POLLEN MORPHOLOGY OF THE CYPRIPEDIOIDEAE AND THE APOSTASIOIDEAE (ORCHIDACEAE)

by Gilbert D. Newton and Norris H. Williams**

INTRODUCTION

Various taxonomic systems have been proposed for the Orchidaceae. Dressler and Dodson (1960) divided the Orchidaceae into two subfamilies: Cypripedioideae and Orchidoideae. The Orchidoideae is the larger subfamily and they considered it the more advanced because the androecium is reduced to one anther. In contrast, the Cypripedioideae contains species with either two or three anthers. Dressler and Dodson considered the Apostasieae and Cypripedieae as tribes in the subfamily Cypripedioideae, but maintained that the Apostasieae may not be related to the Cypripedieae even though they show the same basic plan of flower structure. The Apostasieae have unspecialized four-locular anthers with powdery pollen, the most primitive pollen-unit condition in the Orchidaceae. In the Cypripedieae the grains are not united into larger units, but the pollen is rather viscid, a condition also considered primitive. Dressler (1974) later classified both groups as subfamilies, Apostasioideae and Cypripedioideae, and described them as relict groups, each with a few living genera.

Garay (1972) disagrees with Dressler and Dodson (1960) that the Apostasieae is a tribe in the Cypripedioideae. According to Garay (1972), the only two genera in the group, *Apostasia* and *Neuwiedia*, are not even closely related, but are relict survivors of ancient anagenetic lines. Yet Rao (1974) stated that the external morphology of the flowers and their vasculature show a close affinity between the two genera. Nevertheless, Garay (1972) and Dressler (1974) maintain that the Apostasioideae should be included in the Orchidaceae if the Cypripedioideae are included. Both subfamilies lack pollinia and a rostellum, and both possess three fertile stigmas or stigma lobes.

John Lindley (1830), in a major study conducted in the nineteenth century, compiled the first successful use of pollen characters in a classification system. He described four tribes in the Orchidaceae based on pollen grain size, shape, and degree of compactness. Detailed studies of tropical orchid pollen using light microscopy and scanning electron microscopy (SEM) have been lacking until recently. Caspers and Caspers (1976) examined *Orchis* and *Ophrys* species, and Williams and Broome (1976) surveyed many groups. The latter study demonstrated the presence of various degrees of sculpturing in the orchids, with the Epidendroideae showing the greatest range in diversity of pollen types. Reticulation of the surface was found in the most primitive subtribes, whereas smooth surfaces were seen in the pollinia of the more advanced subtribes. Thus, various types of sculpturing may have taxonomic significance.

Since Cypripedioid taxa were not illustrated by Williams and Broome, the present survey was undertaken. Near completion of this project, Schill and Pfeiffer (1977) published a survey of pollen grains of the Orchidaceae which included the Cypripedioideae. Some of their results are at variance with this study and will be considered in the discussion.

The Cypripedioideae are commonly called the slipper orchids, and according to Luer (1975) consist of four genera: *Selenipedium* (4 species),

---

*Department of Biological Science, Florida State University, Tallahassee, Fla. 32306

Cypripedium (30-40 species), *Phragmipedium* (11 species), and *Paphiopedilum* (70 species). Most of the group is characterized by the presence of a synsepal (an organ formed by the fusion of separate sepals), two petals, a specialized third petal (lip), two fertile anthers, and a disc-like structure (staminode) behind which are the pollen masses and stigma lobes. The plants lack pseudobulbs and the flower lacks a rostellum. Roots are at least 2 mm in diameter and may be a meter in length. Leaves are moderately-to-very thick and green or mottled. *Selenipedium* and *Cypripedium* have plicate leaves, and *Phragmipedium* and *Paphiopedilum* have conduplicate leaves.

A description of the pollen of each species available was made using terminology based on the systems of Walker and Doyle (1975) and Erdtman (1969). Pollen from samples of the Apostasioideae was examined for comparison, as was the pollen from *Curculigo recurvata*, an Indonesian member of the Hypoxidaceae. The Hypoxidaceae has been suggested as a possible ancestor of the Orchidaceae (Garay, 1960). The pollen characters of this group will be correlated with other monocotyledonous groups.

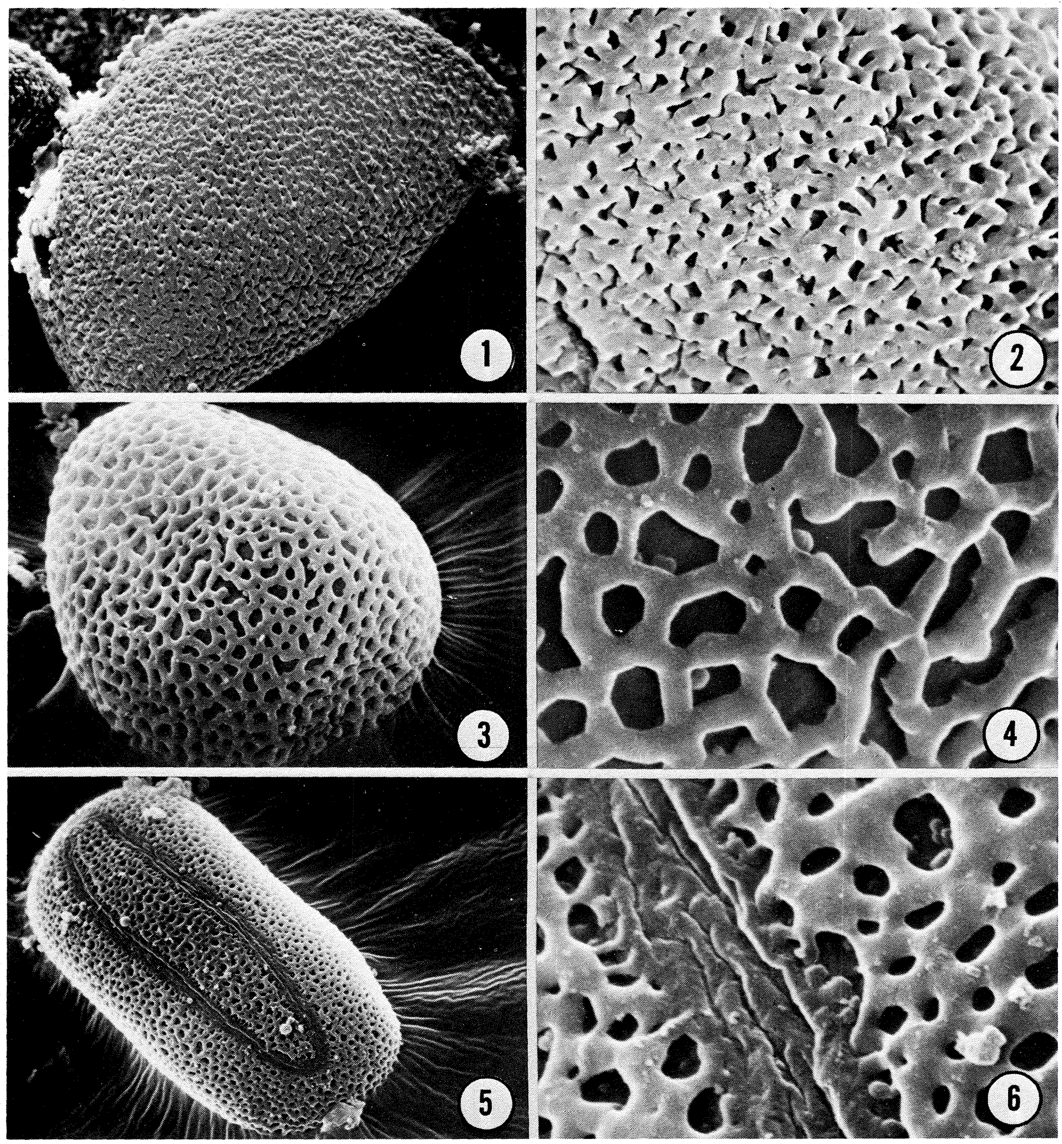
METHODS AND METHODS

Pollen was obtained mainly from cultivated plants at Florida State University in Tallahassee, Florida (FSU), and the Marie Selby Botanical Gardens in Sarasota, Florida (SEL). Additional material was collected from wild growing specimens in the eastern United States and Panama. Available voucher specimens are herbarium specimens and/or flowers preserved in FPA. The species studied are listed in Table 1.

Pollen masses were removed with forceps and stored dry in small vials or preserved in 70% ethyl alcohol (EtOH). The pollen was chemically treated by acetolysis for observation (Erdtman, 1960). This acid hydrolysis destroyed the inner layer of the pollen grain wall (intine) leaving only the outer wall (exine) intact. The pollen was placed in 5 ml of 5% KOH in a series of numbered 13 cc centrifuge tubes. The tubes were placed in a water bath at 90-95°C for 5-10 minutes, removed, and cooled. This cleared the pollen of surface oils and facilitated the observation of surface architecture. The material was then centrifuged in an International Clinical Centrifuge at approximately 2,000 rpm for about three minutes and decanted. The pollen was centrifuged and decanted after each step described below. The material was washed twice with distilled water and twice with glacial acetic acid to prevent an adverse reaction between water and the acetolysis mixture (used in the next step). A freshly prepared acetolysis mixture of one part concentrated sulfuric acid and nine parts acetic anhydride was added to each tube, and the tubes were placed in a water bath at 90-95°C for 5-10 minutes. After cooling, centrifuging, and decanting, the pollen was washed once with glacial acetic acid, twice with distilled water, and once with 70% EtOH.

The acetolyzed pollen was transferred through a graded alcohol series into 100% EtOH. Then it was taken through the following amyl acetate/alcohol (%) series: 25/75, 50/50, 75/25, and finally into a 100% amyl acetate. The pollen was then put into small EM tissue processing capsules and placed in a critical point drying apparatus. The amyl acetate was replaced with carbon dioxide which helped prevent the collapse of the pollen grains (Williams, 1978).

The pollen grains, mounted on SEM stubs, were coated with gold-paladium, and examined with a Cambridge Stereo-scan model S4-10 scanning



Representative pollen samples of *Curculigo* and the Apostasioideae. Figure 1. *Curculigo recurvata* 1700x. Figure 2. *Curculigo recurvata* 4240x. Figure 3. *Apostasia wallichii* 3190x. Figure 4. *Apostasia wallichii* 12,730x. Figure 5. *Neuwiedia veratrifolia* 1790x. Note lid-shaped furrow. Figure 6. *Neuwiedia veratrifolia* 13,650x. Close-up of furrow edge.

electron microscope. Polaroid prints were made using the SEM, and final prints were made by contact printing the Polaroid negatives on Kodak Kodabromide enlarging paper.

Results and Observations

Pollen of 44 species representing the four genera of the Cypripedioideae, the two genera of the Apostasioideae, and *Curculigo recurvata* were examined with the SEM. All the grains were monads, isobilateral, heteropolar, spheroidal to ellipsoidal, and monosulcate. The aperture appears as a deep furrow near one of the poles when visible. The surface of the exine in the majority of species varied from psilate (smooth) to foveolate (pitted) or fossulate (grooved). One species, *Paphiopedilum bullenianum*, was scabrate. The Apostasioideae and *Curculigo recurvata* were reticulate. A summary of the sculpturing types for each species is given in Table 2.

Selenipedium chica (Figure 12) was the only species examined in this genus. The pollen was psilate with a pronounced sulcus at one of the poles. The grains were ellipsoidal.

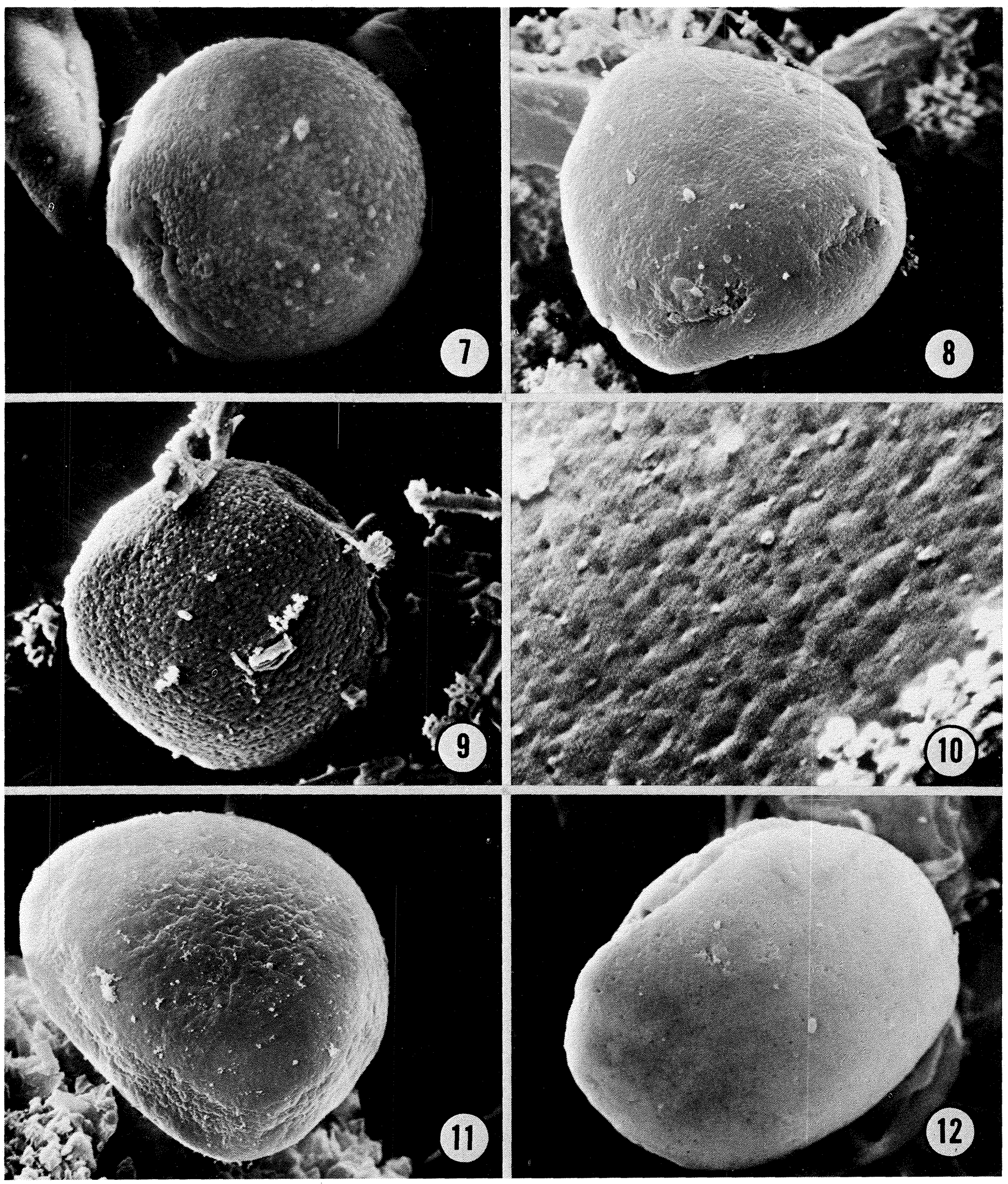
The pollen grain surface in *Cypripedium* was uniformly psilate with one exception. *Cypripedium parviflorum* (Figures 9-10) was characterized by even pitting over the grains. Similar pitting was frequently observed in species of *Paphiopedilum.*

The surface in *Phragmipedium* was consistently psilate in all the species examined with one exception. *Phragmipedium longifolium* (Figure 15) was mostly fossulate. Large grooves tended to break up the surface giving it a rough appearance compared with other *Phragmipedium* species. A hybrid between *P. schlimii* and *P. longifolium* (*Phrag.* × sedenii) had pollen grains of different sizes (Figure 18), but the surface was consistently psilate.

The greatest diversity in the exine sculpturing occurred among the species of *Paphiopedilum*. We distinguished four sculpturing types, and in several cases a combination of these types was evident. The psilate condition was seen in a few *Paphiopedilum* species, and was most pronounced in *P. chamberlainianum* (Figures 27-28) and *P. spicerianum* (Figure 21). The most common type was foveolate in which the surface was broken up by small pits. *Paphiopedilum barbatum* (Figures 25-26) and *P. niveum* (Figures 23-24) are examples. The pitting may also be extensive and deep in large grooves (fossulate), as in *P. concolor* and *P. praestans*. However, it is sometimes difficult to distinguish between foveolate and fossulate as the two types tend to intergrade, and proper terminology for the degree of pitting does not exist.

The presence of small knob-like structures scattered on the surface varies widely within and among several *Paphiopedilum* species. These structures are not part of the sculpturing, but are thought to be the remains of the viscin, a strong elastic substance which binds the pollen grains together in masses. A large portion of the viscin is able to withstand the acetolysis and remains deposited on the pollen grains.

Although the viscin is not part of the sculpturing, it is part of the pollen grain, and occurs in layers on the grain's surface. This sticky adhesive is sloughed off in different layers during acetolysis. This can be seen in *Paphiopedilum godefroyae* (Figures 31-34) in which various grains in the same sample observed were characterized by different degrees of coverage. In Figure 32, the surface is almost completely coated by viscin. Knob-like clumps of viscin are scattered on the surface in Figure 33. Pitting of the surface can be



Representative pollen samples of the Cypripedioideae. Figure 7. *Cypripedium acaule* 1150x. Figure 8. *Cypripedium candidum* 1820x. Figure 9. *Cypripedium parviflorum* 1690x. Figure 10. *Cypripedium parviflorum* 9,400x. Figure 11. *Cypripedium pubescens* 1730x. Figure 12. *Selenipedium chica* 1930x.

seen. Finally, Figure 34 shows a moderately clean surface in which the characteristic pits and grooves can be seen.

Pollen grains of the two Apostasioid species, *Apostasia wallichii* (Figures 3-4) and *Neuwiedia veratrifolia* (Figures 5-6) were monosulcate and reticulate. A prominent lid-shaped furrow can be seen in *N. veratrifolia*. We have not seen this structure in any other pollen grains examined. There was a slight difference in shape between the two species; *Apostasia wallichii* was spheroidal whereas *N. veratrifolia* was ellipsoidal.

A member of the Hypoxidaceae, *Curculigo recurvata* (Figures 1-2), had reticulate pollen grains. They were more similar to the Apostasioideae than the Cypripedioideae.

DISCUSSION

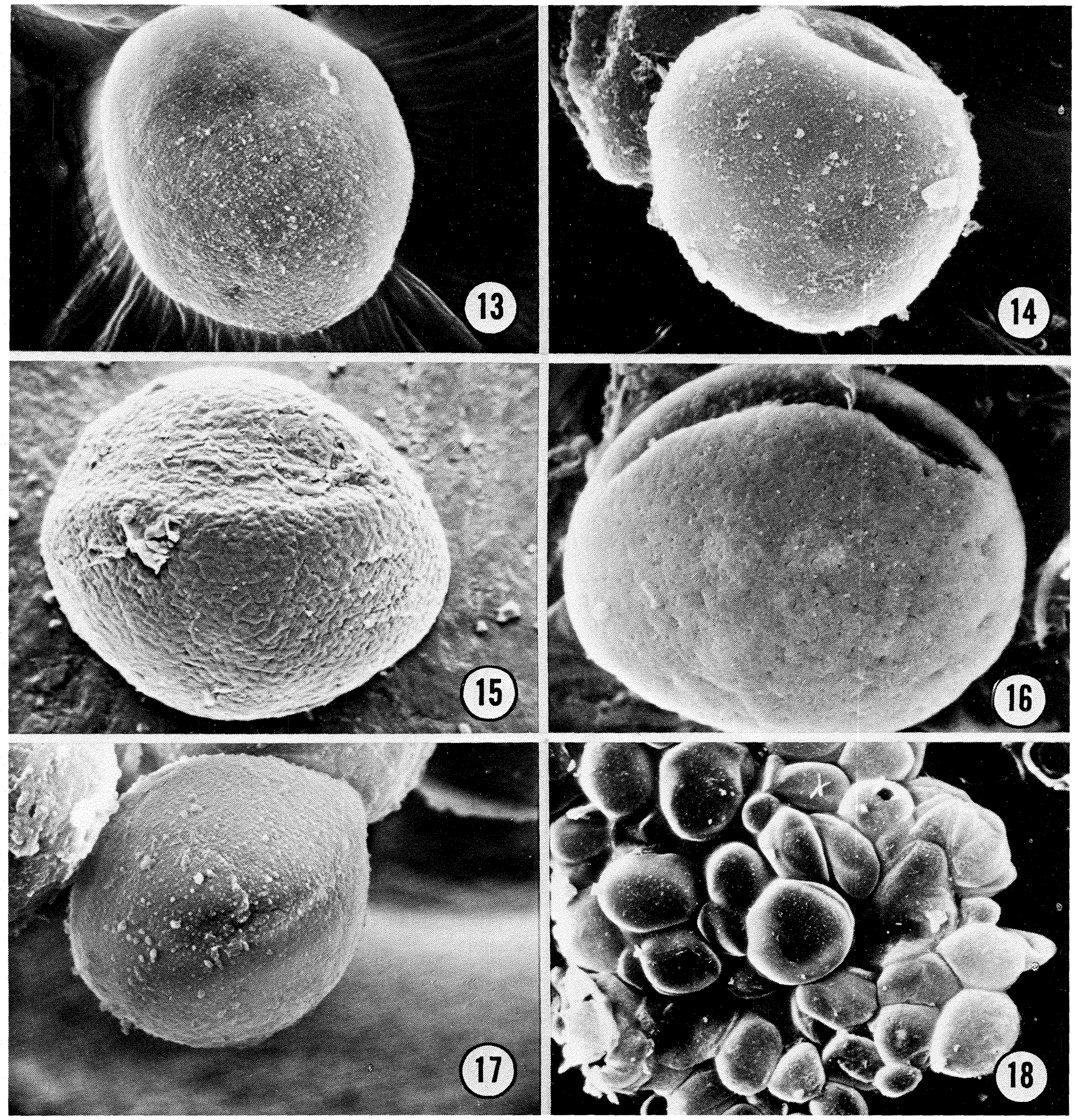
Scanning electron micrographs show that the pollen of the Cypripedioideae is relatively stenopalynous. Pollen of all taxa examined was in monads, monosulcate, heteropolar, isobilateral, and spheroidal to ellipsoidal. Most grains were smooth, sometimes with small pits on the surface, and a few others had small grooves. All of these characteristics are associated with primitive groups (Walker and Doyle, 1975).

Variation in exine sculpturing and density of ornamentation occurred mainly within the genus *Paphiopedilum*. We distinguished four types of sculpturing (see Results), and often a combination of these types was observed on a single pollen grain. Because all parts of the plant are important in establishing relationships (Thorne, 1958), the pollen information given in this paper should aid those describing the phylogeny of *Paphiopedilum*. However, the pollen data must be treated with caution until other lines of evidence are available before establishing phylogenetic relationships of species within this genus.

Schill and Pfeiffer (1977) also described the surface as extremely smooth (laevigate) in *Phragmipedium* and *Paphiopedilum*. However, they did not report the presence of pits or grooves in any of the species examined. They also describe *Cypripedium* as being characterized by two swollen lines on both sides of a deeply etched indentation. It appears from their photographic evidence that the grains are collapsed. We observed the same condition in portions of a sample that had been stored dry. The pollen grains should be rehydrated (Lynch and Webster, 1975) prior to acetolysis and the amyl acetate treatment.

Our study confirms the findings of Williams and Broome (1976) that most of the Cypripedioideae lack the elaborate sculpturing found in other primitive orchids. In fact they showed that the pollen grains of the Neottioideae are usually highly sculptured. In the Epidendroideae they found the greatest range of diversity of pollen types. The more advanced members lacked detailed sculpturing on the outside of the pollinia, while the primitive genera were characterized by a certain degree of sculpturing, such as the presence of small bacula in *Sobralia* pollen grains. Thus, the Cypripedioideae are an exception to the generalization that a smooth surface is an advanced trait within the orchids.

The pollen of *Curculigo recurvata* (Hypoxidaceae) with deeply fossulate to finely reticulate grains is similar to other monocotyledons, particularly the palms (Parthasarathy, 1970), but different from the majority of spe-



Representative pollen samples of the Cypripedioideae (continued). Figure 13. *Phragmipedium hartwegii* 1990x. Figure 14. *Phragmipedium caricinum* 1840x. Figure 15. *Phragmipedium longifolium* 2210x. Figure 16. *Phragmipedium schlimii* 2090x. Furrow sulcus visible along top edge of grain. Figure 17. *Phragmipedium caudatum* 1820x. Figure 18. *Phragmipedium schlimii* x *P. longifolium* 460x - Note different sizes of grains.

cies in the Cypripedioideae. It seems unlikely, based solely on the pollen evidence, that *Curculigo* is a direct ancestor of the slipper orchids.

Pollen of *Apostasia wallichii* and *Neuwiedia veratrifolia* (Apostasioideae) with reticulate grains resembles that of *Curculigo,* a fact mentioned by de Vogel (1969). Garay (1960) suggested that *Curculigo* and *Neuwiedia* have a common ancestor. However, the pollen of the Apostasioideae does not resemble that observed in the Cypripedioideae. Thus, one cannot assume that the Apostasioideae were direct ancestors of the Cypripedioideae based solely on the pollen evidence. Other plant characters must be used to clarify the relationship between these two subfamilies.

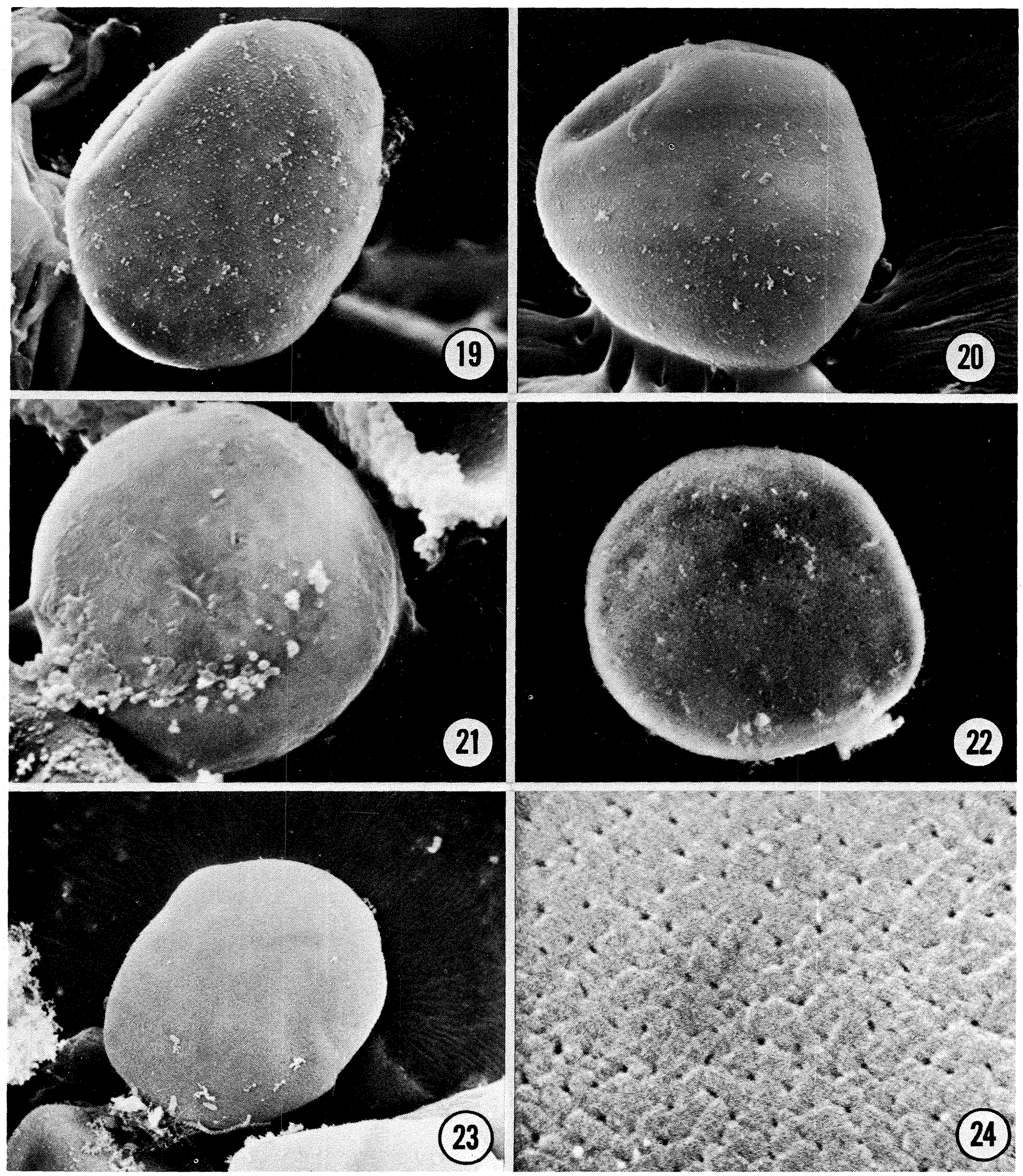
We found that the two most phylogenetically useful characters are the aperture and the sculpturing of the exine. The presence of a single aperture is considered primitive and is found in gymnosperms, monocotyledons, and some dicotyledons (Smith, 1967; Takhtajan, 1969; Cronquist, 1968). According to Walker (1976), there are five "categories" describing the sculpturing of the exine. In this system, psilate pollen is the most primitive, giving rise to foveolate and/or fossulate on one evolutionary branch, and scabrate to verrucate on another. The pollen grains of several species in the Cypripedioideae and Apostasioideae resemble those of other monocotyledons, such as palms (Parthasarathy, 1970). Sowunmi (1972) identified 25 types of pollen morphology for the Palmae. Some of the palm species have fossulate grains and are similar to pollen grains seen in certain *Paphiopedilum* species. The pollen of the Cypripedioideae is similar to other primitive monocotyledons in possessing these traits.

The more elaborate sculpturing is correlated with entomophily, while psilate pollen grains are associated with anemophilous plants (Whitehead, 1969). The presence of spines (seen in such groups as the Compositae) facilitates attachment of the grains to the insect-vector (Stebbins, 1974). However, the Cypripedioideae are insect-pollinated, yet are mostly psilate or foveolate. The presence of spines is unnecessary for adherence to the insect because the pollen grains are held together by viscin which will adhere to almost anything that comes in contact with it.

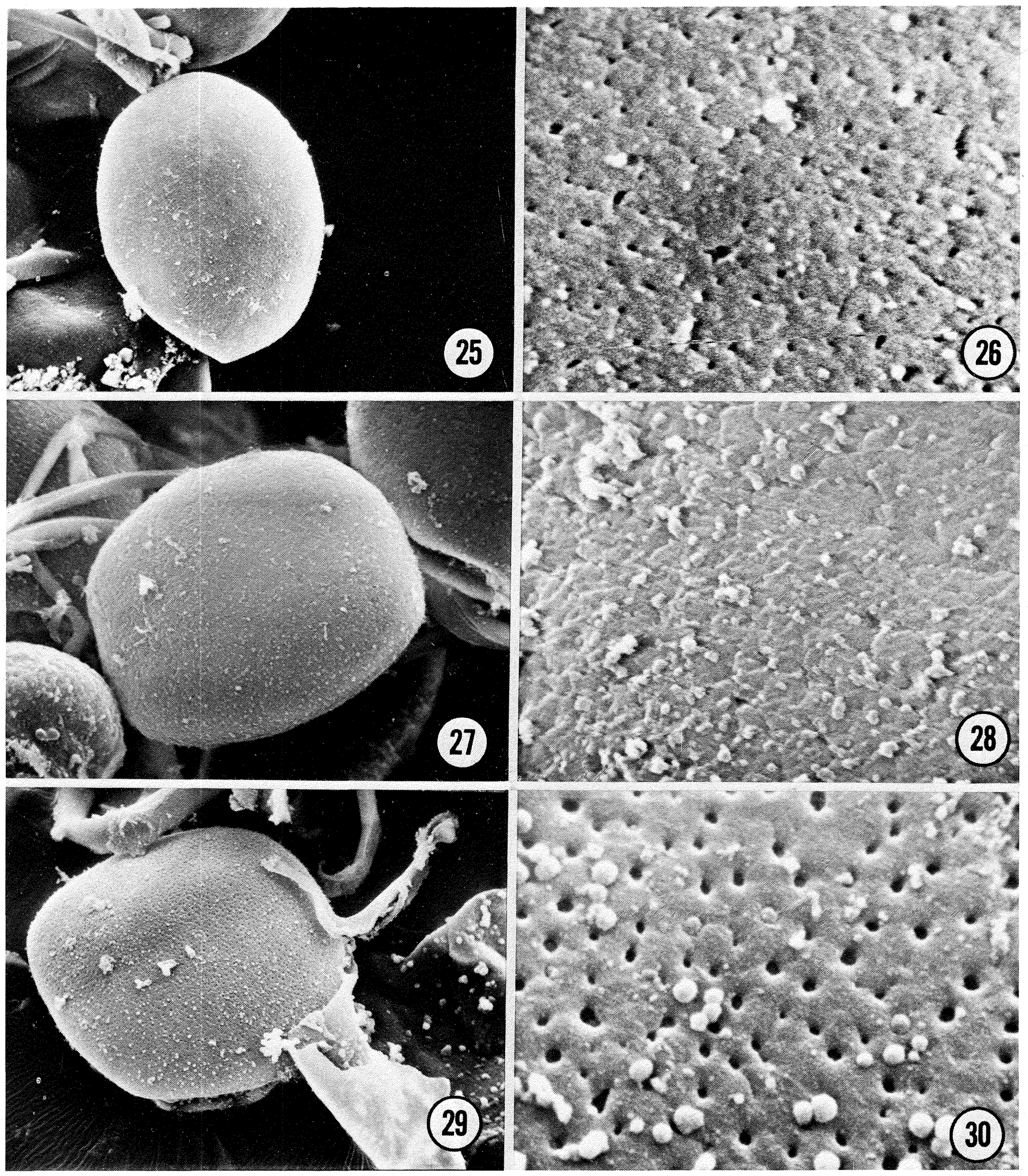
In summary, all the pollen grains of the Cypripedioideae were characterized by primitive traits. Our results support the findings of Williams and Broome (1976) that the Cypripedioideae lack the elaborate sculpturing seen in other orchid groups. Therefore, the Cypripedioideae are an exception to their generalization that the pollen surface of advanced orchids is psilate whereas that of primitive orchids is sculptured. The pollen of the Apostasioideae fits this pattern. Finally, the pollen grains of *Curculigo recurvata* were reticulate and similar to those of the Apostasioideae.

ACKNOWLEDGMENTS

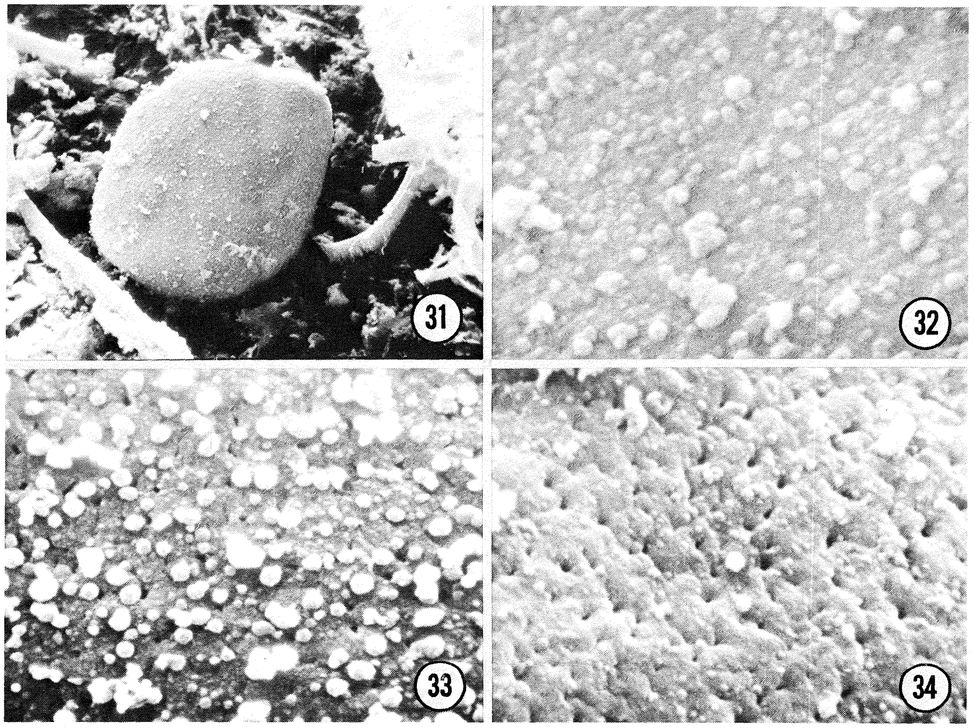
We thank Drs. Loran C. Anderson and L. Raymond Fox for their critical suggestions concerning the research project and manuscript; William Miller who provided technical assistance with the scanning electron microscope; and John T. Atwood who graciously allowed us use of his Cypripedioideae collection. We are indebted to the Marie Selby Botanical Gardens in Sarasota, Florida for providing access to their facilities and collections. Portions of this research were supported by a grant from the American Orchid Society Fund for Education and Research, Inc. to N. H. Williams.



Representative pollen samples of the Cypripedioideae (continued). Figure 19. *Paphiopedilum hennisianum* 1710x. Figure 20. *Paphiopedilum appletonianum* 1610x. Figure 21. *Paphiopedilum spicerianum* 2670x. Figure 22. *Paphiopedilum sukhakulii* 1890x. Figure 23. *Paphiopedilum niveum* 1320x. Figure 24. *Paphiopedilum niveum* 13,160x.



Representative pollen samples of the Cypripedioideae (continued). Figure 25. *Paphiopedilum barbatum* 1170x. Figure 26. *Paphiopedilum barbatum* 11,700x. Figure 27. *Paphiopedilum chamberlainianum* 1500x. Figure 28. *Paphiopedilum chamberlainianum* 7500x. Figure 29. *Paphiopedilum hirsutissimum* 1290x. Figure 30. *Paphiopedilum hirsutissimum* 12,880x.



Representative pollen samples of the Cypripedioideae (continued). Figure 31. *Paphiopedilum godefroyae* 1250x. Figure 32. *Paphiopedilum godefroyae* 12,500x. Figure 33. *Paphiopedilum godefroyae* 12,500x. Figure 34. *Paphiopedilum godefroyae* 12,500x.

Table 1 — Species Examined

Species	Distribution	Source	Voucher Deposited
Curculigo recurvata Dryand.	Malaysia	L. Beidler - cult. at FSU	FSU - 486
Apostasia wallichii R. Br.	S.E. Asia	P. Taylor, Kew	Kew Sp. Coll. no. 26490
Neuwiedia veratrifolia Bl.	S.E. Asia	P. Taylor, Kew	Kew Sp. Coll. no. 31949
Selenipedium chica Rchb. f.	Panama	Norris H. Williams coll. in Panama	FSU - 487
Cypripedium acaule Ait.	N.E. North America	John T. Atwood coll. in Vermont	FSU - 77210
C. arietinum R. Br.	N.E. North America	JTA - coll. in N. Michigan	FSU - 77208
C. californicum A. Gray	N. California	Michael R. Mesler in N. California	No Voucher
C. candidum Muhl. ex Willd.	N. Central United States	JTA - coll. in C. Michigan	FSU - 77203
C. guttatum Sw.	Circumboreal	Chris Sparado coll. on Kodiak Island	FSU - 77715
C. parviflorum Salis.	N.E. United States	JTA - coll. in C. Michigan	FSU - 7632
C. pubescens Willd.	E. United States & Canada	JTA - coll. in N. Michigan	FSU - 77207
Phragmipedium caricinum (Lindl.) Rolfe	South America	SEL	FSU - 77189
P. caudatum (Lindl.) Rolfe	C. America & Andean S. America	JTA - cult. at FSU	FSU - 7616
P. hartwegii (Rchb. f.) L.O. Wms.	N.W. South America	SEL	No Voucher

P. longifolium (Rchb. f.) Rolfe	S. Central America & N. South America	JTA - cult. at FSU	FSU - 77173
P. pearceii (Rchb. f.) Rauh & Sengh.	Andean S. America	SEL	FSU - 7622
P. schlimii (Rchb. f.) Rolfe	Colombia	JTA - cult. at FSU	FSU - 747
P. X*sedenii*		JTA - cult. at FSU	FSU - 7618
Paphiopedilum acmodontum Schoser ex M.W. Wood.	Philippines	JTA - cult. at FSU	FSU - 741-1
P. appletonianum (Gower) Rolfe	Thailand	JTA - cult. at FSU	FSU - 7325
P. argus (Rchb. f.) Pfitz.	Philippines	JTA - cult. at FSU	FSU - 732
P. barbatum (Lindl.) Pfitz.	Malay Peninsula	JTA - cult. at FSU	FSU - 761
P. bullenianum (Rchb. f.) Pfitz.	Borneo	JTA - cult. at FSU	FSU - 763
P. callosum (Rchb. f.) Pfitz.	S.E. Asia	JTA - cult. at FSU	FSU - 751-1
P. chamberlainianum (O'Brien) Pfitz.	Indonesia	JTA - cult. at FSU	FSU - 7641
P. ciliolare (Rchb. f.) Pfitz.	Philippines	JTA - cult. at FSU	FSU - 765
P. concolor (Batem.) Pfitz.	S.E. Asia	JTA - cult. at FSU	FSU - 759
P. exul (O'Brien) Pfitz.	Malay Peninsula	JTA - cult. at FSU	FSU - 7612
P. fairieanum (Lindl.) Pfitz.	E. Himalaya Region	JTA - cult. at FSU	FSU - 7412
P. glaucophyllum (J.J. Smith) Pfitz.	Indonesia	Charles Portes	No Voucher
P. godefroyae (Hemsl.) Pfitz.	S.E. Asia	JTA - cult. at FSU	FSU - 7314
P. haynaldianum (Rchb. f.) Pfitz.	Philippines	JTA - cult. at FSU	FSU - 7310
P. hennisianum (M.W. Wood) Fowlie	Philippines	JTA - cult. at FSU	FSU - 742-3
P. hirsutissimum (Lindl.) Pfitz.	E. Himalaya Region	SEL	SEL-1-75-80
P. lowii (Lindl.) Pfitz.	Non-mainland S.E. Asia	JTA - cult. at FSU	FSU - 77199
P. niveum (Rchb. f.) Pfitz.	Malay Peninsula to Borneo	JTA - cult. at FSU	FSU - 7615
P. mastersianum (Rchb. f.) Pfitz.	Borneo	JTA - cult. at FSU	FSU - 767-1
P. parishii (Rchb. f.) Pfitz.	Burma	JTA - cult. at FSU	FSU - 768
P. praestans (Rchb. f.) Pfitz.	Islands N.W. of New Guinea	JTA - cult. at FSU	FSU - 738
P. spicerianum (Rchb. f.) Pfitz.	E. Himalaya Region	JTA - cult. at FSU	FSU - 662
P. sukhakulii Schoser & Senghas	N.E. Thailand	JTA - cult. at FSU	FSU - 7322
P. tonsum (Rchb. f.) Pfitz.	Sumatra	JTA - cult. at FSU	FSU - 734
P. venustum (Wall.) Pfitz.	E. Himalaya Region	JTA - cult. at FSU	FSU - 748
P. villosum (Lindl.) Pfitz.	E. India to Thailand	JTA - cult. at FSU	No Voucher
P. violascens Schltr.	Papua	JTA - cult. at FSU	FSU - 7631-2

Table 2 — Summary of Pollen Sculpturing Types

Species	Exine	Species	Exine
Curculigo		_Paphiopedilum_	
*recurvata	Fs-R	_acmodontum_	Fs
		*appletonianum	Fv
Apostasia		_argus_	Fv
*wallichii	R	_*barbatum_	Fv
		bullenianum	S
Neuwiedia		_callosum_	Fv
*veratrifolia	R	_*chamberlainianum_	P
		ciliolare	Fs
Selenipedium		_concolor_	Fs
*chica	P	_exul_	Fv
		fairieanum	Fv
Cypripedium		_glaucophyllum_	P
*acaule	P	_*godefroyae_	Fv
arietinum	P	_haynaldianum_	Fs
californicum	P	_*hennisianum_	P
*candidum	P	_*hirsutissimum_	Fv
guttatum	P	_lowii_	Fv
*parviflorum	Fv	_*niveum_	Fv
*pubescens	P	_mastersianum_	Fs
		parishii	Fv
Phragmipedium		_praestans_	Fs
*caricinum	P	_*spicerianum_	P
*caudatum	P	_*sukhakulii_	P
*hartwegii	P	_tonsum_	Fv
*longifolium	Fs	_venustum_	P
pearceii	P	_villosum_	P
*schlimii	P	_violascens_	Fs

P = psilate
Fv = foveolate
Fs = fossulate
S = scabrate
R = reticulate
*See Figures

LITERATURE CITED

Caspers, N. and L. Caspers 1976. Zur Oberflaechenskulpturierung der Pollinien Mediterraner _Orchis_-und _Ophrys_-Arten. Pollen et Spores 18:203-215.

Cronquist, A. J. 1968. The evolution and classification of flowering plants. Houghton Mifflin. Boston. 396 pp.

De Vogel, E. F. 1969. Monograph of the tribe Apostasieae (Orchidaceae). Blumea 17: 313-350.

Dressler, R. L. 1974. Classification of the orchid family. Pages 259-270 _in_ M. Ospina ed., Proceedings of the Seventh World Orchid Conference. Medellin, Colombia.

Dressler, R. L. and C. H. Dodson 1960. Classification and phylogeny in the Orchidaceae. Ann. Missouri Bot. Gard. 27:25-68.

Erdtman, G. 1960. The acetolysis method — a revised description. Svensk Bot. Tidskr. 54:561-564.

__________1969. Handbook of palynology. Munksgaard. Copenhagen. 486 pp.

Garay, L. A. 1960. On the origin of the Orchidaceae.

__________1972. On the origin of the Orchidaceae II. J. Arnold Arbor. 53:202-215.

Lindley, J. 1830. The genera and species of orchidaceous plants. London. Cited in Wodehouse, R. P. 1959, Pollen grains. Hafner Publishing Co., New York. 574 pp.

Luer, C. A. 1975. The native orchids of the United States and Canada excluding Florida. The New York Botanical Garden. 361 pp.

Lynch, S. P. and G. L. Webster 1975. A new technique of preparing pollen for scanning electron microscopy. Grana 15:127-136.

Parthasarathy, M. V. 1970. Fine structure of pollen surface in palms. Principes 14:55-62.

Rao, V. S. 1974. The relationships of the Apostasiaceae on the basis of floral anatomy. Bot. J. Linn. Soc., 68:319-327.

Schill, R. and W. Pfeiffer 1977. Untersuchungen an Orchideenpollinien unter besonderer Beruecksichtigung ihrer Feinskulpturen. Pollen et Spores 19:5-118.

Smith, A. C. 1967. The presence of primitive angiosperms in the Amazon Basin and its significance in indicating migrational routes. Atas do Simpósio sôbre a Biota Amazônica 4:37-59.

Sowunmi, M. A. 1972. Pollen morphology of the Palmae and its bearing on taxonomy. Rev. Paleobot. and Palyn. 13:1-80.

Stebbins, G. L. 1974. Flowering plants: evolution above the species level. The Belknap Press of Harvard Univ., Cambridge, Mass. 399 pp.

Takhtajan, A. 1969. Flowering plants: origin and dispersal. Smithsonian Institution Press. Washington, D. C. 310 pp.

Thorne, R. F. 1958. Some guiding priciples of angiosperm phylogeny. Brittonia 10: 72-77.

Walker, J. W. 1976. Evolutionary significance of the exine in the pollen of primitive angiosperms. Pages 251-308 *in* I. K. Ferguson and J. Muller eds., The evolutionary significance of the exine. Academic Press. N.Y.

Walker, J. W. and J. A. Doyle 1975. The bases of angiosperm phylogeny: palynology. Ann. Missouri Bot. Gard. 62:664-723.

Whitehead, D. R. 1969. Wind pollination in the angiosperms; evolutionary and environment considerations. Evolution 23:28-35.

Williams, N. H. 1978. Pollen structure and the systematics of the neotropical Gesneriaceae. Selbyana 2(2-3): 310-322.

Williams, N. H. and C. R. Broome 1976. Scanning electron microscope studies of orchid pollen. Amer. Orchid Soc. Bull. 45:699-707.