereas, pollen and seed morphology remain relatively stable, other morphological and physiological features can become diversified, so that patterns of the former serve as better taxonomical and phylogenetic markers (Healey et al., 1980; Ackerman and Williams, 1981). Indeed, several recent palynological surveys of the Orchidaceae have shown that pollen morphology and organization may be quite useful especially at higher taxonomic categories (Williams and Broome, 1976; Schill and Pfeiffer, 1977; Newton and Williams, 1978; Schill, 1978; Balogh, 1979; Ackerman and Williams, 1980). Furthermore, the testa and embryos of different genera and species may vary in size, shape, color, and the ratios between their volumes. The walls of testa cells can be smooth or reticulated, and when reticulation is present, its pattern may be distinctive (Healey et al., 1980). However, little has been published about the pollen or seed surfaces of the *Ophrys* genus, and Orchidaceae, as a whole.

For the orchids of Turkey and southeastern Mediterranean countries, many morphological investigations (Sezik, 1967, 1969a, b, c, 1981, 1982, 1984, 1988; Renz and Taubenheim, 1984; Kreutz, 2000) as well as a few caryological and pollen germination studies (Güler and Başak, 1997; Aybeke, 2000, 2002) have been performed, but none has yet attempted to examine the morphology of pollen and/or seed. Moreover, a number of systematical problems still remain unresolved in many orchids, such as *Ophrys*. Furthermore, very diverse types of sculpturing are apparent because of possible exine variations in plants with different phytogeographic origins. Thus, this issue is important to the general characterization of pollen surface features (Schill and Pfeiffer, 1977). Therefore, the aim of this work was to study the pollen and seed surfaces of some *Ophrys* species.

MATERIALS AND METHODS

Seeds collected from the mature capsules of natural plants (Table 1) were prepared for scanning electron microscopy (SEM), and observed as previously described by Arditti et al. (1979, 1980). The colors and dimensions (length and width) were recorded for seeds and embryos under an Olympus stereomicroscope and a Zeiss Jena light microscope, respectively. An Analysis of Variance was performed on the data, which were then statistically analyzed using Scheffe's test for paired comparisons of species (Sokal and Rohlf, 1995). Polliniums also were collected from fresh materials (Table 1). Their dimensions as well as those of the pollen were recorded according to a partially modified procedure from Schill et al. (1992). For SEM preparations, the method was somewhat modified from that of Ackerman and Williams (1981). Briefly, specimens were mounted on double-sided cellophane tape on aluminum stubs, and coated with gold-palladium. The coated grains were then observed and photographed with a Jeol-JSMT-330 scanning electron microscope. Pollen terminology followed that defined by Walker and Doyle (1975) and Punt et al. (2007).

RESULTS

Pollinium and Pollen Morphology

In all taxa, pollen grains are collected in a mass, i.e., the pollinium, and so do not readily separate from each other.

Ophrys transhyrcana Czerniak

Pollinium, 245.0 ± 16.21 μm long and 132.1 ± 12.44 μm wide; pollen grains, 21.2 ± 2.22 μm long and 13.28 ± 3.51 μm wide; monoaperturate, semitectate; verrucate, at distally scabrate (Fig. 1A).

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Table 1. Sources of materials examined.

Species	Locality	Collector	Material[a]
O. sphegodes	Turkey: Edirne, Keşan, Yerlisu village	Aybeke	p, s
O. sphegodes	Albania: Vlore	Cassophotyckyz	p, s
O. sphegodes	Turkey: Keşan, Danışment	Aybeke	s
O. transhyrcana	Bulgaria: Varna	Ahmedov	p, s
O. transhyrcana	Macedonia: Priep	Kasım	p, s
O. transhyrcana	Albania: Vlore	Cassophotyckyz	s
O. mammosa	Turkey: Edirne, Mecidiye	Aybeke	p
O. mammosa	Turkey: Edirne, Lalapaşa, Hasanağa	Aybeke, Güler	p, s
O. mammosa	Turkey: Kırklareli, Dereköy	Aybeke	s
O. mammosa	Turkey: Çanakkale, Gelibolu, Fındıklı	Aybeke	s
O. mammosa	Macedonia: Priep	Kasım	p, s
O. mammosa	Greece: Gümülcine	M. Ali	p, s
O. epirotica	Greece: Alexandroupolis	M. Ali	p, s
O. epirotica	Greece: Volos	Hüseyin	p, s
O. pseudomammosa	Bulgaria: Dobrich	Ahmedov	s
O. pseudomammosa	Bulgaria: Kırcaali	Ahmedov	p
O. oestrifera	Turkey: Edirne, Keşan, Maltepe	Aybeke, Güler	p
O. oestrifera	Turkey: Tekirdağ, Malkara, Keþan	Güler	p
O. oestrifera	Turkey: Çanakkale, Ilgardere	Aybeke	p, s
O. oestrifera	Turkey: Edirne, Erikli	Aybeke, Güler	s
O. oestrifera	Macedonia: Ohri, Stypek	Kasım	p, s
O. oestrifera	Albania: Palase	Cassophotyckyz	s
O. oestrifera	Albania: Lukove	Cassophotyckyz	p
O. oestrifera	Kosovo: Shterpca	Cunephe	s
O. cornuta	Kosovo: Koçaniku	Cunephe	p, s
O. cornuta	Albania: Palase	Cassophotyckyz	p, s
O. cornuta	Albania: Lukove	Cassophotyckyz	p, s
O. apifera	Turkey: Edirne, Keşan, Çeltik village	Aybeke, Güler	p, s
O. apifera	Greece: Oristida	Huseyin	p
O. apifera	Turkey: Çanakkale, Gelibolu, Tayfur	Aybeke	p, s

[a] p, pollinium; s, seed.

Ophrys sphegodes Miller

Pollinium, 315.0 ± 46.44 μm long and 152.1 ± 28.44 μm wide; pollen grains, 19.2 ± 3.28 μm long and 14.96 ± 2.21 μm wide; monoaperturate, simple, porate, tenuate-porate in some pollens; tectate-perforate; rugulate-fossulate and irregularly scabrate, often tending to be verrucate along with other grains, also in some pollen grains on which strap-like bands are connected with other grains (Fig. 1B).

Ophrys epirotica (Renz) Devillers-Tersch. and Devillers

Pollinium, 267.1 ± 26.0 μm long and 106.26 ± 12.34 μm wide; pollen grains, 11.7 ± 2.53 μm long and 12.04 ± 4.3 μm wide; monoaperturate, ca. 11.58 μm; tectate-perforate to semitectate; verrucate- scabrate, but near to margins gemmate (Fig. 1C).

Ophrys mammosa Desf.

Pollinium, 412.74 ± 122.58 μm long and 169.38 ± 45.72 μm wide; pollen grains, 21.64 ± 3.48 μm long and 17.01 ± 3.44 μm wide; mono- or biaperturate, ca. 9.553 ± 2.337 μm; somewhat tenuate-porate; distally semitectate, but tending to tectate-perforate especially along margins of grains; rugulate-granulate, additionally pilate on distal portions (Fig. 1D).

Ophrys pseudomammosa Renz

Pollinium, 421.21 ± 45.65 μm long and 135.67 ± 41.65 μm wide; pollen grains, 19.7 ± 3.41 μm long and 9.23 ± 2.2 μm wide; bi- or pentaperturate, ca. 6.31 ± 1.4 μm; tectate-perforate; rugulate-verrucate, and irregularly suprascabrate (Fig. 2A).

Ophrys oestrifera Bieb. subsp. ***oestrifera***

Pollinium, 435.6 ± 81.36 μm long and 166.05 ± 40.14 μm wide; pollen grains, 20.85 ± 3.23 μm long and 15.76 ± 3.36 μm wide; simple, porate, ca. 7.134 μm; tectate-imperforate; in general psilate (Fig. 2B).

Ophrys cornuta Stev. Ex M. Bieb.

Pollinium, 312.2 ± 43.98 μm long and 195.06 ± 78.54 μm wide (Fig. 2C); pollen grains, 14.45 ± 2.11 μm long and 8.54 ± 1.1 μm wide; tenuate-porate, ca. 3.24 μm; tectate-perforate; psilate and supragranulate (Fig. 2D).

Ophrys apifera Hudson

Pollinium, 375.16 ± 36.35 μm long and 126.16 ± 22.24 μm wide; pollen grains, 16.74 ± 3.53 μm long and 13.07 ± 2.214 μm wide; monoaperturate, ca. 11.58 μm; tectate-

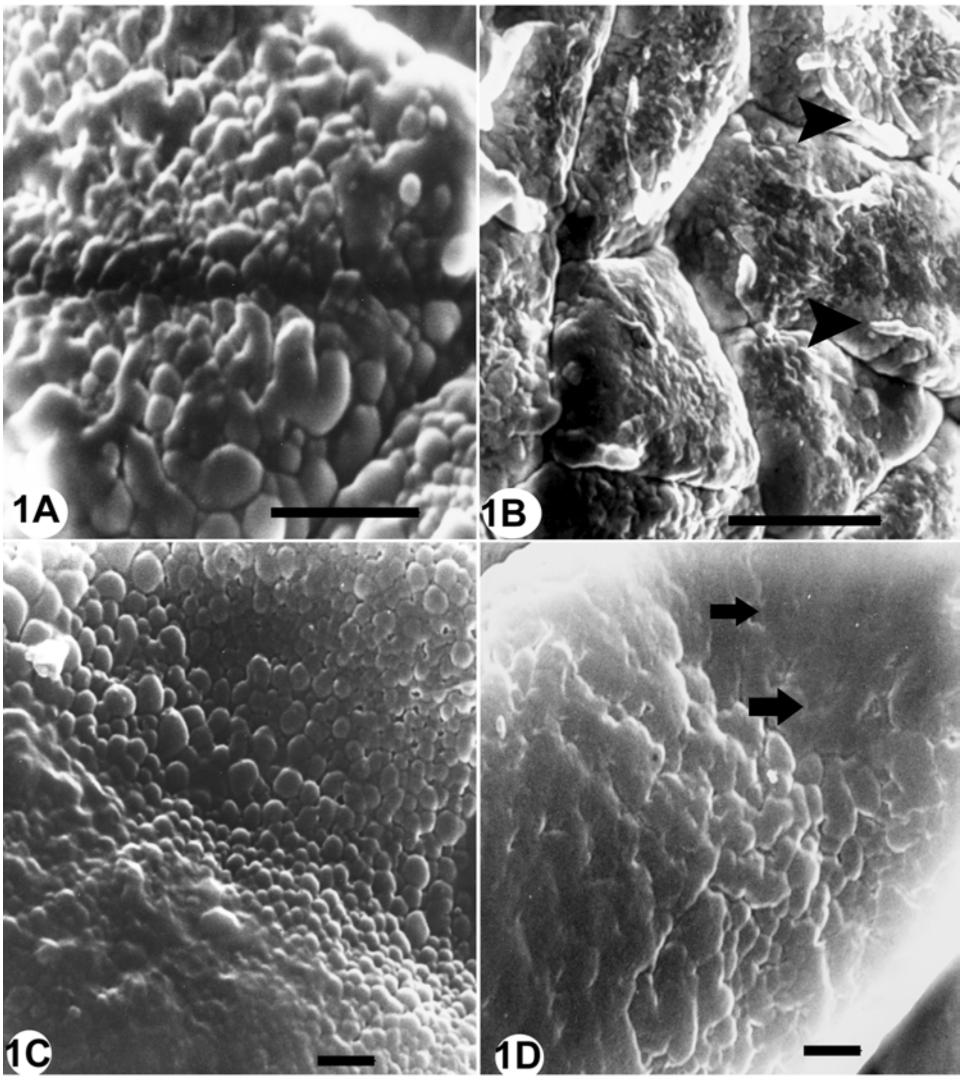

Figure 1. Scanning electron micrographs of *O. transhyrcana* pollen, × 7500 (**A**); *O. sphegodes* group of pollen in pollinium (× 2000), note the strap-like bands (arrowheads) (**B**); *O. epirotica* pollen, × 7500 (**C**); and *O. mammosa* pollen (× 7500), with semi-tectate exine toward aperture (arrows) (**D**).

imperforate to tectate-perforate; rugulate-scabrate, but near to margins partially verrucate (Fig. 3A).

Seed Morphology

Seed surfaces of all the *Ophrys* taxa are reticulated, but those reticulations, a thickening on the periclinal walls, are not similar. Observations of the testa surface showed that the reticulations are irregular in *O. sphegodes*, *O. mammosa*, *O. pseudomammosa*, and *O. oestrifera* subsp. *oestrifera* but regular in *O. transhyrcana* and *O. epirotica*. Those of *O. apifera* are the most regular and are often parallel to each other (Fig. 4, 5). In addition, the degree of anastomosing of testa reticulations in *O. apifera* is less than for the other taxa. In *O. cornuta*, the reticulations are crosswise. With regard to their morphological characteristics, the testa cells are generally rectangle, although they are pentagonal or hexagonal in *O. oestrifera* subsp. *oestrifera* and oval (longitudinal axis of testa) in *O. cornuta*.

Seeds and embryos from all taxa are various shades of brown (Table 2, 3). This morphometric analysis showed that seeds are longest, widest, and smallest in *Ophrys mammosa*, *O. pseudomammosa*, and *O. sphegodes*, respectively (Table 2). As for the embryos, these are longest and widest from *O. apifera*. Based on calculations for truncation values (i.e., length/width), *O. pseudomammosa* and *O. mammosa* have thinner and longer seeds and embryos, respectively, while *O. cornuta* has the widest and the shortest seeds and embryos (Table 2, 3). In general, the chalazal pole from all taxa is slightly narrower than the diameter of the middle portion of the seeds. Differences are not significant among seed volumes except for *O. sphegodes*, where those values are about one-fifth of the others. Likewise, when the numbers of cells on the longest axis of the testa are compared, these differences are minor except for *O. mammosa*.

Testa cell size is relatively uniform within the taxa, except for *O. oestrifera*. This statistical analysis showed some general variations among four parameters -- the lengths and

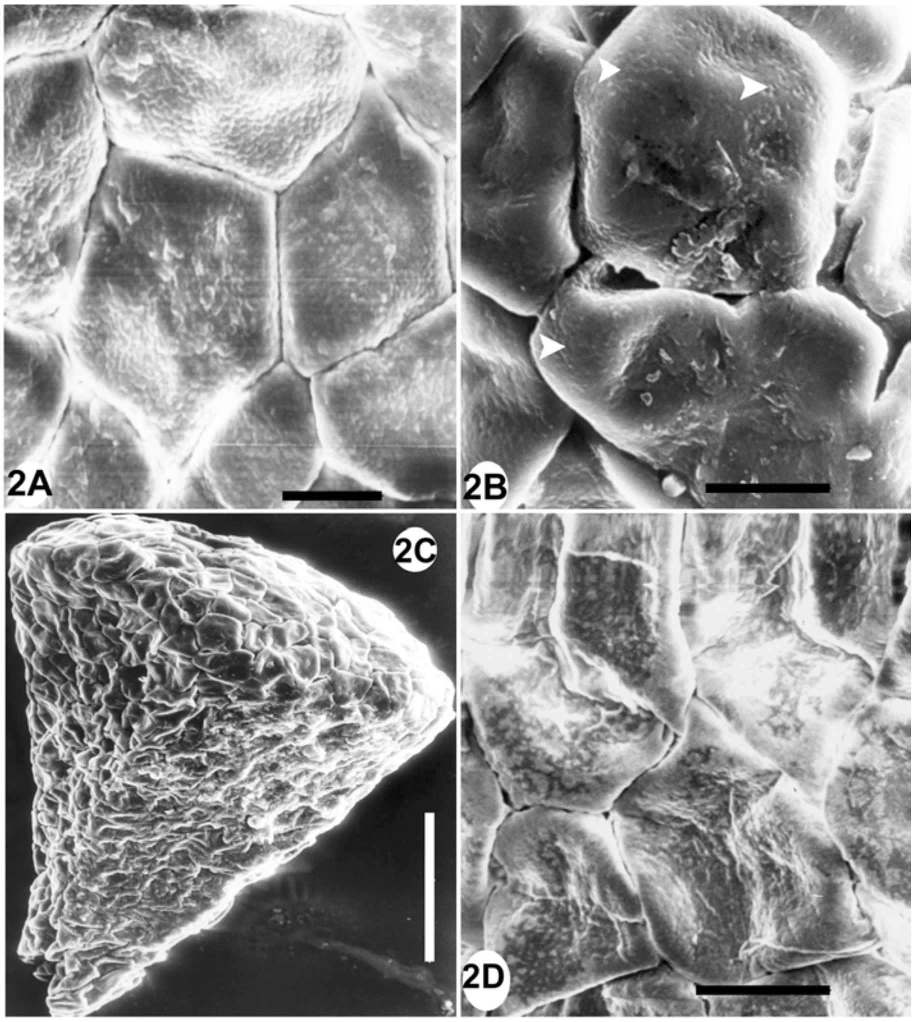

Figure 2. Scanning electron micrographs of *O. pseudomammosa* pollen, × 2000 (**A**); two pollen grains of *O. oestrifera* subsp. *oestrifera* (× 2000), with tectate exine and minute pits on tectum (arrowheads) (**B**); *O. cornuta* pollinium, × 500 (**C**); and *O. cornuta* pollen, × 2000 (**D**).

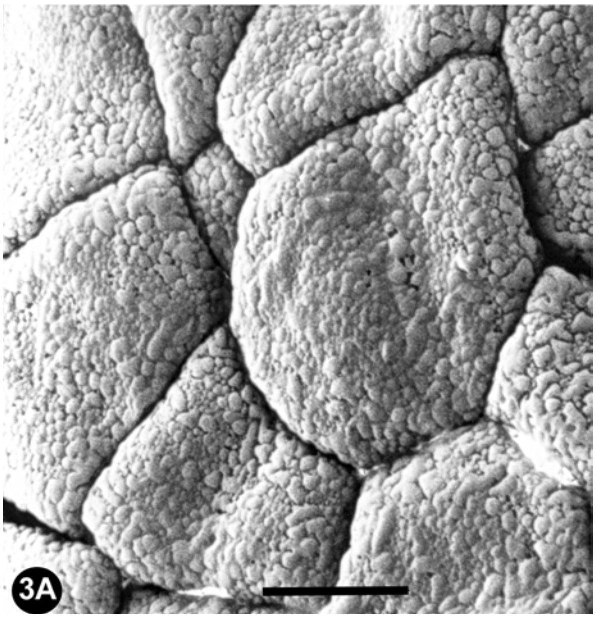

Figure 3. (**A**) Scanning electron micrograph of *O. apifera* pollen with tectate-imperforate exine, showing gradual transition to tectate-perforate, especially on margins (× 2000).

widths of seeds and embryos. However, the pairing of *O. mammosa* with *O. oestrifera* subsp. *oestrifera* does not reveal any statistical differences in their seed widths. Similarly, no differences are seen when embryo lengths are compared between *O. oestrifera* subsp. *oestrifera* and *O. apifera* (Table 4, 5).

DISCUSSION

The pollinium of *O. oestrifera* is remarkably larger than from the other taxa, while *O. transhyrcana* has the smallest and *O. epirotica* has the narrowest ones. In contrast, the dimensions of pollen grains from all taxa are nearly equal while their exine surfaces are species-specific, especially in their fine detail. This is also true of the surfaces at the distal portions and margins of the grains. These taxa do exhibit some differences in their tecta, and especially their sculptural characteristics. Similar results have been reported from other studies of the Orchidoideae subfamily (including *Ophrys*), with the widest variety being observed in the features of the pollen walls (Schill and Pfeiffer, 1977; Burns-Balogh, 1983). Previously, Hesse and Burns-Balogh (1984)

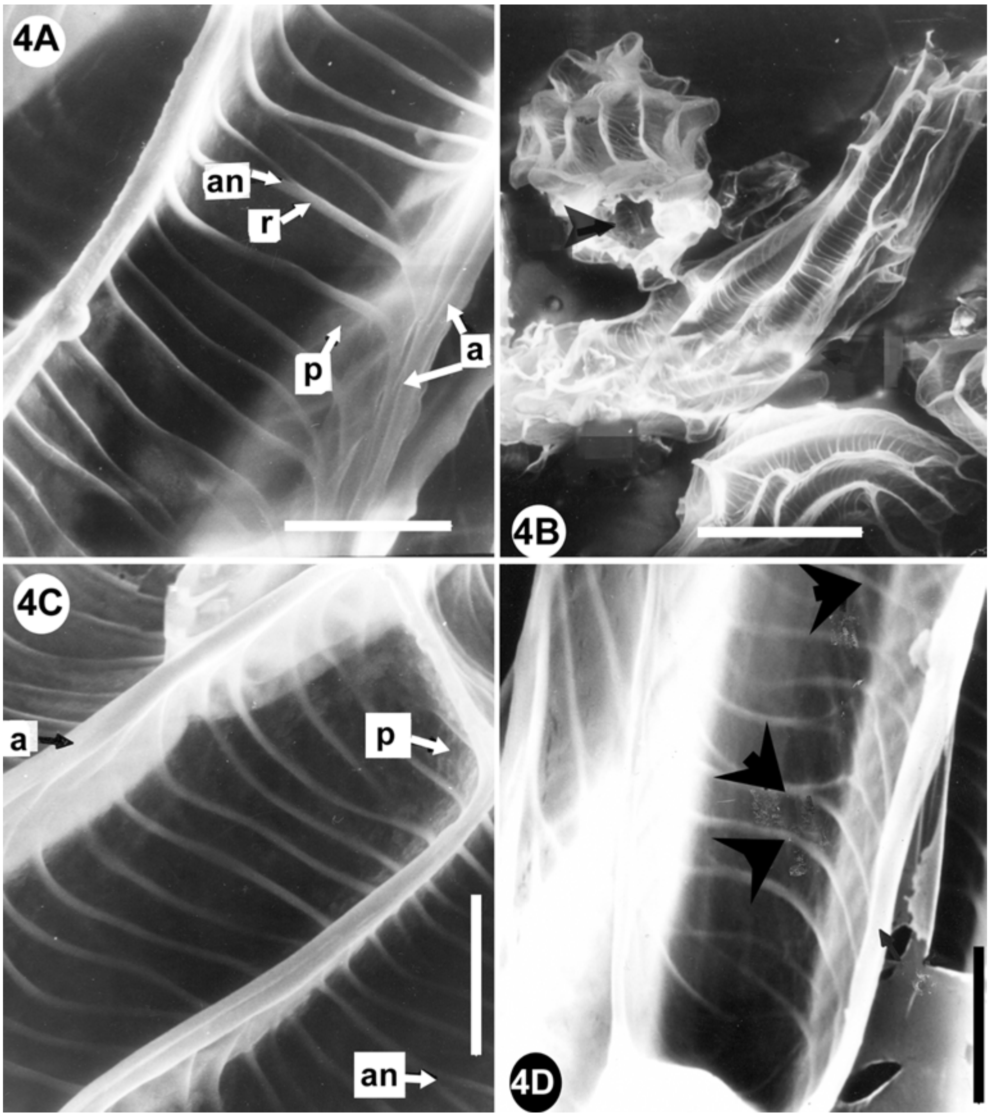

Figure 4. Scanning electron micrographs of seeds from *O. transhyrcana*, × 2000 (**A**); *O. sphegodes* testa and chalazal pole, note interval for mychorrizal fungus infection at arrowhead (× 200) (**B**); *O. epirotica*, irregular reticulations, × 2000 (**C**); and *O. mammosa*, with reticulations anastomozing in testa cell walls at arrowheads (× 2000) (**D**). a, anticlinal wall; an, anastomose; p, periclinal wall; r, reticulation.

found no known distinguishable pollen features unique to a particular genus in the subfamily because of the inherent diversity of the tectum, which ranges from tectate-imperforate to semi-tectate. The sculptural details for some *Ophrys* taxa vary from verrrucose-hamulate to laevigate-scabrate (terminology in their own work), but these are not precise (Schill and Pfeiffer, 1977). The current study, however, does demonstrate some differences in such details. Despite the palynological handicaps, it has only been within the last years that any appreciable examination with TEM, SEM, and LM has been done on orchid pollen ultrastructures. Of particular importance is the overview presented by Schill and Pfeiffer (1977). Burns-Balogh (1983) also has synthesized an evolutionary theory on orchid exine development and pollen ultrastructure, and has suggested that it is not possible to reconstruct phylogenies using exine structures because they are similar in Orchidoideae and Epidendroideae. Nevertheless, even if such great efforts are not sufficient with regard to orchid pollen phylogeny, they make it possible to overcome some systematical problems through careful review. For example, pollen has sometimes been depicted, in various taxonomic categories, as granular, mealy, free, loose, or sectile (Reichenbach, 1852). However, those terms have often been used imprecisely, and so researchers have not always been able to accurately described pollen organizational patterns without conducting SEM surveys. As for the pollinarium, which has been one of the major and most stable characters used at higher taxonomic categories since the work of Adanson (1763), its importance has been emphasized especially with respect to the pollination mechanism (Johnson and Edwards, 2000).

The morphometric characters of the seeds differ between taxa. For very close taxa in particular – i.e., *O. transhyrcana*, *O. epirotica*, *O. sphegodes*, and *O. mammosa* – these can be reasonably separated according to values for truncation, volume, percent air space, and lengths and widths. All seeds

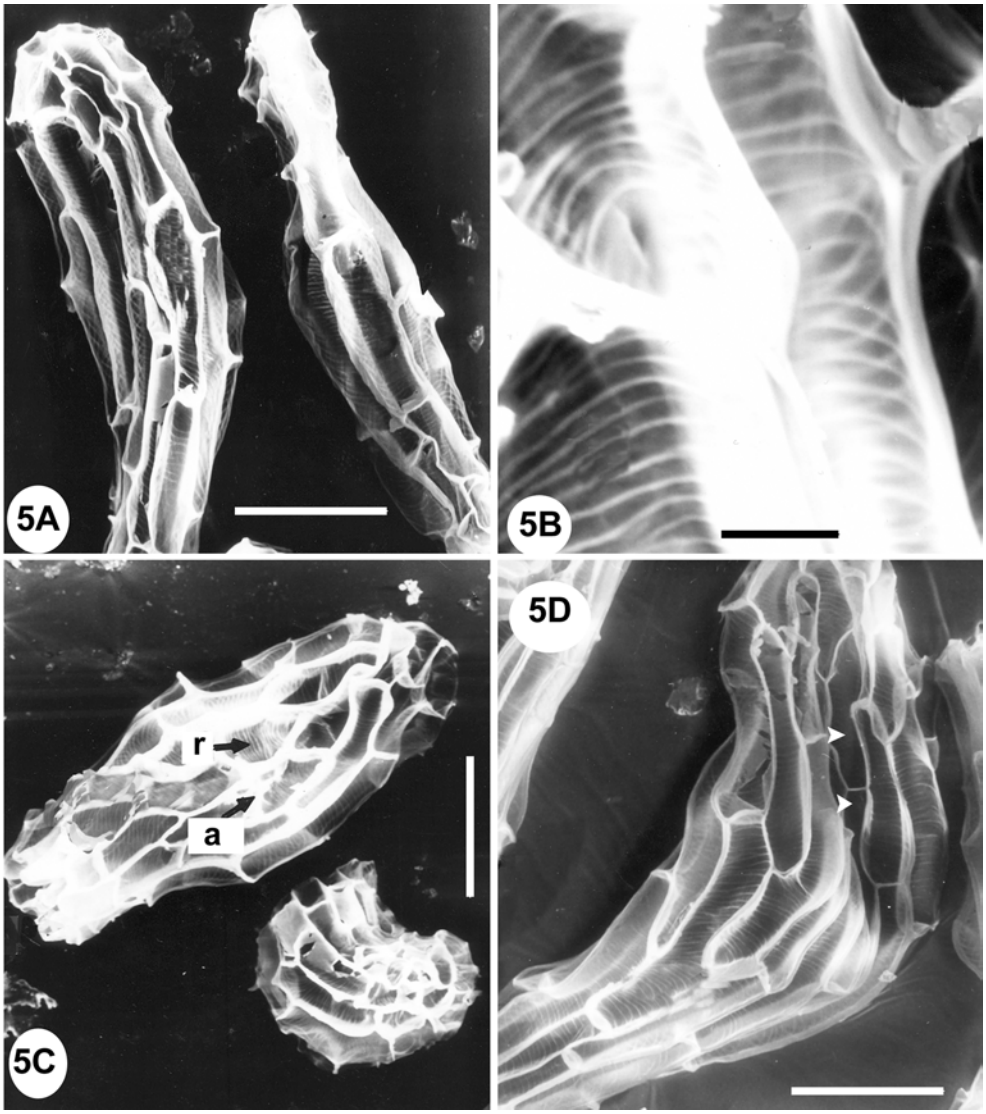

Figure 5. Scanning electron micrographs of *O. pseudomammosa* pollen, general view, × 200 (**A**); *O. oestrifera* subsp. *oestrifera* pentagonal and hexagonal testa cells, × 2000 (**B**); *O. cornuta* with reticulations (**r**) in oval testa cell and anticlinal walls (**a**) × 200 (**C**); and *O. apifera*, with the most regular and parallel reticulations on periclinal cell walls, at arrowheads (× 200) (**D**).

Table 2. Colors and numerical parameters for *Ophrys* seeds.

Species	Color^{a}	Length^{b}	Width^{b}	L/W^{c}	^{d}	e	f	Vol^{g}
O. transhyrcana Yellow		0.523/0.56	0.125/0.085	2.581	4.86	8.57	2.4	1.547
O. sphegodes	Brown-yellow	0.340/0.098	0.089/0.024	3.28	3.96	11.98	1.9	0.704
O. epirotica	Brown	0.421/0.012	0.147/0.004	3.28	4.24	9.41	2.01	2.847
O. mammosa	Brown-yellow	0.557/0.142	0.138/0.023	4.036	6.86	9.19	2.81	2.77
O. pseudo-mammosa	Brown-yellow	0.512/0.086	0.254/0.045	3.452	4.21	10.2	3.41	1.021
O. oestrifera	Brown	0.491/0.121	0.129/0.027	3.806	4.76	12.07	1.8	2.13
O. cornuta	Brown	0.512/0.063	0.215/0.035	2.501	4.62	12.04	2.4	2.62
O. apifera	Brown	0.408/0.119	0.151/0.031	2.701	3.86	11.16	1.96	2.43

^{a}Transparent testa cell wall, ^{b}mm/SD, standard deviation, ^{c}Length/width’ ^{d}No. of cells at longest axis of testa, ^{e}Avg. length of testa cell (mm), ^{f}Cells mm^{-1} length of testa, ^{g}Volume ($mm \times 10^{-3}$).

are usually fusiform, with their chalazal poles being only slightly reduced in diameter. The testa cells are generally elongate rectangles or penta-hexagonal with raised and thickened anticlinal walls (Arditti et al., 1980; Chase and Pippen, 1988). For these taxa, the values for ratios of “seed/embryo volume” and “percent air space” are higher. The greater seed volume generally results because of wider rather than longer testa; higher testa volumes can also come

Table 3. Colors and numerical parameters for *Ophrys* embryos.

Species	Color	Length[a]	Width[a]	L/W[b]	Vol.[c]	Sv/ Ev[d] %	air sp.[e]
O. transhyrcana Brown		0.112/0.014	0.087/0.014	1.54	0.54	6.8	89.54
O. sphegodes	Dark brown	0.0765/0.0201	0.053/0.0198	1.443	0.1	6.4	84.37
O. epirotica	Light Brown	0.124/0.014	0.076/0.0174	1.325	0.35	8.2	79.25
O. mammosa	Brown	0.107/0.0227	0.062/0.0202	1.725	0.21	13.19	92.41
O. pseudo-mammosa	Brown	0.135/0.034	0.065/0.025	1.901	0.43	7.25	83.64
O. oestrifera	Dark brown	0.129/0.0217	0.079/0.0179	1.632	0.42	5.09	80.35
O. cornuta	Brown	0.153/0.024	0.076/0.024	1.345	0.59	9.8	80.97
O. apifera	Brown	0.139/0.008	0.097/0.0197	1.432	0.68	3.57	72.06

[a]mm/SD, Standard deviation, [b]Length/width. [c]Volume (mm^{-3} X10^{-3}), [d]Seed volume /embryo volume, [e]Percent air space: (seed volume-embryo volume) × 100, seed volume.

Table 4. Statistical test results from pairs comparisons among the taxa.

Taxa	O. tra.	O. sphe.	O. epiro.	O. mam.	O. pseud.	O. oest.	O. cor.	O. apif.	Criterion
O. tra.	-	-49.2[a]	19.5	64.8[a]	73.2[a]	-91.2[a]	84.2[a]	42.0[a]	Seed width
	-	35.8[a]	-21.5[a]	-19.3	-28.7[a]	15.7[a]	10.9	-64.8[a]	Seed length
	-	87.9[a]	-25.8[a]	9.8	-43.8[a]	84.5[a]	-52.9[a]	-69.3[a]	Emb. width
	-	-179.3[a]	25.8	-42.6	5.8[a]	-5.97	52.7[a]	47.9[a]	Emb. length
O. sphe.	52.7	-	63.8[a]	63.9[a]	85.2[a]	74.5[a]	89.6[a]	62.10[a]	Seed width
	58.7	-	4.25	72.0[a]	2.58[a]	8.54[a]	86.7[a]	64.79[a]	Seed length
	96.3	-	8.96	74.0[a]	25.8[a]	46.9[a]	85.7[a]	44.1[a]	Emb. width
	78.5	-	74.9[a]	73.9[a]	-85.2[a]	47.9[a]	29.6[a]	63.0[a]	Emb. length
O. epiro.	63.8	54.7[a]	-	25.8[a]	-43.9[a]	-89.6[a]	159.3[a]	25.8[a]	Seed width
	58.7	52.0[a]	-	-45.7[a]	86.9[a]	-75.3[a]	156.3[a]	52.9[a]	Seed length
	45.9	73.2[a]	-	15.2	25.7[a]	-12.7	-25.9[a]	-1.96	Emb. width
	15.7	25.7[a]	-	12.9	-125.6[a]	25.7	128.9[a]	-25.3	Emb. length
O. mam.	12.9	48.0[a]	52.3[a]	-	152.9[a]	8.09	45.9[a]	-14.10[a]	Seed width
	15.9	21.72[a]	25.9[a]	-	45.9[a]	66.10[a]	25.7[a]	152.4[a]	Seed length
	52.9	9.0[a]	75.9[a]	-	14.7	-16.5[a]	-20.5[a]	-35.1[a]	Emb. Width
	12.0	-32.1[a]	45.9[a]	-	78.9[a]	30.9[a]	49.8[a]	-21.9[a]	Emb. Length

[a]Significant at the 5% level.

Table 5. Statistical test results from pairs comparisons between the taxa.

Taxa	O. tra.	O. sphe.	O. epiro.	O. mam.	O. pseud.	O. oest.	O. cor.	O. apif.	Criterion
O. pseud.	-82.0	85.7[a]	56.2[a]	25.7[a]	-	-2.68	29.7[a]	-46.5[a]	Seed width
	58.7[a]	45.7[a]	-59.7[a]	-86.3[a]	-	47.9[a]	85.6[a]	79.5[a]	Seed length
	58.2[a]	89.6[a]	78.9[a]	-85.6[a]	-	79.5[a]	73.5[a]	-196.8[a]	Emb. Width
	25.8[a]	85.7[a]	-83.7[a]	87.0[a]	-	-96.3[a]	70.8[a]	-96.7[a]	Emb. length
O. oest.	1.69	-39.90[a]	36.9[a]	25.8[a]	-79.3[a]	-	46.8[a]	22.2[a]	Seed width
	78.4[a]	151.12[a]	-49.7[a]	92.5[a]	69.8 [a]	-	85.1[a]	89.39[a]	Seed length
	83.4[a]	25.5[a]	8.36	96.8[a]	52.0 [a]	-	15.9	-18.60[a]	Emb. width
	86.2[a]	-52.8[a]	78.9[a]	-25.3[a]	-59.6 [a]	-	89.6[a]	10.2	Emb. length
O. cor.	25.9[a]	36.9[a]	55.8[a]	82.0[a]	45.6 [a]	28.7	-	-25.0	Seed width
	53.6[a]	-93.2[a]	-85.4[a]	20.4	45.9 [a]	58.7[a]	-	78.9[a]	Seed length
	-25.7[a]	-85.7[a]	58.1[a]	20.5	-45.9[a]	78.5[a]	-	78.9[a]	Emb. width
	57.8[a]	58.7[a]	56.3[a]	-78.0[a]	45.1[a]	56.8[a]	-	-189.2[a]	Emb. length
O. apif.	14.7	53.9[a]	56.7	52.4[a]	96.7[a]	25.1	18.4[a]	-	Seed width
	52.3[a]	-25.7	69.1[a]	25.7[a]	-28.1[a]	8.15[a]	25.7	-	Seed length
	45.3[a]	69.7[a]	-53.4[a]	-82.4[a]	-2.15	63.3[a]	29.4[a]	-	Emb. width
	-45.7[a]	83.1[a]	-35.4	8.76	-63.4[a]	52.1[a]	69.4[a]	-	Emb. length

[a]Significant at the 5% level.

from greater length rather than width. In this case, the testa cell values are determined during seed maturation instead of during radial expansion (Arditti et al., 1980). A greater percent of air space may increase seed buoyancy and aid in wind dispersion. For example, two Australian terrestrial orchids -- *Cryptostylis subulata* and *Thelymitra* (Fitzgerald, 1888; Hatch, 1951, 1952; Dockrill, 1969) – are carried on prevailing winds or by jet streams across the Tasman Sea to New Zealand (2000 km in total). Other studies have shown that a lighter weight enables orchid seeds to traverse great distances, logically pointing to buoyancy as an important factor in the aerial distribution of the Orchidaceae (Burgeff, 1936; Went, 1957; Close et al., 1978; Arditti et al., 1979).

With regard to seed morphology, reticulations are present on the testa surfaces of all the taxa examined here, although their shapes differ among species. The reticulations observed in *Calypso* are fine and scalariform, while those in *Corallorhiza* are less pronounced, not as complex, and more diffuse (Arditti et al., 1980). In *Dactylorhiza sambucina*, reticulation is described as dichotomous (Haas, 1977a), while that of *Piperia* tends to be transverse, anastomozing, and species-specific with respect to detail. That pattern is similar in *Spiranthes*, but nevertheless easily recognizable as distinct (Healey et al., 1980). For the *Nigritella* genus, the testa cell walls of *N. nigra* are reticulated while those of *N. miniata* are smooth (Haas, 1977b). In members of the Oncidinae, the great majority of oncidioid species have smooth, unsculptured testa cells, and their anticlinal walls are irregularly thickened. Those species have a similar form of ornamentation on all portions of their testa cells (Chase and Pippen, 1988). Therefore, all of these observations suggest that the presence and nature of reticulation can serve as good taxonomic markers, at least at the generic level. Furthermore, reticulation has been reported, but not in detail, for the tribes of Ophrydoideae, Polychondroideae, and Kerosphaeroideae (the spelling of Schultes and Pease, 1963) as well as several subtribes (Senghas et al., 1974; Barthlott, 1976; Arditti et al., 1979, 1980). Although the anticlinal walls of the testa cells of most orchid seeds are raised, they do not become elongate; this is true of the taxa in the present study. The exceptions to this generality are the extension observed in oncidioid twig epiphytes (Chase, 1986). Hooked seeds appear to be restricted to the vandoid orchids, the most advanced in the Orchidaceae (Dressler, 1981, 1986a, b; Burns-Balogh and Funk, 1986), but they also can be found in the seeds of *Orchis crista-galli* and *Psygmorchis* seeds (Ziegler, 1981).

Therefore, based on previous research and the results presented here, one can conclude that orchid seeds differ considerably in their size, morphology, structure, and fine details. For that reason, morphometric and statistic analyses should be combined with scanning electron microscopy to elucidate systematic relationships. In addition, these observations show that a functional correlation exists between orchid seed morphology and their wetability and aerodynamics that, in turn, affects their dispersion (Beer, 1863; Arditti, 1967, 1979; Senghas et al., 1974; Barthlott, 1976; Arditti et al., 1980). Such morphological data are potentially useful sources for study of this previously unexplored large family (Beer, 1863; Clifford and Smith, 1969; Barthlott, 1976; Ackerman and Williams, 1981; Chase 1986, 1987). In fact, these new findings for *Ophrys* will allow researchers to carry out morphometric and morphological investigations of orchid seed and pollen from different origins, and will undoubtedly be necessary for examining other, previously unknown aspects of this family.

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